

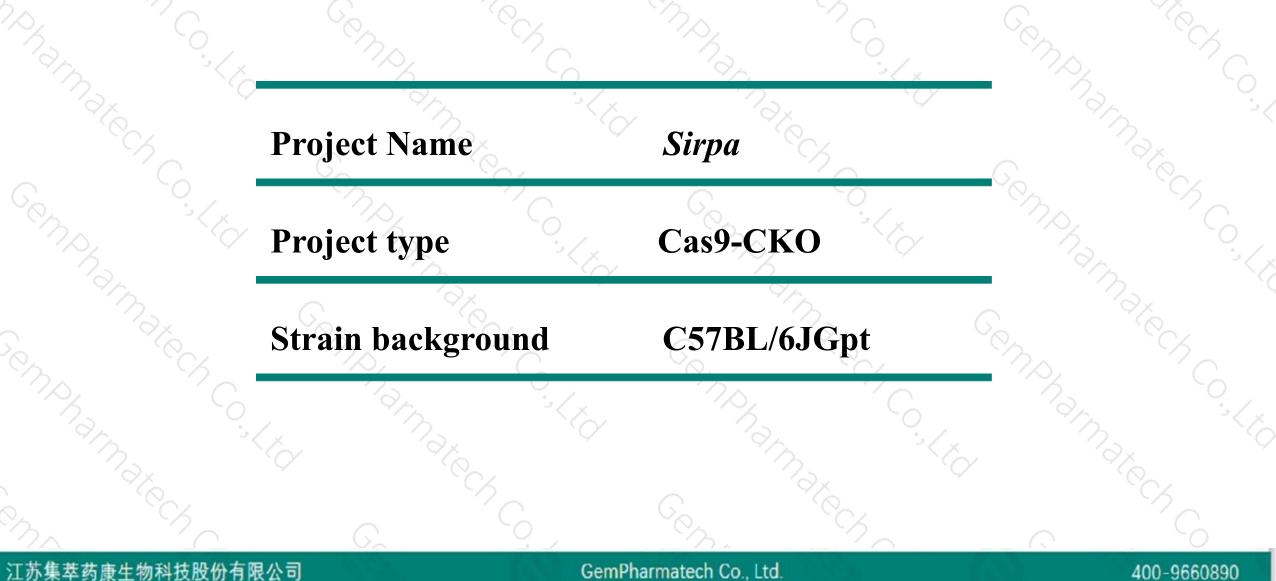
Sirpa Cas9-CKO Strategy Romphamater Control

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enphamatech,

Project Overview



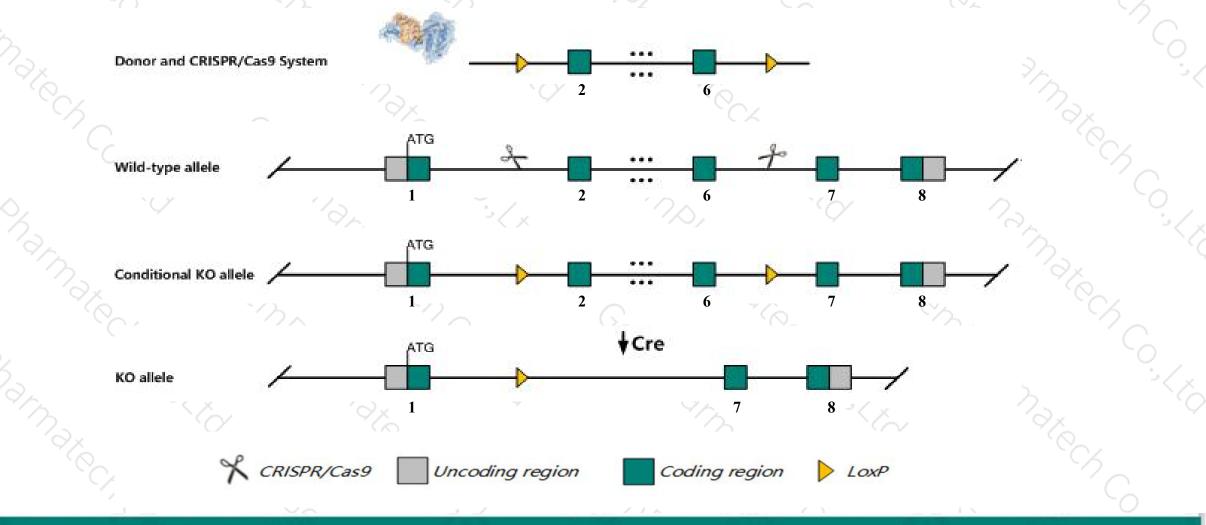


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Conditional Knockout strategy



