

Smg1 Cas9-CKO Strategy

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Reviewer: Rui Xiong

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Project Overview



Project Name Smg1

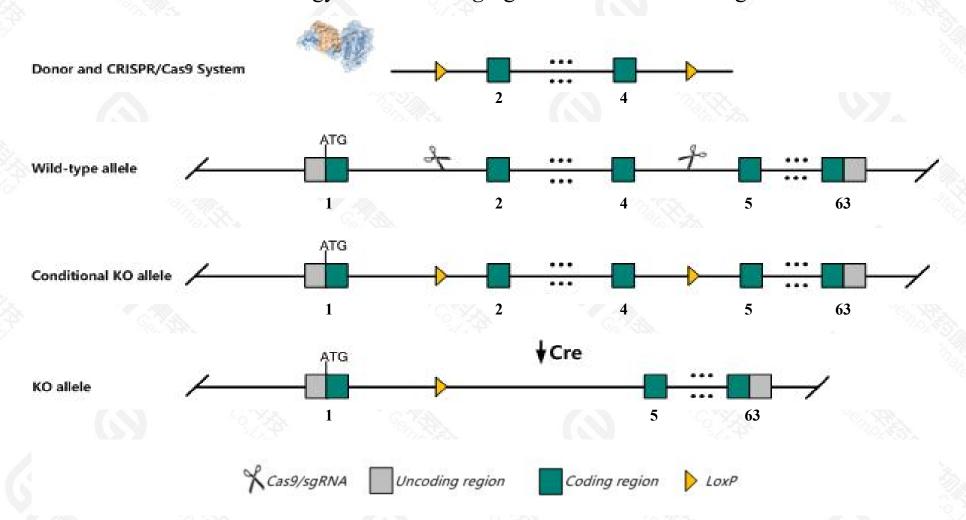
Project type Cas9-CKO

Strain background C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Smg1 gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Smg1* gene has 10 transcripts. According to the structure of *Smg1* gene, exon2-exon4 of *Smg1*201(ENSMUST00000032891.15) transcript is recommended as the knockout region. The region contains 457bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Smg1* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor was constructed.Cas9, sgRNA and Donor 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.
- > The flox mice was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

Notice



- > According to the existing MGI data, mice homozygous for a gene trap allele exhibit early embryonic lethality. Mice heteroygous for a gene trap allele exhibit abnormal tooth development, chronic inflammation, increased body weight, increased incidence of tumor formation and premature death.
- ➤ Transcript *Smg1-202* may not be affected.
- > The *Smg1* gene is located on the Chr7. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)



Smg1 SMG1 homolog, phosphatidylinositol 3-kinase-related kinase (C. elegans) [Mus musculus (house mouse)]

Gene ID: 233789, updated on 17-Dec-2020

Summary



Official Symbol Smg1 provided by MGI

Official Full Name SMG1 homolog, phosphatidylinositol 3-kinase-related kinase (C. elegans) provided by MGI

Primary source MGI:MGI:1919742

See related Ensembl:ENSMUSG00000030655

Gene type protein coding
RefSeq status PROVISIONAL
Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;

Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as 2610207105Rik, 5430435M13Rik, C130002K18Rik, mKIAA0421

Expression Ubiquitous expression in cerebellum adult (RPKM 16.3), thymus adult (RPKM 14.4) and 28 other tissuesSee more

Orthologs <u>human</u> all

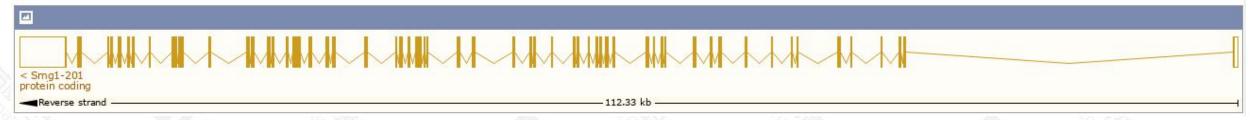
Transcript information (Ensembl)



The gene has 10 transcripts, all transcripts are shown below:

