

# Runx1t1 Cas9-CKO Strategy

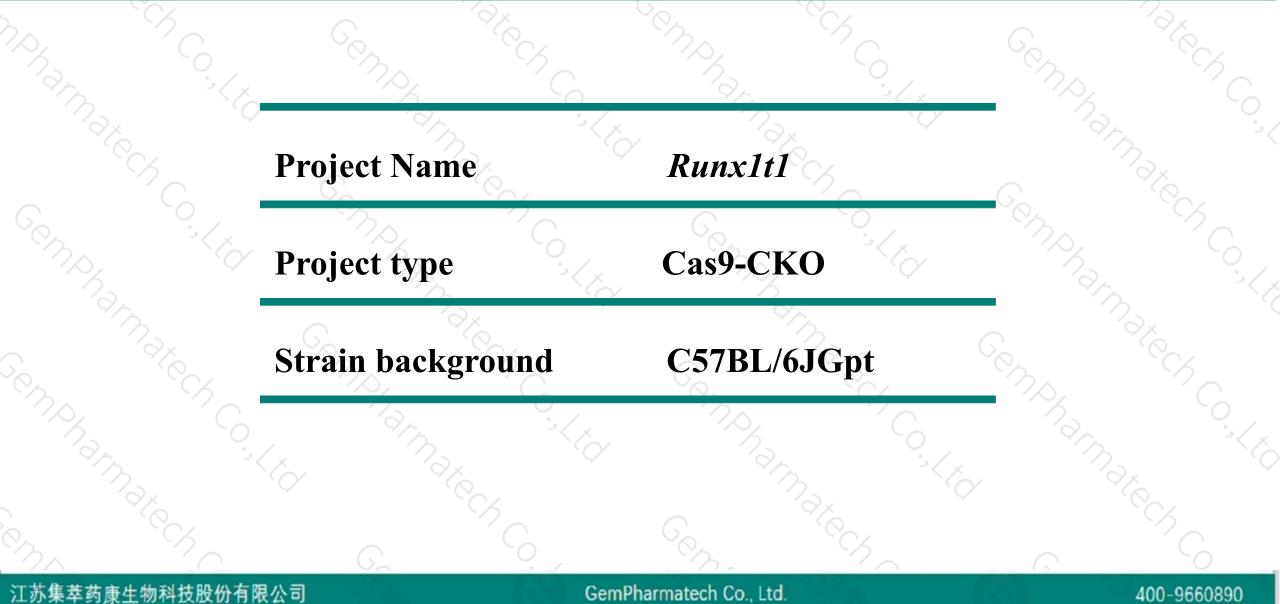
Designer: Reviewer:

**Design Date:** 

Daohua Xu Huimin Su 2019-9-25

# **Project Overview**



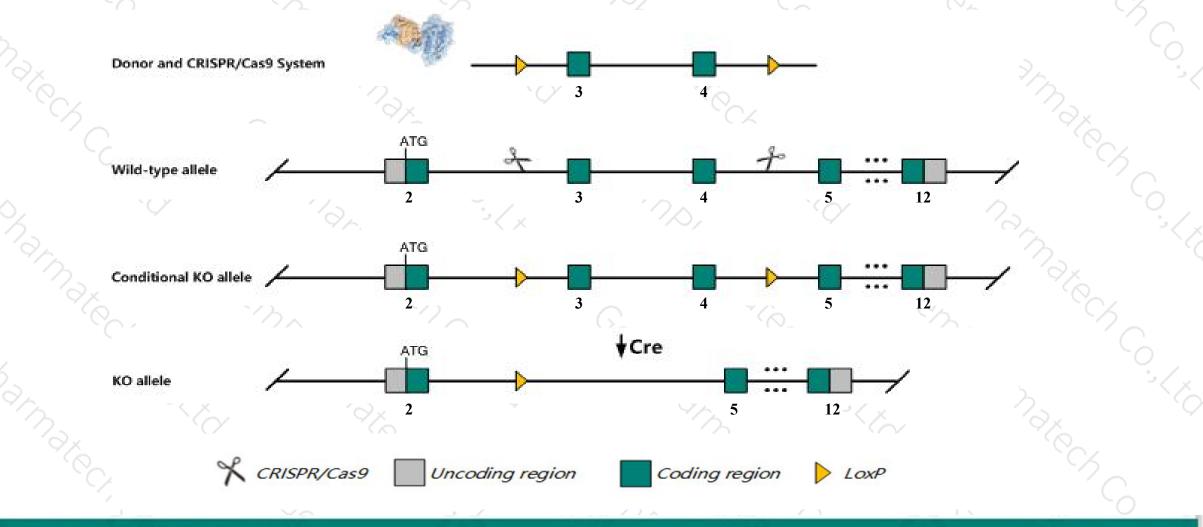


## **Conditional Knockout strategy**



400-9660890

This model will use CRISPR/Cas9 technology to edit the Runx1t1 gene. The schematic diagram is as follows:



