Rora Cas9-CKO Strategy

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Project Overview

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

Conditional Knockout strategy

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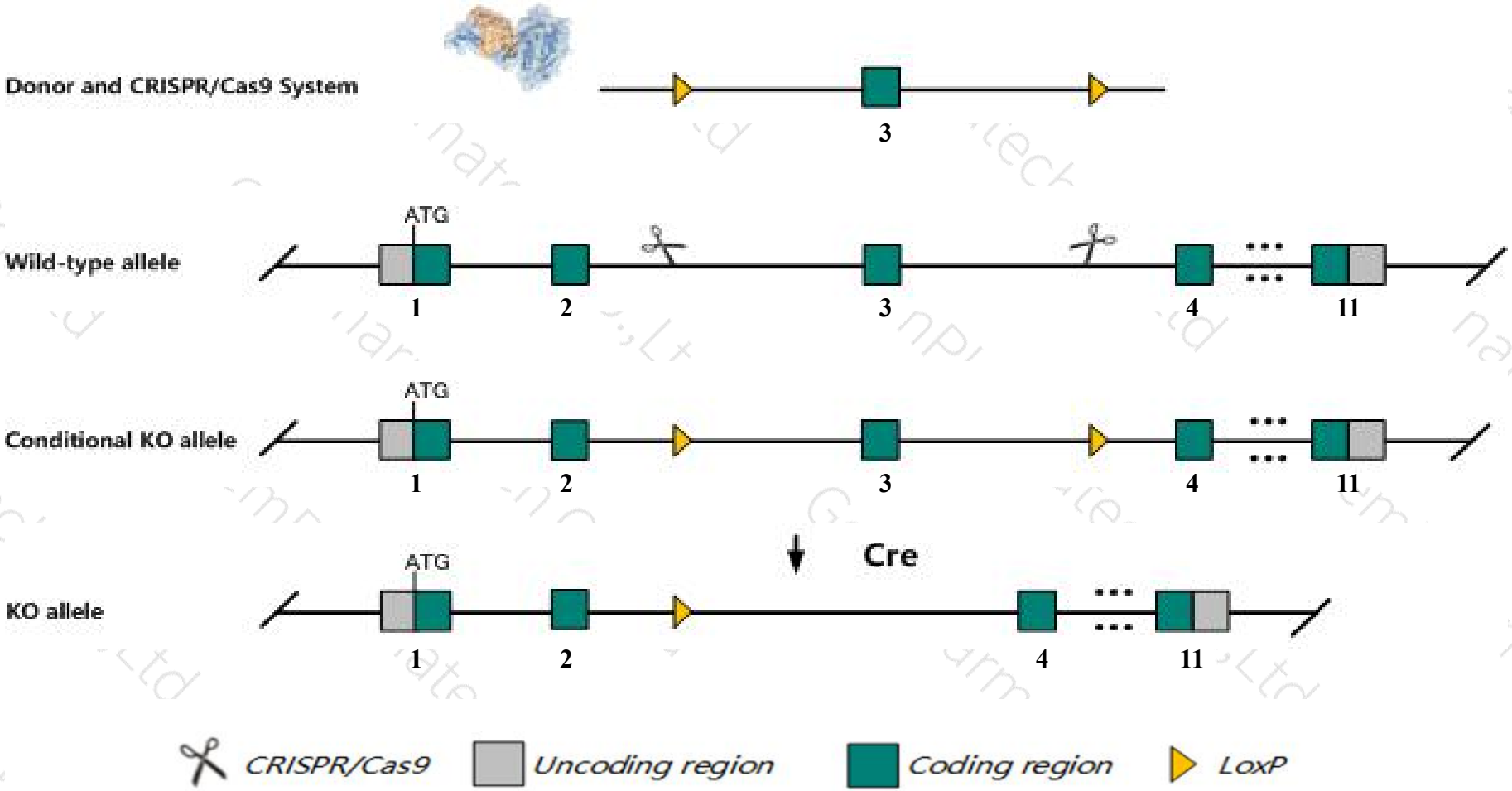
This model will use CRISPR/Cas9 technology to edit the *Rora* gene. The schematic diagram is as follows:



Technical routes

- The *Rora* gene has 6 transcripts. According to the structure of *Rora* gene, exon3 of *Rora-201* (ENSMUST00000034766.13) transcript is recommended as the knockout region.The region contains 86bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Rora* 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,Homozygotes for null mutations exhibit ataxia, cerebellar dysgenesis, impaired Purkinje and granule cell development, olfactory defects, hypoalphalipoproteinemia, and death around 4 weeks. Heterozygotes show slow Purkinje cell dedritic atrophy and loss.

