

Skap2 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Skap2

Project type

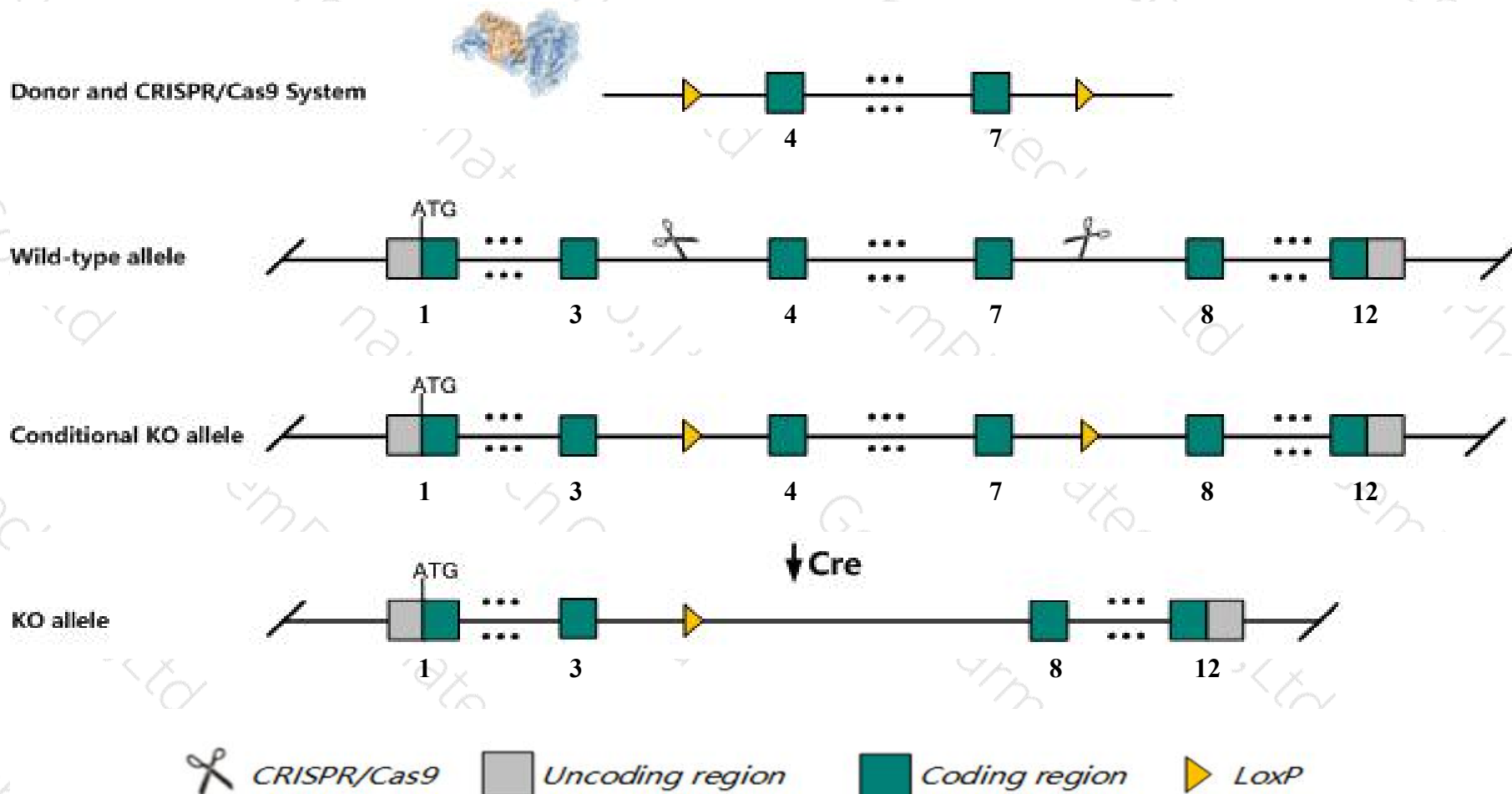
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Skap2* gene. The schematic diagram is as follows:



Technical routes

- The *Skap2* gene has 7 transcripts. According to the structure of *Skap2* gene, exon4-exon7 of *Skap2*-205 (ENSMUST00000204778.2) transcript is recommended as the knockout region. The region contains 395bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Skap2* gene. The brief process is as follows: CRISPR/Cas9 system 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 will be 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.

