

# Stim2 Cas9-KO Strategy

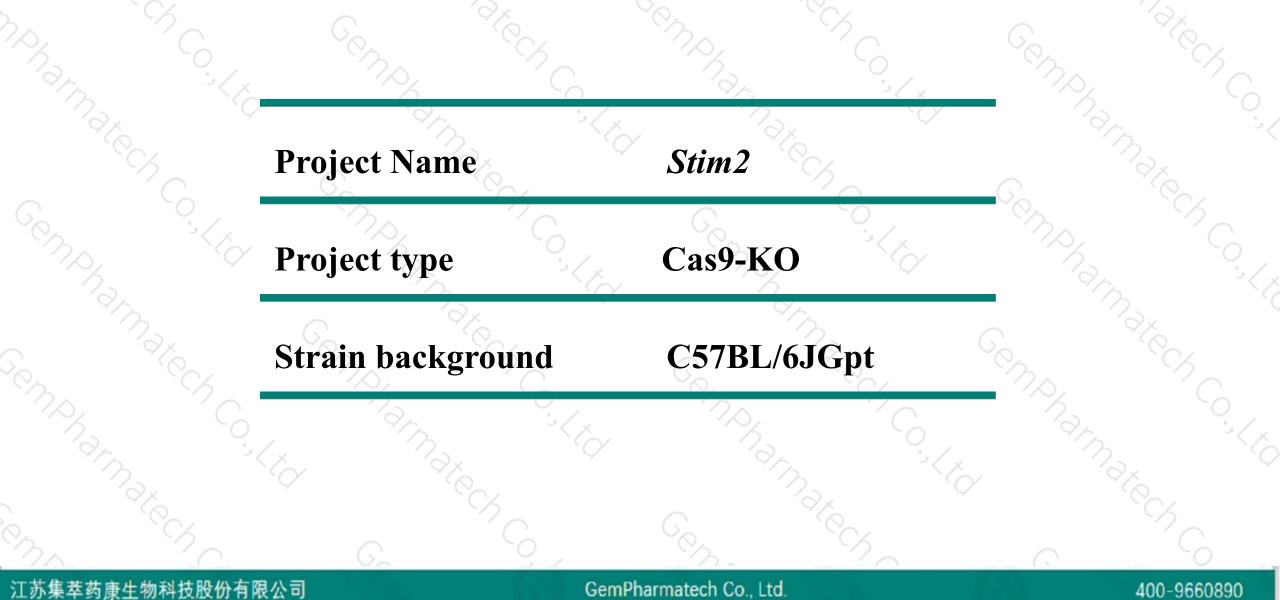
Designer: Shilei Zhu

**Reviewer: Linyan Wu** 

Design Date:2020-8-28

### **Project Overview**

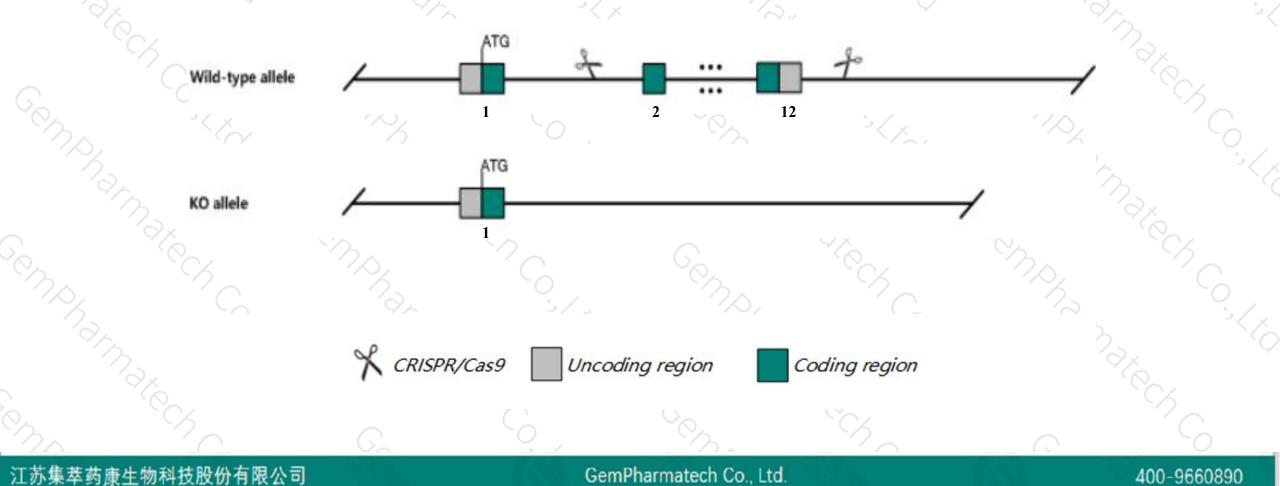




# **Knockout** strategy



This model will use CRISPR/Cas9 technology to edit the Stim2 gene. The schematic diagram is as follows:





> The *Stim2* gene has 5 transcripts. According to the structure of *Stim2* gene, exon2-exon12 of *Stim2-*201(ENSMUST00000117661.8) transcript is recommended as the knockout region. The region contains 2090bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Stim2* gene. The brief process is as follows: CRISPR/Cas9 system 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 null allele exhibit a slight growth delay and premature death while embryonic fibroblasts show reduced store-operated Ca2+ influx. Mice homozygous for a different null allele show increased neuron survival under hypoxic conditions and resistance to ischemic brain injury.
  The *Stim2* gene is located on the Chr5. 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.

# **Gene information (NCBI)**



### Stim2 stromal interaction molecule 2 [Mus musculus (house mouse)]

Gene ID: 116873, updated on 13-Mar-2020

