Irx1 Cas9-CKO Strategy

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Project Overview

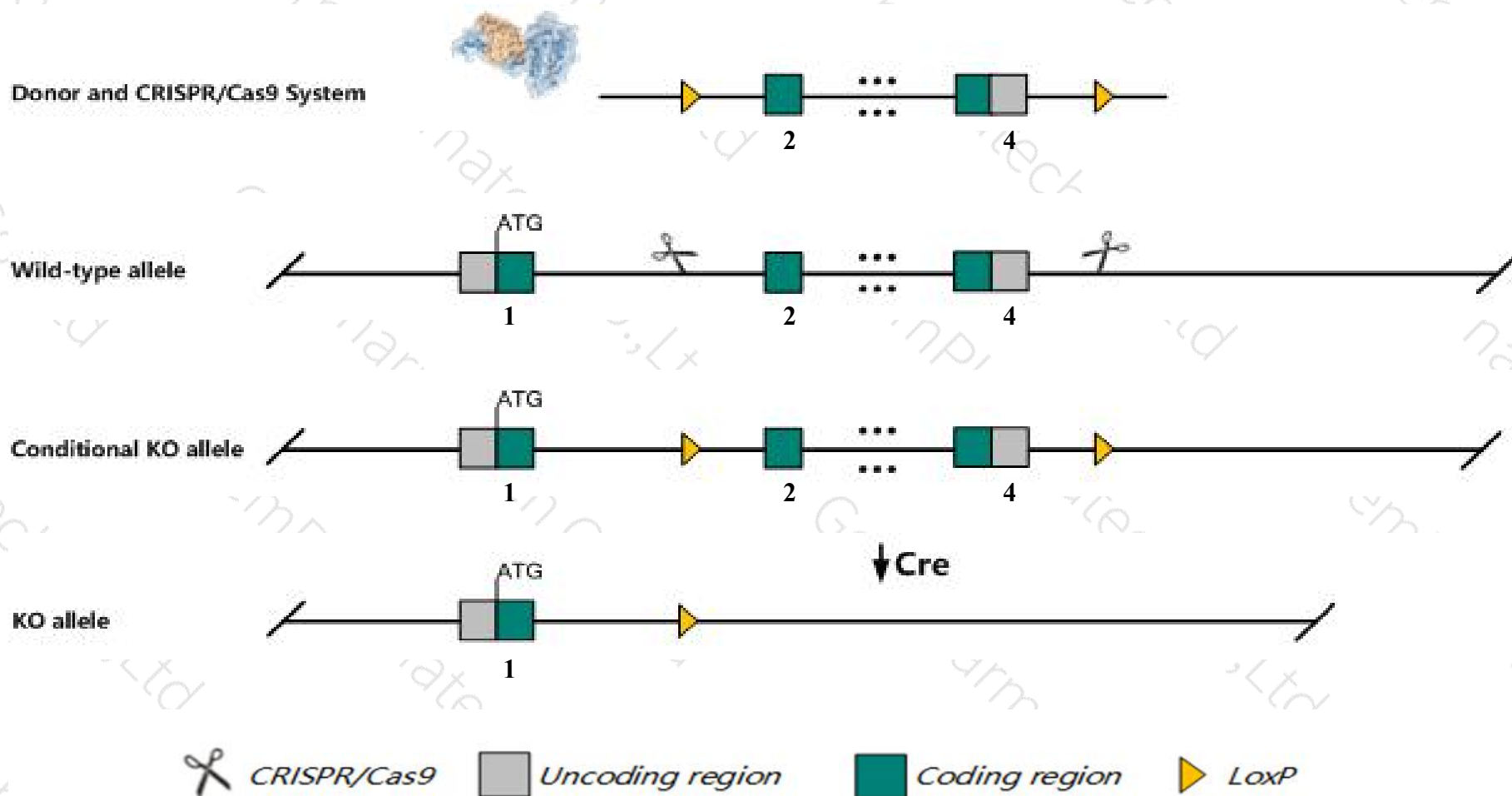|	
Project Name	*Irx1*
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Irx1* gene. The schematic diagram is as follows:

Technical routes

➤ The *Irx1* gene has 3 transcripts. According to the structure of *Irx1* gene, exon2-exon4 of *Irx1-201* (ENSMUST00000077337.8) transcript is recommended as the knockout region.The region contains most of the coding sequence.Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Irx1* 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,Mice homozygous for a knock-out allele exhibit complete neonatal lethality associated with delayed lung maturation, cyanosis, respiratory distress, atelectasis and decreased surfactant secretion, and show defects in dental epithelial cell differentiation and delayed growth of the lower incisors.

➢ The *Irx1* gene is located on the Chr13. 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）

Irx1 Iroquois homeobox 1 [Mus musculus (house mouse)]

Gene ID: 16371, updated on 31-Jan-2019

Summary

|	
Official Symbol	Irx1 provided by MGI
Official Full Name	Iroquois homeobox 1 provided by MGI
Primary source	MGI:MGI:1197515
See related	Ensembl:ENSMUSG00000060969
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
Expression	Biased expression in lung adult (RPKM 102.7), mammary gland adult (RPKM 39.1) and 8 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Irx1-201	ENSMUST00000077337.8	2373	480aa	Protein coding	CCDS26627	P81068	TSL:1 GENCODE basic APPRIS P1
Irx1-203	ENSMUST0000223460.1	820	273aa	Protein coding	-	A0A1Y7VIS3	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3
Irx1-202	ENSMUST00000223379.1	498	86aa	Protein coding	-	A0A1Y7VJ15	CDS 5' incomplete TSL:2