- According to the existing MGI data, mice homozygous for a null allele are embryonic lethal. homozygotes for a gene-trapped allele show impaired b-cell responses and b-cell adhesion, decreased susceptibility to eae, abnormal dendritic cell physiology, fast extinction of fear memory, and impaired social memory.
- Transcript 203 CDS 5' and 3' incomplete the influences is unknown.
- The *Skap2* gene is located on the Chr6. 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)

Skap2 src family associated phosphoprotein 2 [Mus musculus (house mouse)]

Gene ID: 54353, updated on 13-Mar-2020

Summary



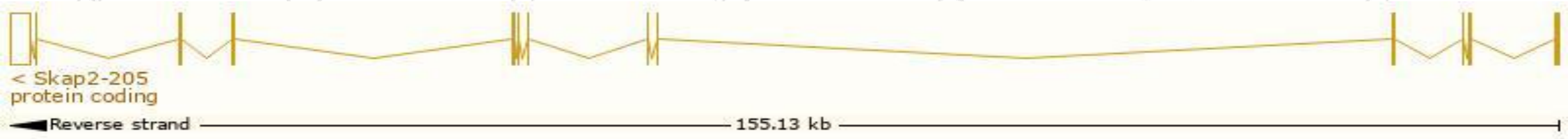
Official Symbol	Skap2 provided by MGI
Official Full Name	src family associated phosphoprotein 2 provided by MGI
Primary source	MGI:MGI:1889206
See related	Ensembl:ENSMUSG00000059182
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2610021A10Rik, AA960083, BB137539, RA70, SKAP-HOM, Saps, Scap2, mSKAP55R
Expression	Ubiquitous expression in placenta adult (RPKM 10.2), liver E18 (RPKM 9.4) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

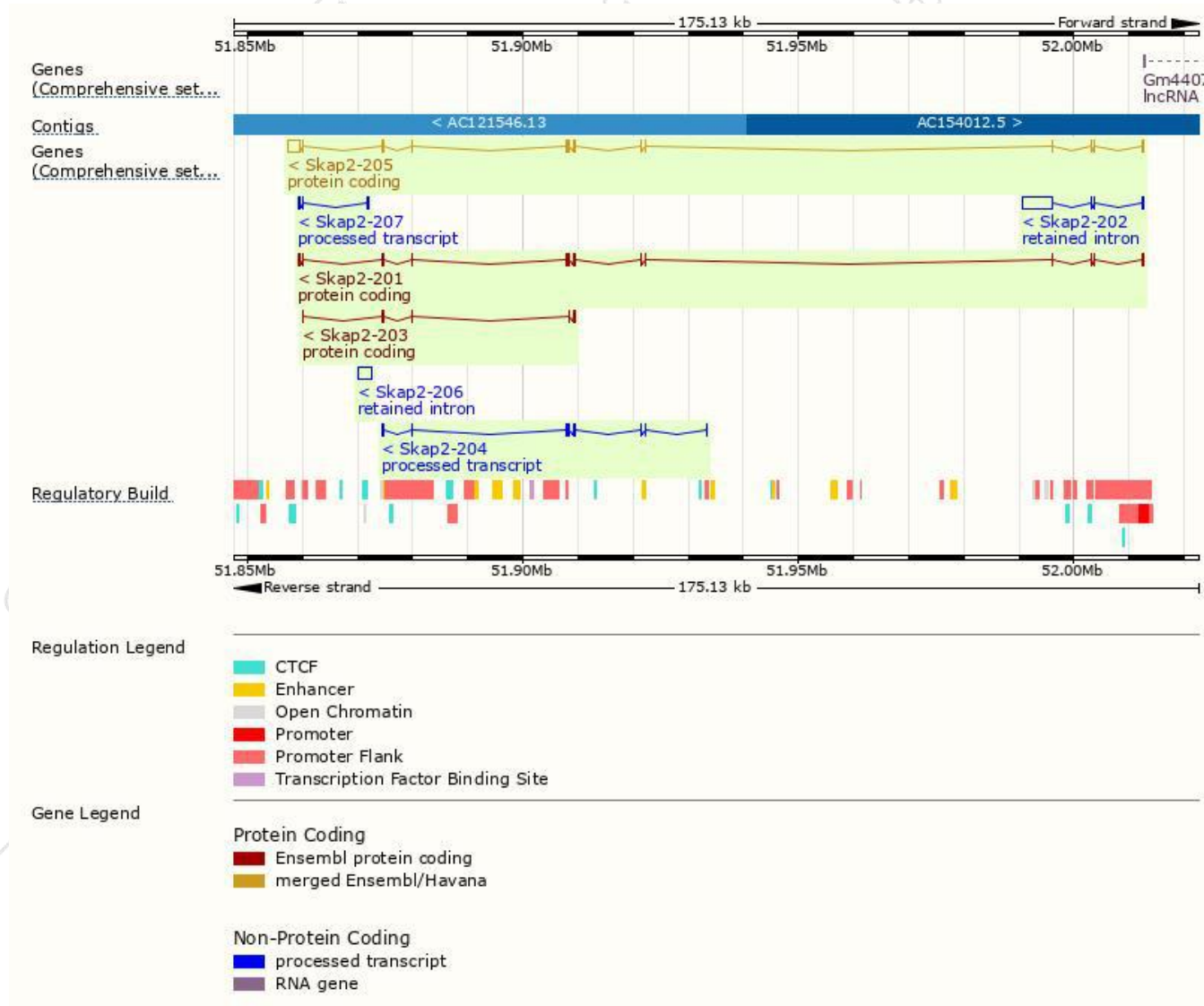
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Skap2-205	ENSMUST00000204778.2	3382	358aa	Protein coding	CCDS20137	Q3UND0	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Skap2-201	ENSMUST00000078214.7	1583	351aa	Protein coding	-	Q3UND0	TSL:1 GENCODE basic
Skap2-203	ENSMUST00000203948.1	380	126aa	Protein coding	-	A0A0N4SVW8	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Skap2-204	ENSMUST00000204178.1	751	No protein	Processed transcript	-	-	TSL:3
Skap2-207	ENSMUST00000205174.1	661	No protein	Processed transcript	-	-	TSL:3
Skap2-202	ENSMUST00000203313.1	5896	No protein	Retained intron	-	-	TSL:1
Skap2-206	ENSMUST00000205112.1	2443	No protein	Retained intron	-	-	TSL:NA