### Summary

Official Symbol	Stim2 provided by MGI
<b>Official Full Name</b>	stromal interaction molecule 2 provided by MGI
Primary source	MGI:MGI:2151156
See related	Ensembl:ENSMUSG0000039156
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;
	Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Expression	Ubiquitous expression in lung adult (RPKM 8.3), thymus adult (RPKM 7.1) and 28 other tissuesSee more
Orthologs	human all

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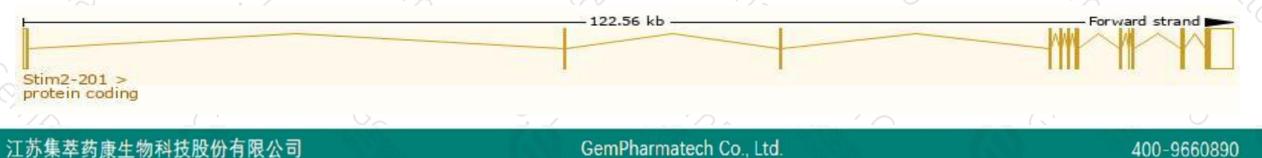
# **Transcript information (Ensembl)**



### The gene has 5 transcripts, all transcripts are shown below:

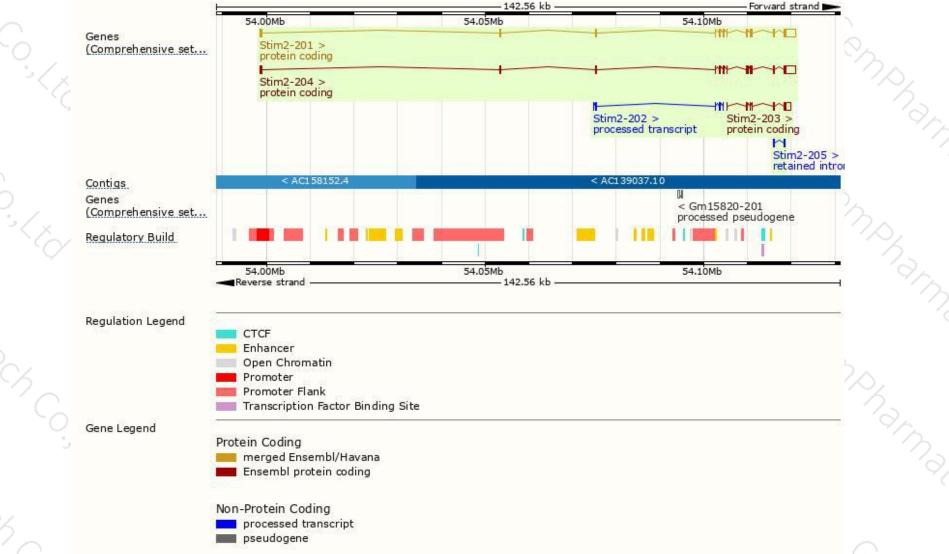
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Stim2-201	ENSMUST00000117661.8	4934	<u>746aa</u>	Protein coding	CCDS39089	A5CVE4 P83093	TSL:1 GENCODE basic APPRIS P2
Stim2-204	ENSMUST00000201469.3	4892	<u>754aa</u>	Protein coding	-	<u>11E4X8</u>	TSL:5 GENCODE basic APPRIS ALT2
Stim2-203	ENSMUST00000201198.1	2338	<u>384aa</u>	Protein coding	1	F6WBP9	CDS 5' incomplete TSL:1
Stim2-202	ENSMUST00000200873.1	553	No protein	Processed transcript			TSL:3
Stim2-205	ENSMUST00000202342.1	652	No protein	Retained intron	2	-	TSL:2