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



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The Sirpa gene has 14 transcripts. According to the structure of Sirpa gene, exon2-exon6 of Sirpa-203 (ENSMUST00000103202.9) transcript is recommended as the knockout region. The region contains 1150bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Sirpa* 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, Homozygous null mice display mild thrombocytopenia, fatty livers, decreased body weight, decreased proportion of single positive T cells, enhanced peritoneal macrophage phagocytosis and impaired Langerhans cell migration.
- The Sirpa gene is located on the Chr2. 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)



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Sirpa signal-regulatory protein alpha [Mus musculus (house mouse)]

Gene ID: 19261, updated on 9-Apr-2019

Summary

Official SymbolSirpa provided by MGIOfficial Full Namesignal-regulatory protein alpha provided by MGIPrimary sourceMGI:MGI:108563See relatedEnsembl:ENSMUSG0000037902Gene typeprotein codingRefSeq statusVALIDATEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
Muroidea; Murinae; Mus; MusAlso knownasAl835480, Bit, CD172a, P84, Ptpns1, SHP-1, SHPS-1, SIRPExpressionBroad expression in cortex adult (RPKM 59.8), frontal lobe adult (RPKM 57.5) and 25 other tissues
See more

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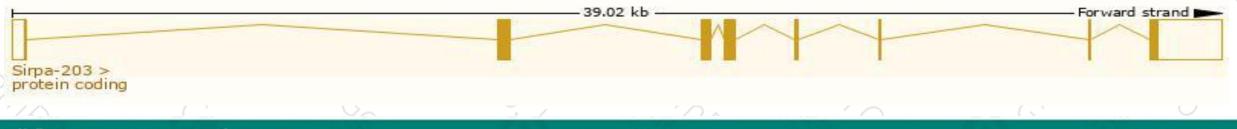
Transcript information (Ensembl)



The gene has 14 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sirpa-203	ENSMUST00000103202.9	3982	<u>509aa</u>	Protein coding	CCDS16729	<u>Q6P6l8</u>	TSL:1 GENCODE basic APPRIS P3
Sirpa-211	ENSMUST00000161620.7	3824	<u>513aa</u>	Protein coding	CCDS71142	E0CYM8	TSL:1 APPRIS ALT2
Sirpa-214	ENSMUST00000179001.7	3812	<u>509aa</u>	Protein coding	CCDS16729	<u>Q6P618</u>	TSL:5 GENCODE basic APPRIS P3
Sirpa-204	ENSMUST00000103203.7	3674	<u>509aa</u>	Protein coding	CCDS16729	Q6P618	TSL:5 GENCODE basic APPRIS P3
Sirpa-201	ENSMUST00000049262.13	3644	<u>513aa</u>	Protein coding	CCDS71142	E0CYM8	TSL:1 GENCODE basic APPRIS ALT2
Sirpa-202	ENSMUST00000099113.9	3342	<u>295aa</u>	Protein coding	CCDS71143	E9QPT7	TSL:3 GENCODE basic
Sirpa-209	ENSMUST00000160276.1	876	<u>291aa</u>	Protein coding	CCDS50708	A0A0R4J1Z7	TSL:1 GENCODE basic
Sirpa-213	ENSMUST00000163034.7	677	<u>172aa</u>	Protein coding	CCDS79845	E0CX65	TSL:3 GENCODE basic
Sirpa-208	ENSMUST00000153491.7	720	<u>158aa</u>	Protein coding	7	A2ANC1	CDS 3' incomplete TSL:5
Sirpa-206	ENSMUST00000136153.7	521	No protein	Processed transcript	-	-	TSL:5
Sirpa-210	ENSMUST00000160952.1	430	No protein	Processed transcript	-	2	TSL:5
Sirpa-207	ENSMUST00000136554.1	400	No protein	Processed transcript	14	-	TSL:5
Sirpa-205	ENSMUST00000127751.7	339	No protein	Processed transcript	1	-	TSL:3
Sirpa-212	ENSMUST00000162764.1	642	No protein	Retained intron	-	-	TSL:3
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The strategy is based on the design of Sirpa-203 transcript, The transcription is shown below

