

Rspry1 Cas9-CKO Strategy

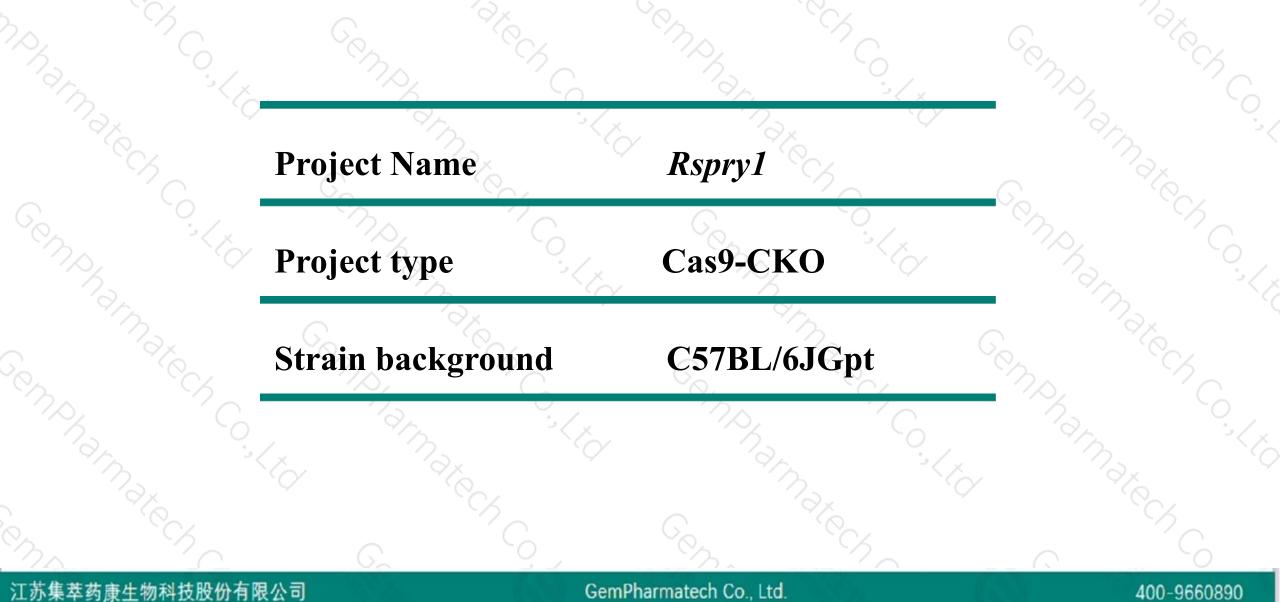
Designer: Reviewer:

Design Date:

Daohua Xu Huimin Su 2019-12-19

Project Overview

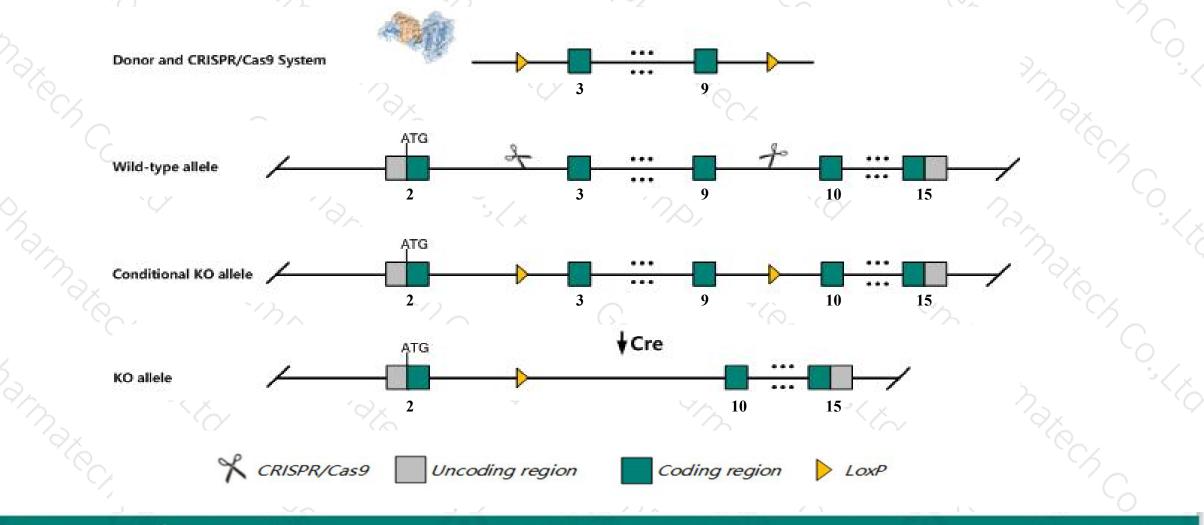




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Rspry1* gene. The schematic diagram is as follows:



