Runx1 Cas9-CKO Strategy

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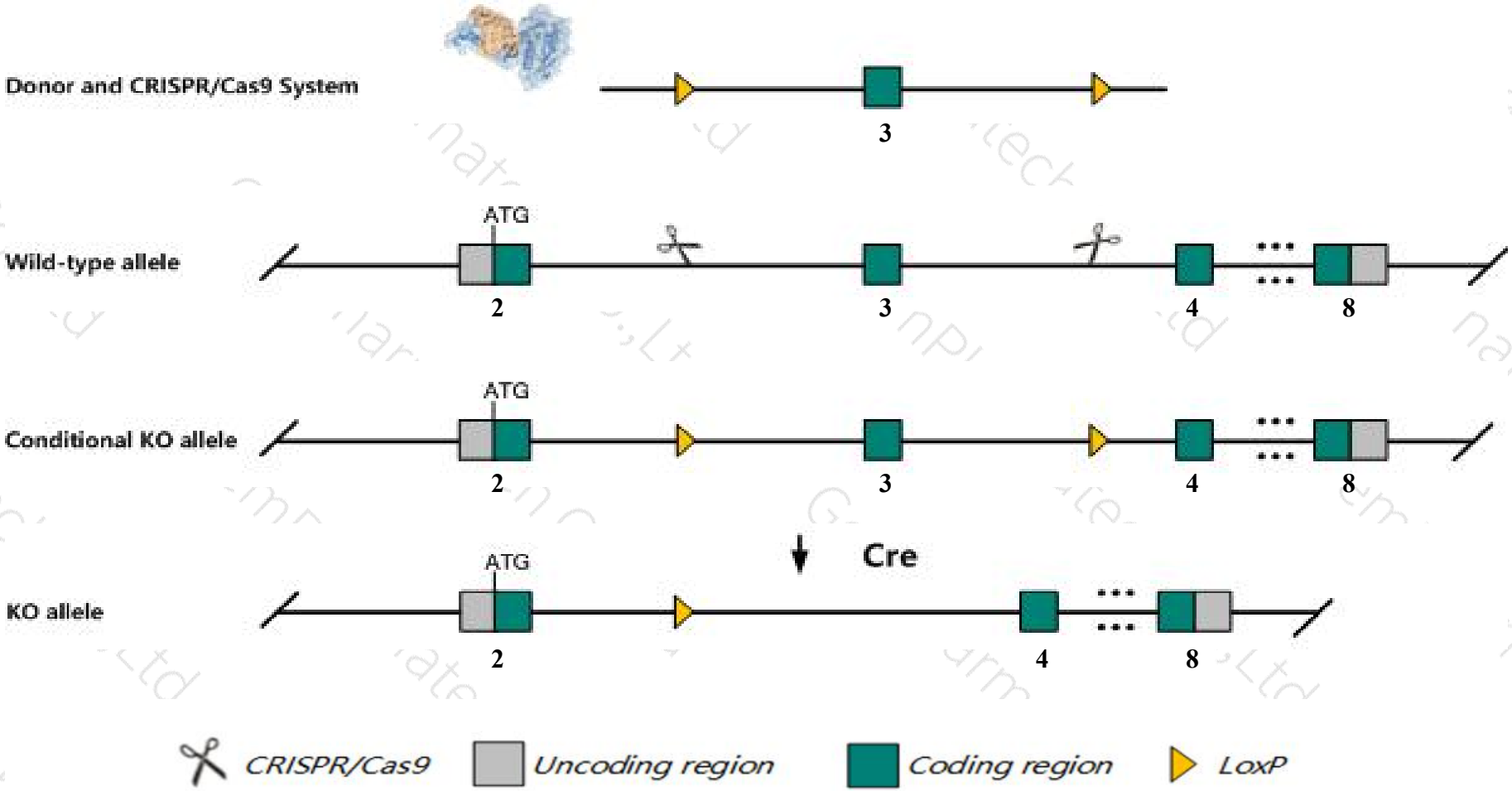
Project Overview

Project Name	***Runx1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Runx1* gene. The schematic diagram is as follows:



Technical routes

➢ The *Runx1* gene has 7 transcripts. According to the structure of *Runx1* gene, exon3 of *Runx1-201* (ENSMUST00000023673.13) transcript is recommended as the knockout region.The region contains 254bp coding sequence. Knock out the region will result in disruption of protein function.

➢This strategy does not affect transcript 204.

➢ In this project we use CRISPR/Cas9 technology to modify *Runx1* 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.

