

Rnmt Cas9-CKO Strategy

Designer:

Reviewer:

Design Date:

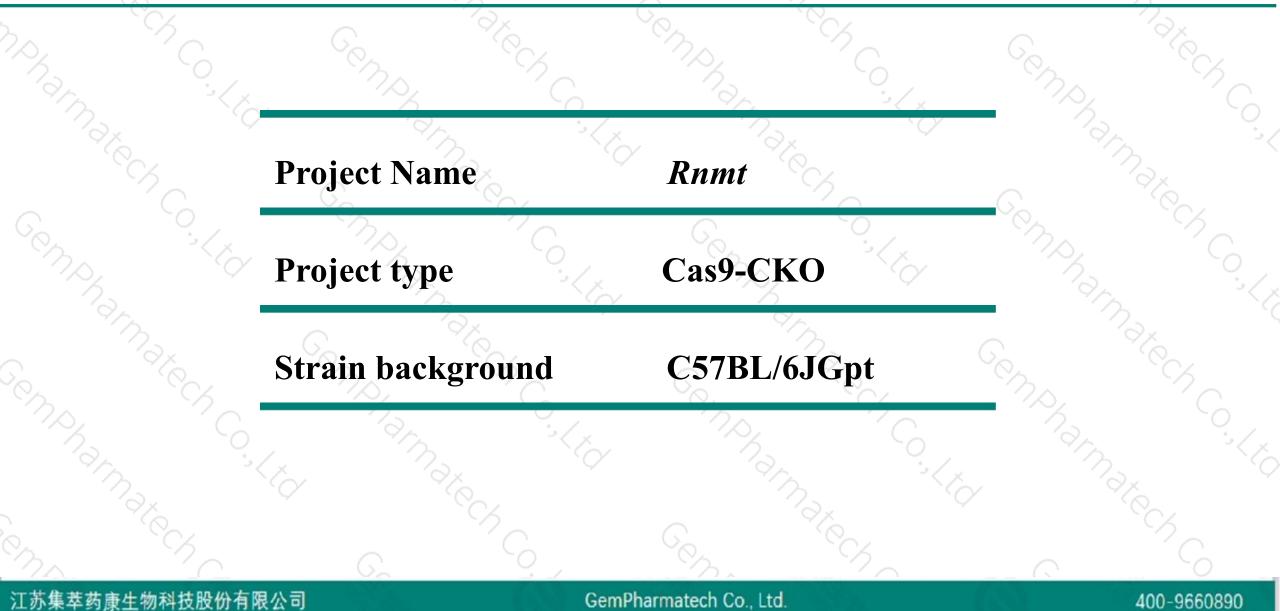
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2020-5-6