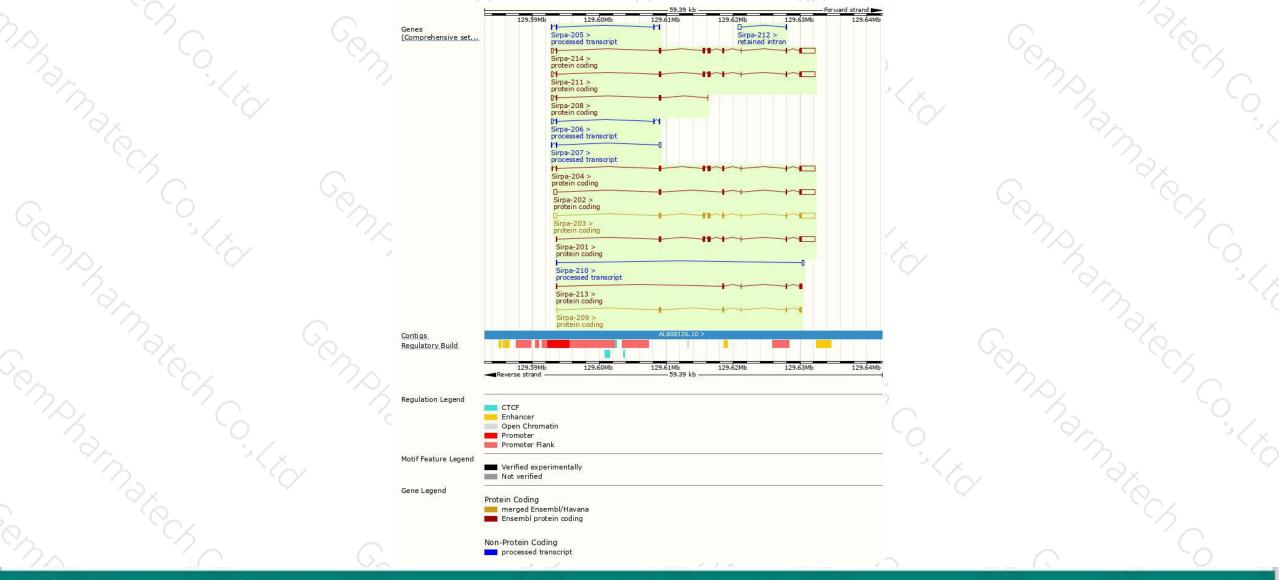


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Genomic location distribution





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Protein domain



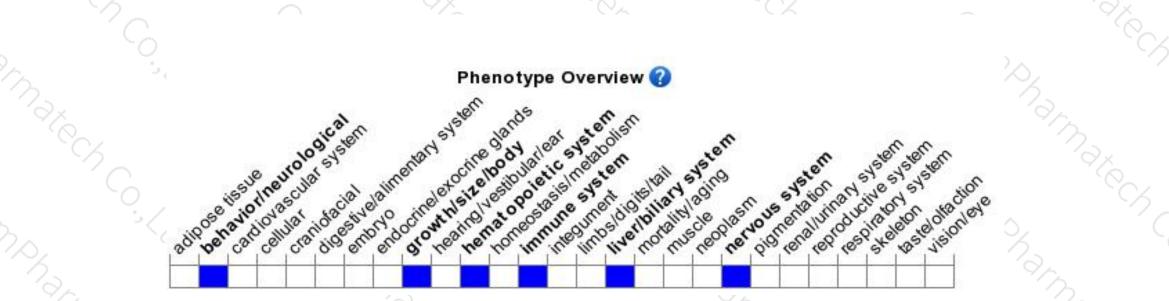


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Mouse phenotype description(MGI)





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, Homozygous null mice display mild thrombocytopenia, fatty livers, decreased body weight, decreased proportion of single positive T cells, enhanced peritoneal macrophage phagocytosis and impaired Langerhans cell migration.

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