- The *Rora* gene is located on the Chr9. 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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Rora RAR-related orphan receptor alpha [Mus musculus (house mouse)]

Gene ID: 19883, updated on 26-Feb-2019

Summary

Official Symbol	Rora provided by MGI
Official Full Name	RAR-related orphan receptor alpha provided by MGI
Primary source	MGI:MGI:104661
See related	Ensembl:ENSMUSG00000032238
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	9530021D13Rik, Nr1f1, ROR1, ROR2, ROR3, nmf267, sg, staggerer, tmgc26
Summary	The protein encoded by this gene is a member of the NR1 subfamily of nuclear hormone receptors. It can bind as a monomer or as a homodimer to hormone response elements upstream of several genes to enhance the expression of those genes. The encoded protein has been shown to interact with NM23-2, a nucleoside diphosphate kinase involved in organogenesis and differentiation, as well as with NM23-1, the product of a tumor metastasis suppressor candidate gene. Also, it has been shown to aid in the transcriptional regulation of some genes involved in circadian rhythm. Three transcript variants encoding different isoforms have been found for this gene. [provided by RefSeq, Feb 2014]
Expression	Broad expression in cerebellum adult (RPKM 8.5), cortex adult (RPKM 3.0) and 19 other tissues See more
Orthologs	human all