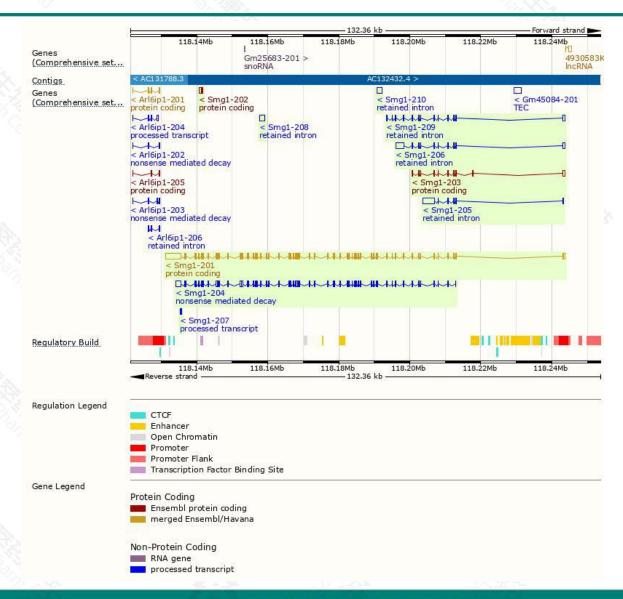
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Smg1-201	ENSMUST00000032891.15	15557	3658aa	Protein coding	CCDS40102		TSL:5 , GENCODE basic , APPRIS P1
Smg1-203	ENSMUST00000179047.3	1516	335aa	Protein coding	(4)		CDS 3' incomplete , TSL:5 ,
Smg1-202	ENSMUST00000178025.2	785	<u>96aa</u>	Protein coding	853		CDS 5' incomplete , TSL:3 ,
Smg1-204	ENSMUST00000179331.9	12147	<u>59aa</u>	Nonsense mediated decay	17.0		CDS 5' incomplete , TSL:5 ,
Smg1-207	ENSMUST00000207162.2	447	No protein	Processed transcript	323		TSL:3,
Smg1-205	ENSMUST00000179847.2	3911	No protein	Retained intron	528		TSL:1,
Smg1-206	ENSMUST00000180009.8	3630	No protein	Retained intron	-		TSL:1,
Smg1-209	ENSMUST00000208209.2	2287	No protein	Retained intron	(20)		TSL:5,
Smg1-210	ENSMUST00000208930.2	1534	No protein	Retained intron			TSL:NA ,
Smg1-208	ENSMUST00000208025.2	1460	No protein	Retained intron	-		TSL:NA ,

The strategy is based on the design of *Smg1-201* transcript, the transcription is shown below:



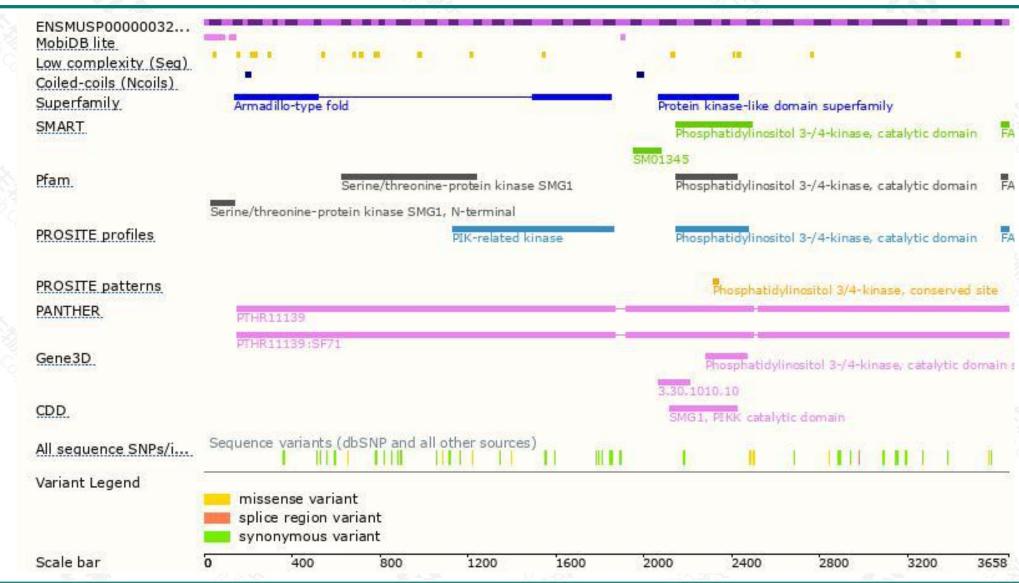
Genomic location distribution





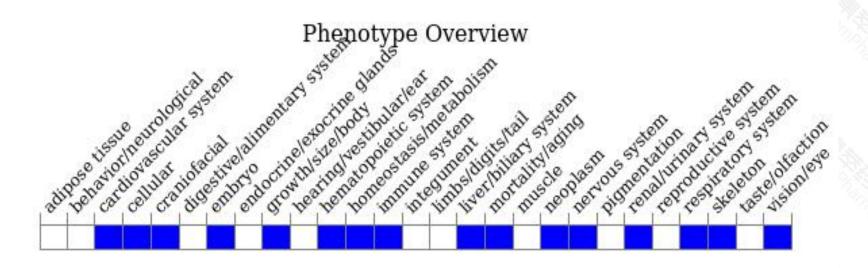
Protein domain





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 trap allele exhibit early embryonic lethality. Mice heteroygous for a gene trap allele exhibit abnormal tooth development, chronic inflammation, increased body weight, increased incidence of tumor formation and premature death.



If you have any questions, you are welcome to inquire.

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