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The Runx1t1 gene has 6 transcripts. According to the structure of Runx1t1 gene, exon3-exon4 of Runx1t1-204 (ENSMUST00000105566.8) transcript is recommended as the knockout region. The region contains 380bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Runx1t1* 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 disruption of this gene results in increased perinatal lethality and surviving animals show severe growth retardation. The midgut is absent in 25% of mutant animals which could explain increased perinatal mortality. Surviving animals display thinned intestinal walls and dilated lumens.
- The Runx1t1 gene is located on the Chr4. 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.

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# **Gene information (NCBI)**



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#### Runx1t1 RUNX1 translocation partner 1 [Mus musculus (house mouse)]

Gene ID: 12395, updated on 31-Jan-2019

#### Summary

Official Symbol	Runx1t1 provided by MGI
Official Full Name	RUNX1 translocation partner 1 provided by MGI
Primary source	MGI:MGI:104793
See related	Ensembl:ENSMUSG0000006586
Gene type	protein coding
<b>RefSeq status</b>	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	Cbfa2t1h, ETO, MTG8
Expression	Broad expression in whole brain E14.5 (RPKM 6.0), CNS E18 (RPKM 4.8) and 15 other tissues See more
Orthologs	human all

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# **Transcript information (Ensembl)**



The gene has 6 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Runx1t1-204	ENSMUST00000105566.8	6090	<u>604aa</u>	Protein coding	CCDS51125	B1AXH8	TSL:5 GENCODE basic APPRIS ALT2
Runx1t1-201	ENSMUST0000006761.9	5783	<u>584aa</u>	Protein coding	CCDS51124	<u>Q3UQX8</u>	TSL:1 GENCODE basic APPRIS ALT2
Runx1t1-202	ENSMUST0000098256.3	3317	<u>577aa</u>	Protein coding	CCDS17978	B1AXH9 Q61909	TSL:1 GENCODE basic APPRIS P3
Runx1t1-203	ENSMUST0000098257.9	2332	<u>604aa</u>	Protein coding	CCDS51125	B1AXH8	TSL:1 GENCODE basic APPRIS ALT2
Runx1t1-206	ENSMUST00000150583.7	2062	No protein	IncRNA	5	7	TSL:1
Runx1t1-205	ENSMUST00000139736.1	405	No protein	IncRNA		-	TSL:3

The strategy is based on the design of Runx1t1-204 transcript, The transcription is shown below

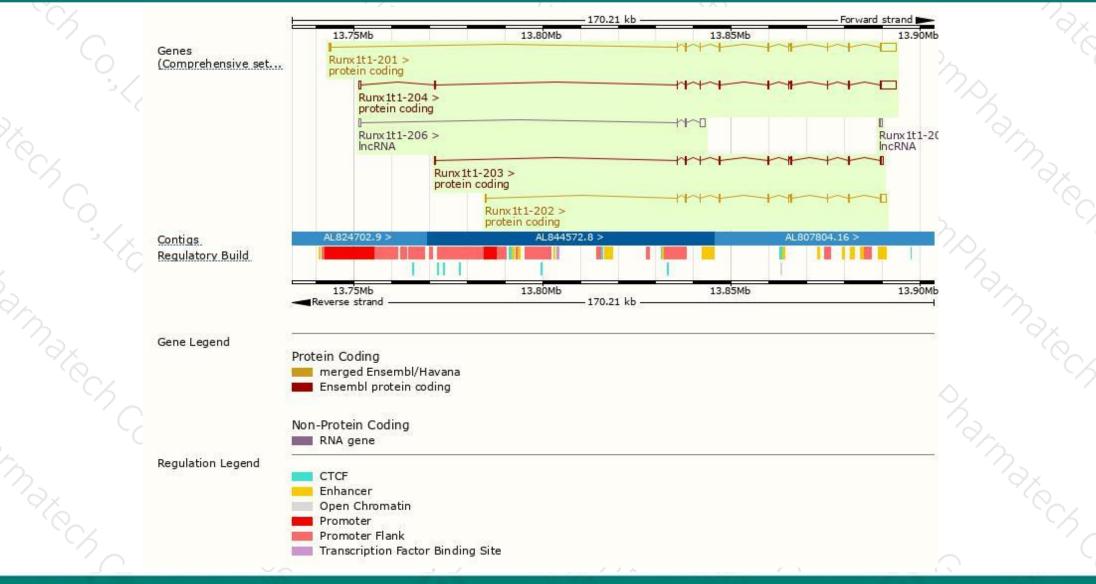


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### **Genomic location distribution**