The strategy is based on the design of *Skap2-205* transcript,The transcription is shown below



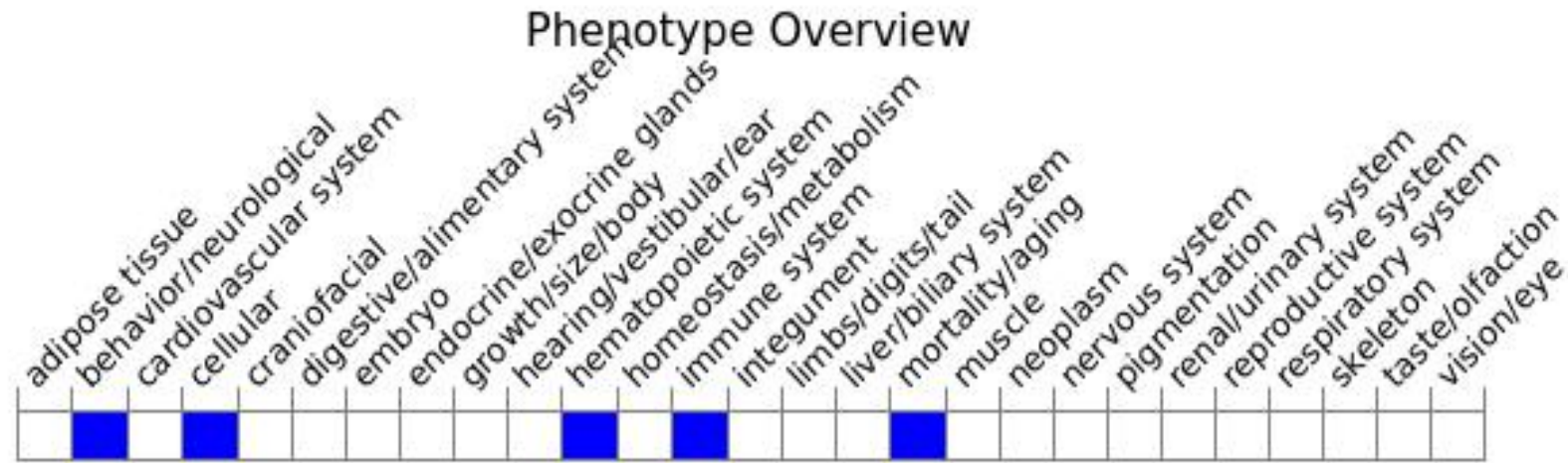
Genomic location distribution



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 null allele are embryonic lethal. Homozygotes for a gene-trapped allele show impaired B-cell responses and B-cell adhesion, decreased susceptibility to EAE, abnormal dendritic cell physiology, fast extinction of fear memory, and impaired social memory.

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

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