Notice

- According to the existing MGI data,Mutations affect hematopoiesis, and in some cases result in defective angiogenesis and intraventricular hemorrhage. Null homozygotes die by embryonic day 12.5; heterozygotes have reduced erythroid and myeloid progenitor numbers.

- The *Runx1* gene is located on the Chr16. 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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Runx1 runt related transcription factor 1 [Mus musculus (house mouse)]

Gene ID: 12394, updated on 9-Apr-2019

Summary

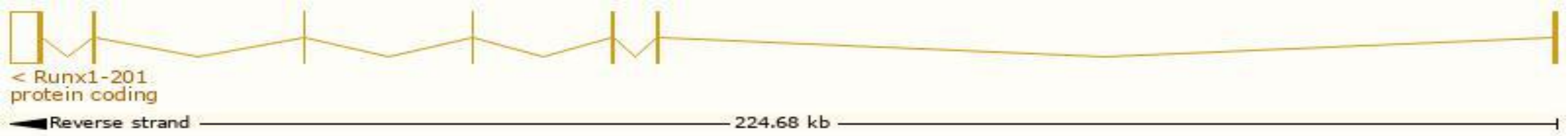
Official Symbol	Runx1 provided by MGI
Official Full Name	runt related transcription factor 1 provided by MGI
Primary source	MGI:MGI:99852
See related	Ensembl:ENSMUSG00000022952
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AML1, CBF-alpha-2, Cbfa2, Pebp2a2, Pebpa2b
Expression	Broad expression in thymus adult (RPKM 11.2), lung adult (RPKM 5.4) and 16 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

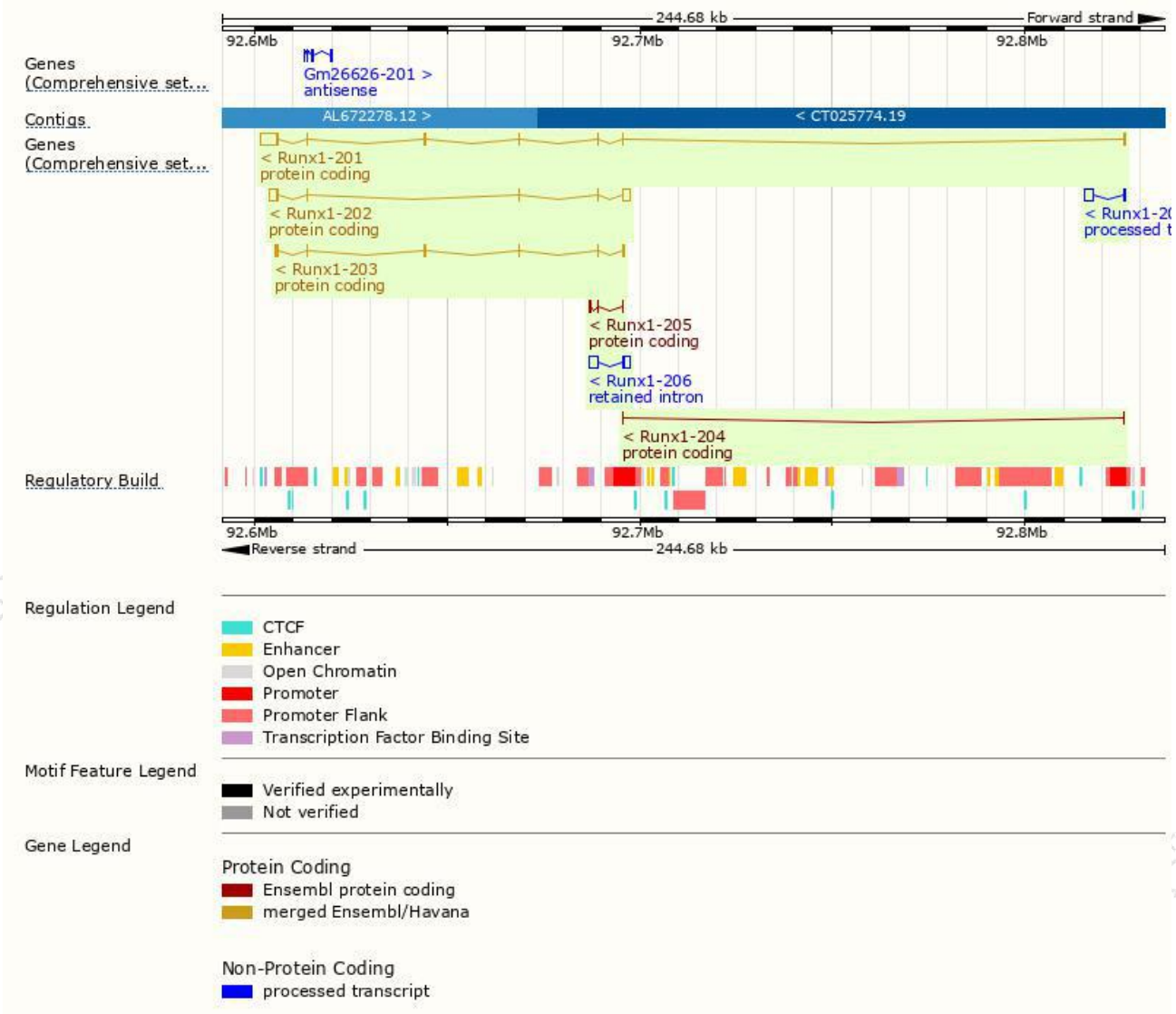
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Runx1-201	ENSMUST00000023673.13	5861	465aa	Protein coding	CCDS49917	Q3UM65	TSL:1 GENCODE basic APPRIS P4
Runx1-202	ENSMUST00000113956.9	4738	387aa	Protein coding	CCDS28339	G3X9W7	TSL:1 GENCODE basic
Runx1-203	ENSMUST00000168195.7	1985	451aa	Protein coding	CCDS49916	G3UWD2	TSL:1 GENCODE basic APPRIS ALT2
Runx1-205	ENSMUST00000187242.2	716	63aa	Protein coding	-	A0A087WPL0	CDS 5' incomplete TSL:5
Runx1-204	ENSMUST00000186296.1	336	68aa	Protein coding	-	A0A087WR05	CDS 3' incomplete TSL:3
Runx1-207	ENSMUST00000189679.1	2538	No protein	Processed transcript	-	-	TSL:1
Runx1-206	ENSMUST00000189126.1	3815	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of *Runx1-201* transcript,The transcription is shown below