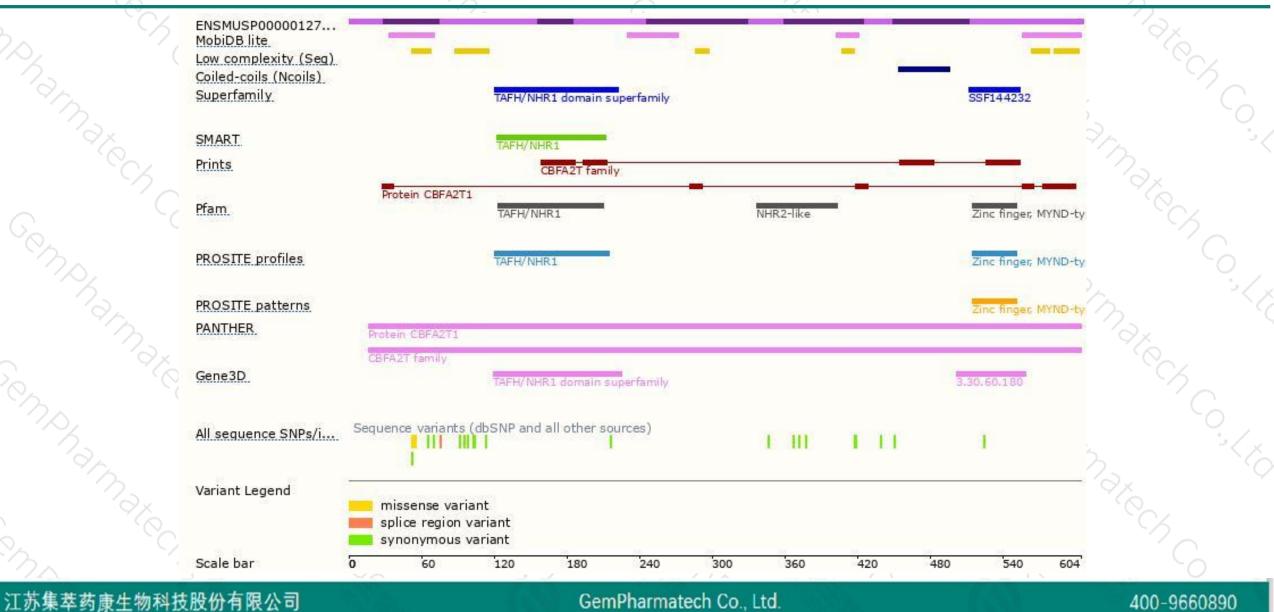


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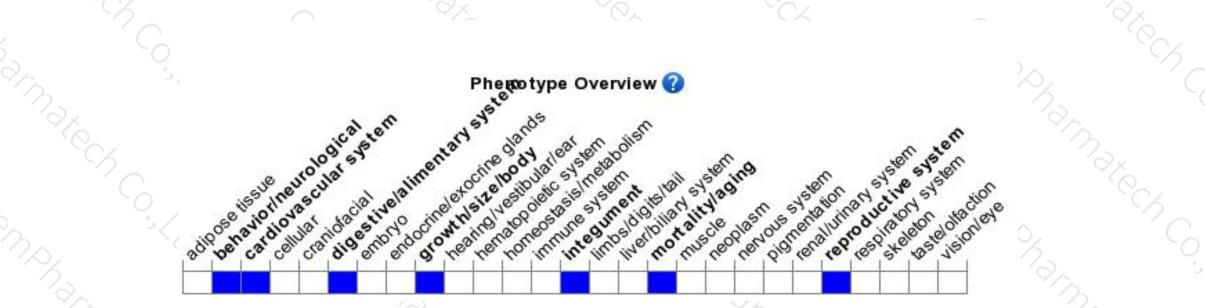
### **Protein domain**





### 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