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The Rspry1 gene has 7 transcripts. According to the structure of Rspry1 gene, exon3-exon9 of Rspry1-201 (ENSMUST0000060389.9) transcript is recommended as the knockout region. The region contains 667bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Rspry1* 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.



- The Rspry1 gene is located on the Chr8. 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)



\$?

Rspry1 ring finger and SPRY domain containing 1 [Mus musculus (house mouse)]

Gene ID: 67610, updated on 31-Jan-2019

Summary

Rspry1 provided by MGI
ring finger and SPRY domain containing 1 provided by MGI
MGI:MGI:1914860
Ensembl:ENSMUSG0000050079
protein coding
VALIDATED
Mus musculus
Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
Muroidea; Muridae; Murinae; Mus; Mus
4930470D19Rik, Al608258
Ubiquitous expression in testis adult (RPKM 9.0), CNS E11.5 (RPKM 8.2) and 28 other tissues See more
human all

Transcript information (Ensembl)



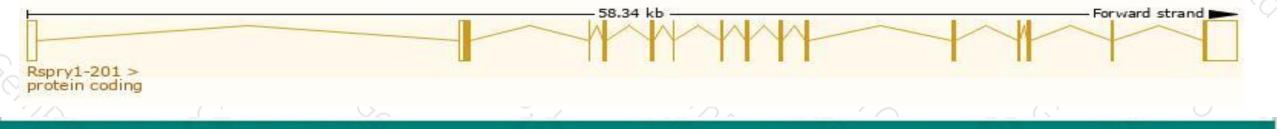
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The gene has 7 transcripts, all transcripts are shown below:

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Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rspry1-201	ENSMUST0000060389.9	3852	<u>576aa</u>	Protein coding	CCDS22544	Q8BVR6	TSL:1 GENCODE basic APPRIS P1
Rspry1-205	ENSMUST00000211983.1	3397	<u>576aa</u>	Protein coding	CCDS22544	Q8BVR6	TSL:1 GENCODE basic APPRIS P1
Rspry1-202	ENSMUST00000121101.1	3092	<u>117aa</u>	Protein coding	<u>84</u>	Q8BVR6	TSL:1 GENCODE basic
Rspry1-207	ENSMUST00000212729.1	2800	<u>452aa</u>	Protein coding	62	Q8BVR6	TSL:1 GENCODE basic
Rspry1-204	ENSMUST00000211941.1	4157	No protein	Retained intron	67		TSL:NA
Rspry1-206	ENSMUST00000212014.1	3181	No protein	Retained intron	8 .	-	TSL:NA
Rspry1-203	ENSMUST00000154035.1	745	No protein	Retained intron	2 .	<u>.</u>	TSL:3