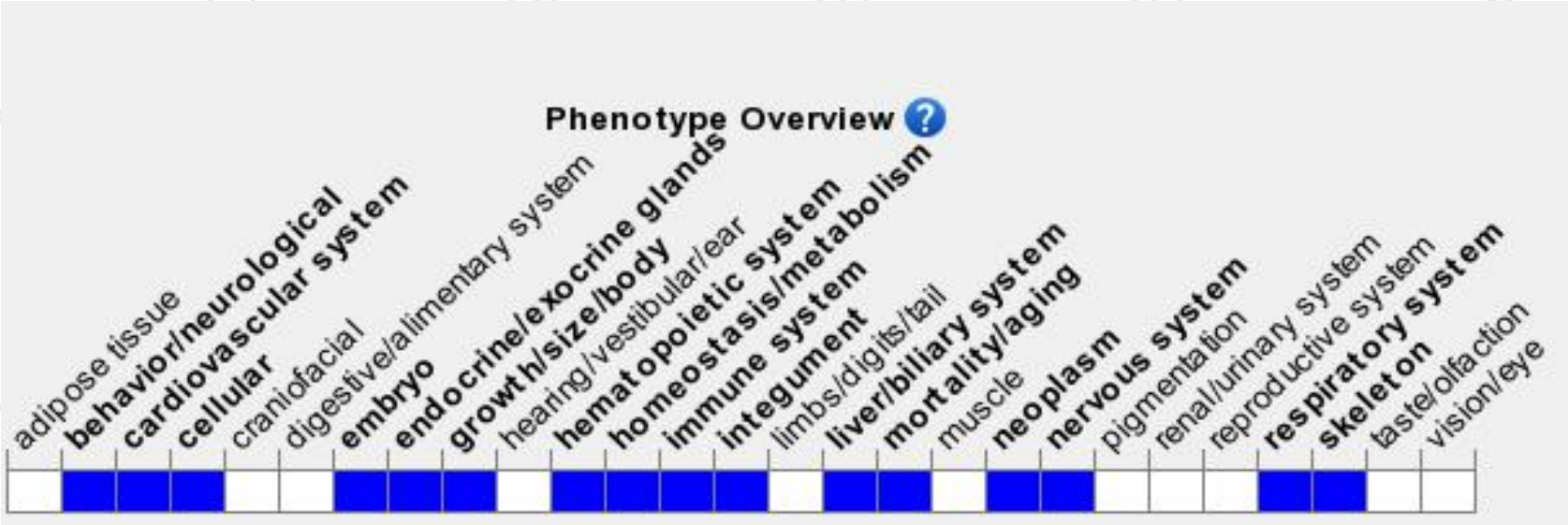


Genomic location distribution



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.

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