Project Overview



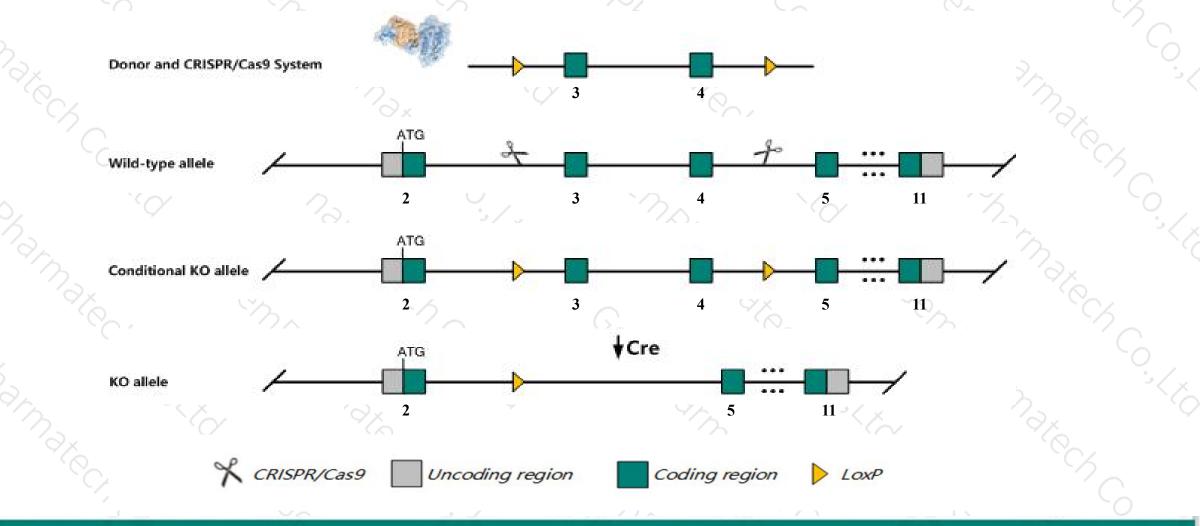


Conditional Knockout strategy



400-9660890

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



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The Rnmt gene has 8 transcripts. According to the structure of Rnmt gene, exon3-exon4 of Rnmt-201 (ENSMUST0000009679.10) transcript is recommended as the knockout region. The region contains 268bp coding sequence. Knock out the region will result in disruption of protein function.

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



- ≻Transcripts 206,207 may not be affected. The effect of transcripts 203,204 is unknown.
- The Rnmt gene is located on the Chr18. 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)



\$?

Rnmt RNA (guanine-7-) methyltransferase [Mus musculus (house mouse)]

Gene ID: 67897, updated on 13-Mar-2020

Summary

127212733 1722 18 33								
Official Symbol	Rnmt provided by <u>MGI</u>							
Official Full Name	VA (guanine-7-) methyltransferase provided byMGI							
Primary source	<u>GI:MGI:1915147</u>							
See related	nsembl:ENSMUSG0000009535							
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							
Also known as	2610002P10Rik, Al848273, Rg7mt1, mKIAA0398							
Expression	Broad expression in CNS E18 (RPKM 9.2), CNS E14 (RPKM 8.1) and 25 other tissues See more							
Orthologs	human all							

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Transcript information (Ensembl)



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The gene has 8 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rnmt-201	ENSMUST0000009679.10	5008	<u>465aa</u>	Protein coding	CCDS37852		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
Rnmt-202	ENSMUST00000025427.13	1852	<u>410aa</u>	Protein coding	CCDS50314	Q9D0L8	TSL:1 GENCODE basic
Rnmt-205	ENSMUST00000139111.1	965	<u>240aa</u>	Protein coding	120	D3YYS7	CDS 3' incomplete TSL:3
nmt-204	ENSMUST00000131075.7	757	<u>205aa</u>	Protein coding	121	<u>D3Z478</u>	CDS 3' incomplete TSL:5
Rnmt-203	ENSMUST00000129849.1	537	<u>124aa</u>	Protein coding		<u>D3Z4K1</u>	CDS 3' incomplete TSL:2
nmt-207	ENSMUST00000148937.1	3791	No protein	Processed transcript	(1 1)		TSL:1
nmt-208	ENSMUST00000151833.1	2474	No protein	Retained intron	120	20	TSL:1
nmt-206	ENSMUST00000139554.1	748	No protein	Retained intron	1027	2	TSL:3