The strategy is based on the design of *Stim2-201* transcript, the transcription is shown below:



### **Genomic location distribution**





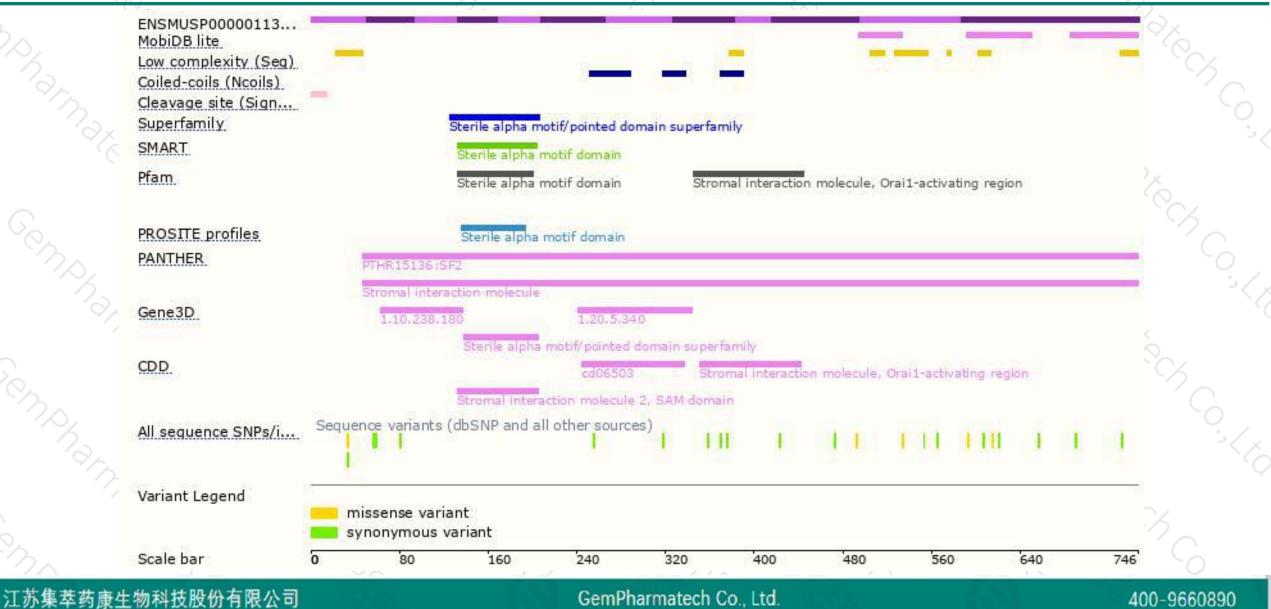
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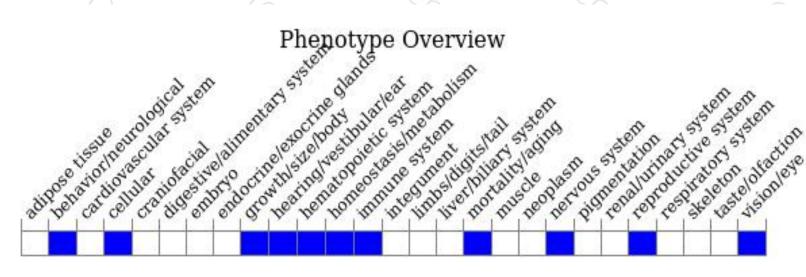
### **Protein domain**





### Mouse phenotype description(MGI)





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

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