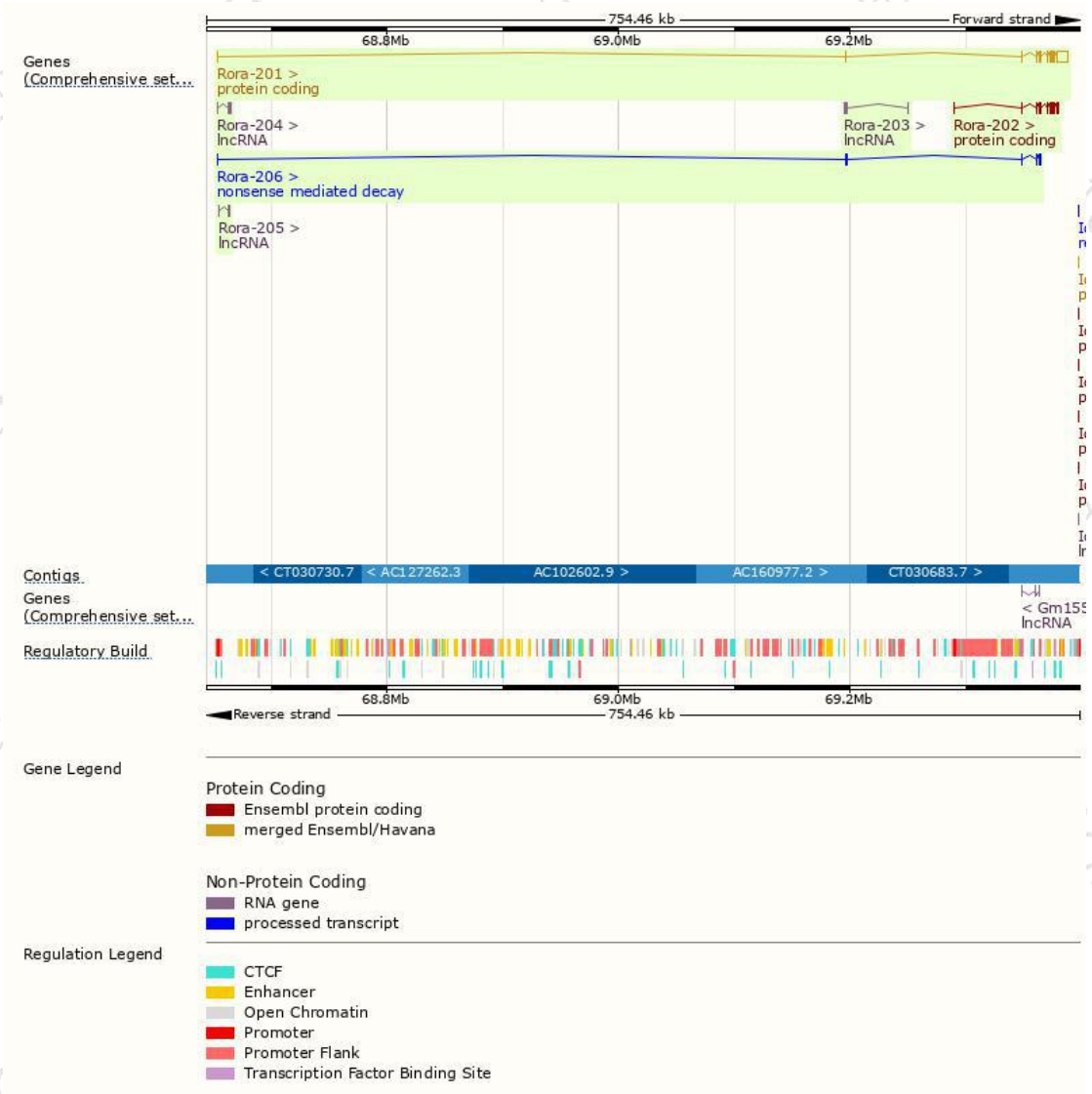
Transcript information（Ensembl）

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

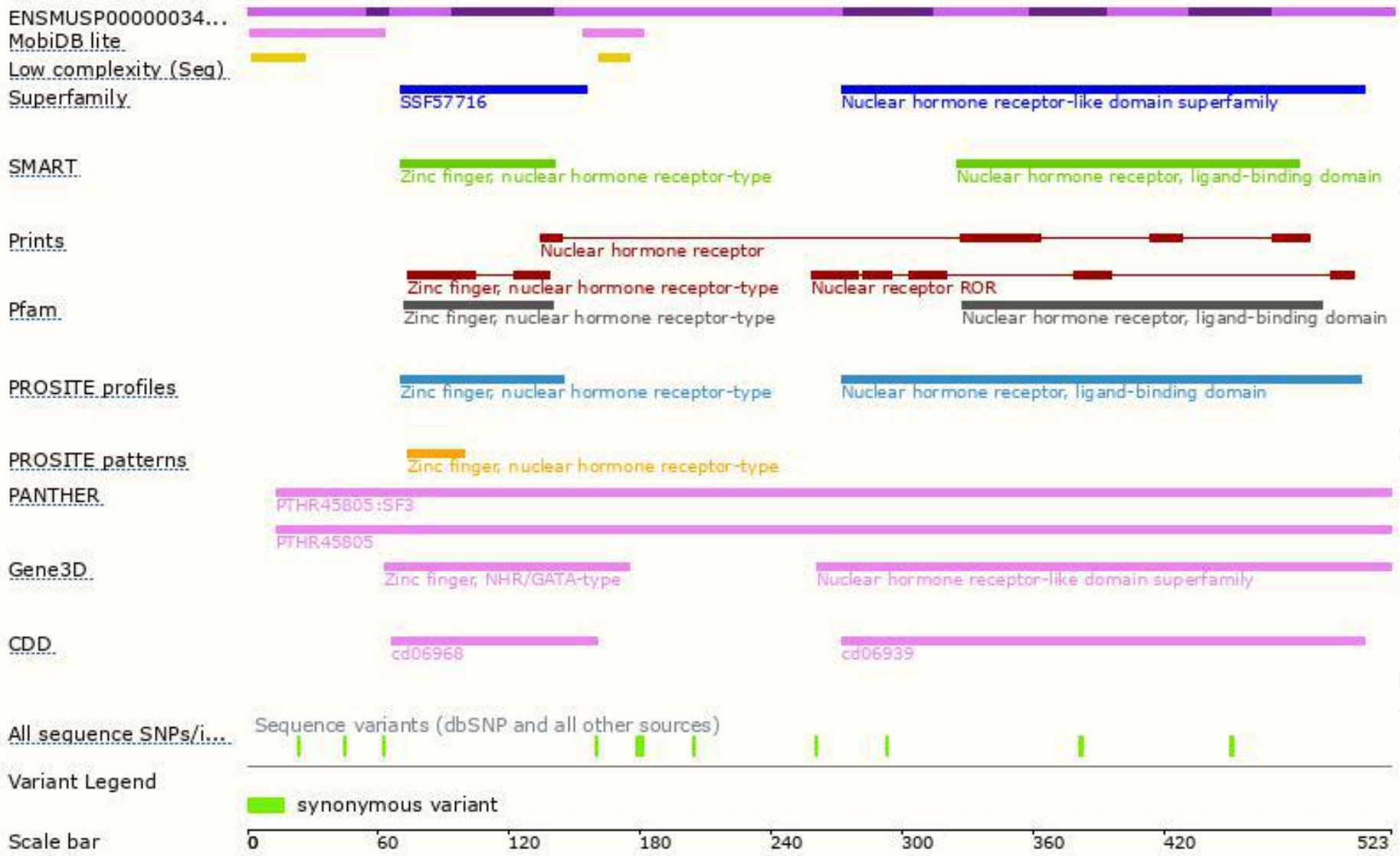
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rora-201	ENSMUST00000034766.13	10878	523aa	Protein coding	CCDS23314	P51448	TSL:1 GENCODE basic
Rora-202	ENSMUST00000113624.2	2591	467aa	Protein coding	CCDS72268	P51448 Q3U1P4	TSL:1 GENCODE basic APPRIS P1
Rora-206	ENSMUST00000174296.1	751	62aa	Nonsense mediated decay	-	G3UZ02	CDS 5' incomplete TSL:5
Rora-204	ENSMUST00000140351.7	1422	No protein	lncRNA	-	-	TSL:3
Rora-203	ENSMUST00000132355.1	561	No protein	lncRNA	-	-	TSL:2
Rora-205	ENSMUST00000143507.1	508	No protein	lncRNA	-	-	TSL:5