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

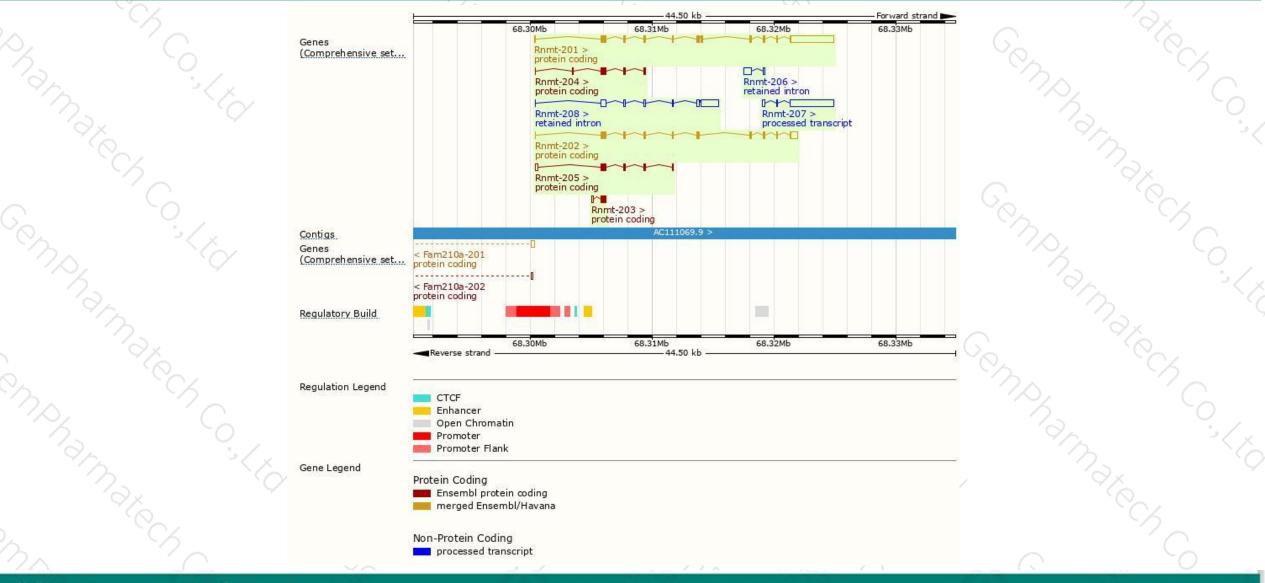
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Rnmt-201 > protein coding						1
10	_(^ /		(\sim

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Genomic location distribution



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Protein domain



			S'OL							
2	ENSMUSP00000009 MobiDB lite Low complexity (Seg) Superfamily		S-;	adenosyl-L-methionine-de	ependent methyltransferas	e .	2°.			
	<u>Pfam</u>		mRNA (gua	anine-N(7))-methyltransfe	erase domain					
	PROSITE profiles		mRNA (guanin	ne-N(7))-methyltransferas	e domain					
	PIRSF	mRNA cap gua	nine-N7 methyltransferas	e, eukaryotes						
	PANTHER	mRNA cap guanine-N7 methyltransferase								
	CDD.	PTHR1218	9:8F2	cd02440	-					
	All sequence SNPs/i	Sequence variants	(dbSNP and all other so	ources)	I I II					
<	Variant Legend	missense vari								
	Scale bar	0 40	80 120 16	50 200 240	280 320	360 400	465			
	°C/		S.	George a	°C/		$^{\sim}$ C			

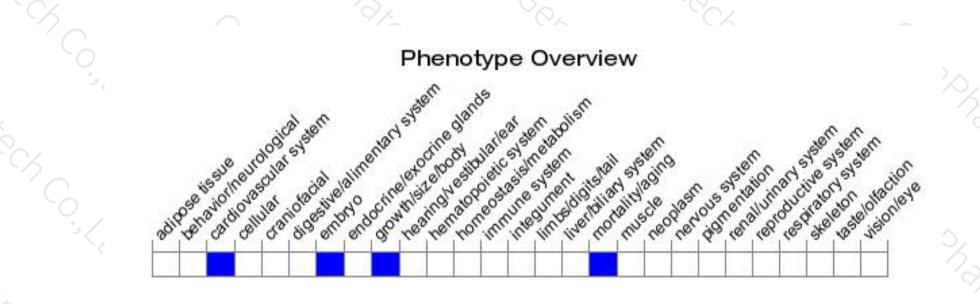
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Mouse phenotype description(MGI)





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



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