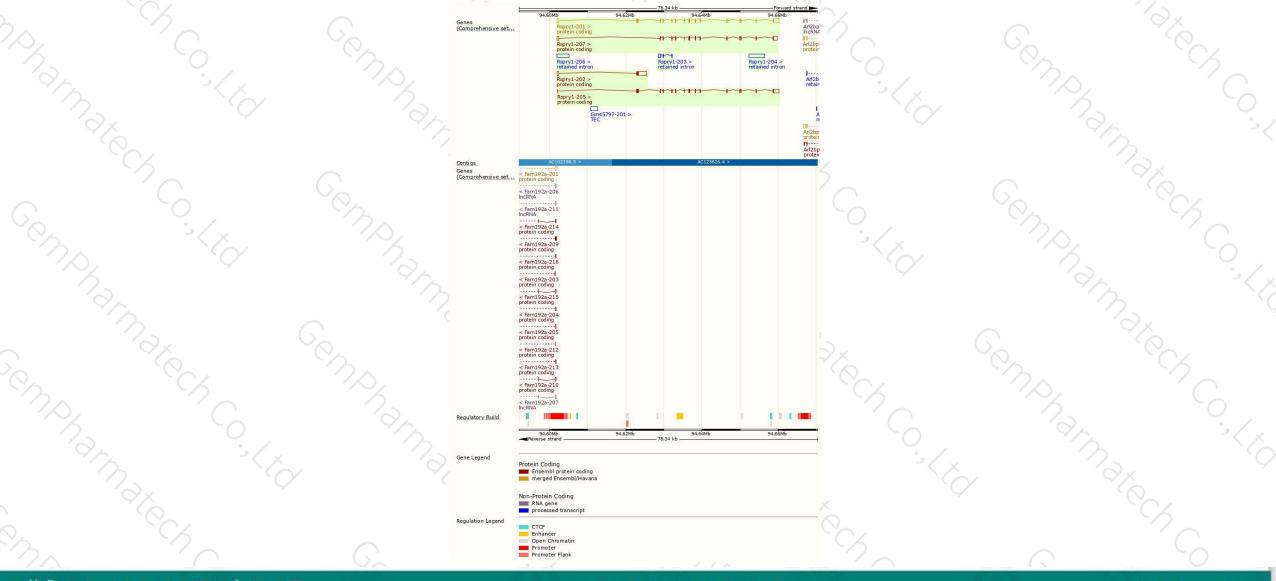
The strategy is based on the design of Rspry1-201 transcript, The transcription is shown below



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Genomic location distribution





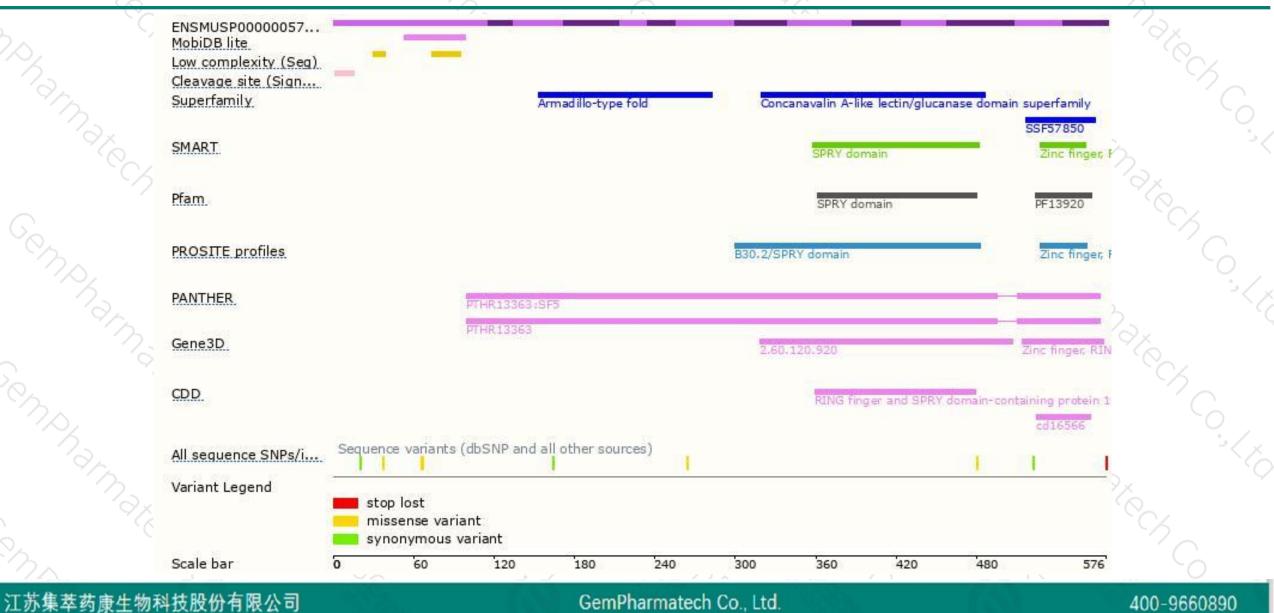
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Protein domain







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