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

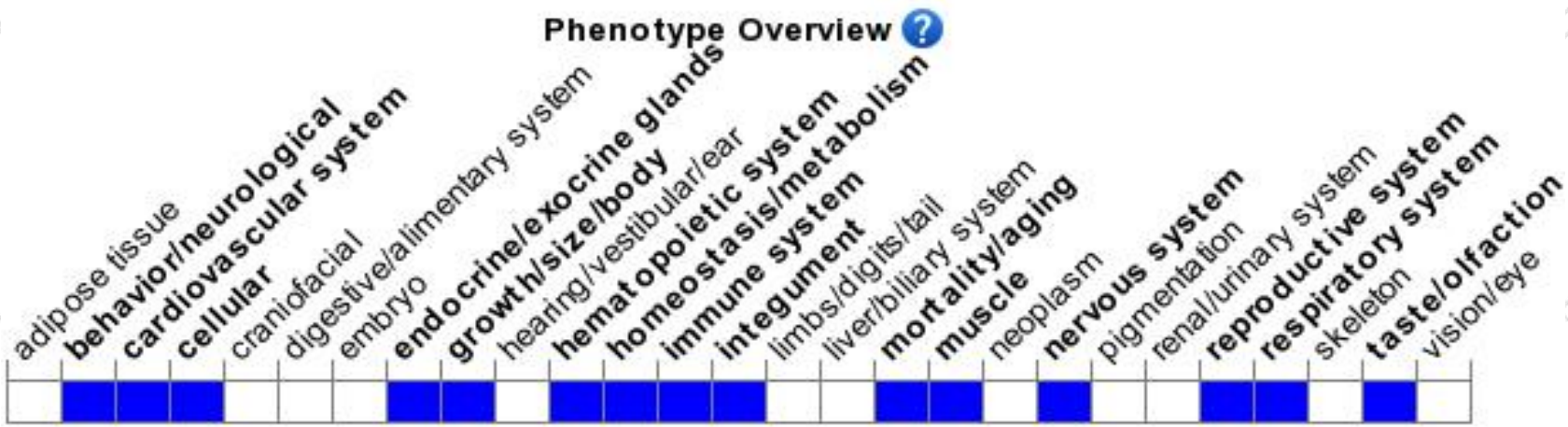
Genomic location distribution



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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