The strategy is based on the design of *Irx1-201* transcript,The transcription is shown below

< Irx1-201
protein coding

◄──Reverse strand

5.80 kb
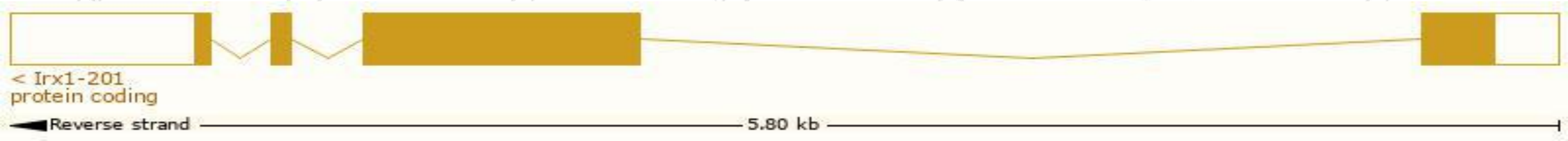
Genomic location distribution
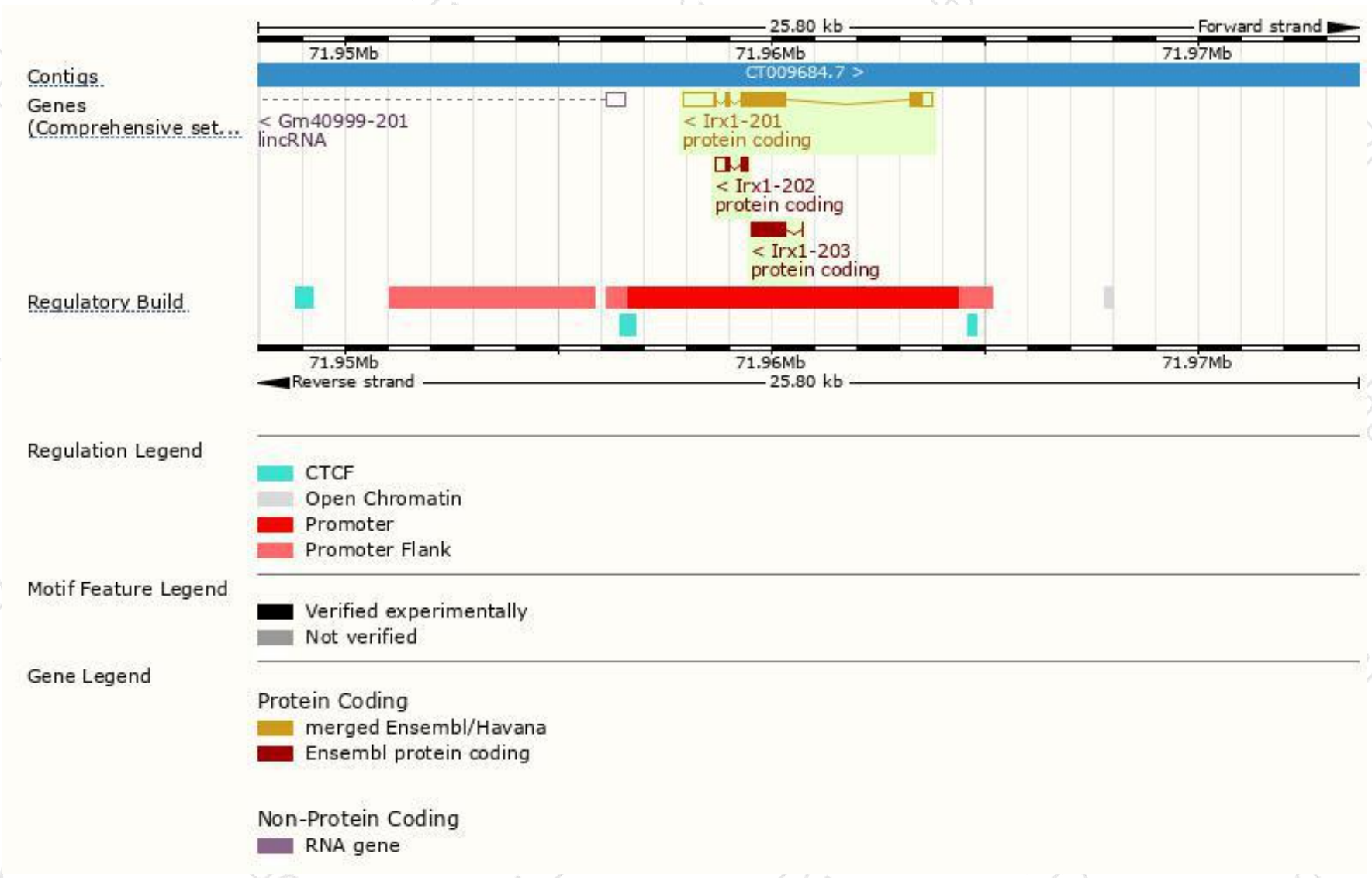
Protein domain

ENSMUSP0000076...
MobiDB lite
Low complexity (Seg)
Conserved Domains
hmmpanther
PTHR11211:SF13
PTHR11211

Superfamily domains
Homeobox-like domain superfamily
SMART domains
Homeobox domain   Iroquois-class homeodomain protein

Pfam domain
Homeobox KN domain
PROSITE profiles
Homeobox domain
PROSITE patterns
Homeobox, conserved site
Gene3D
1.10.10.60

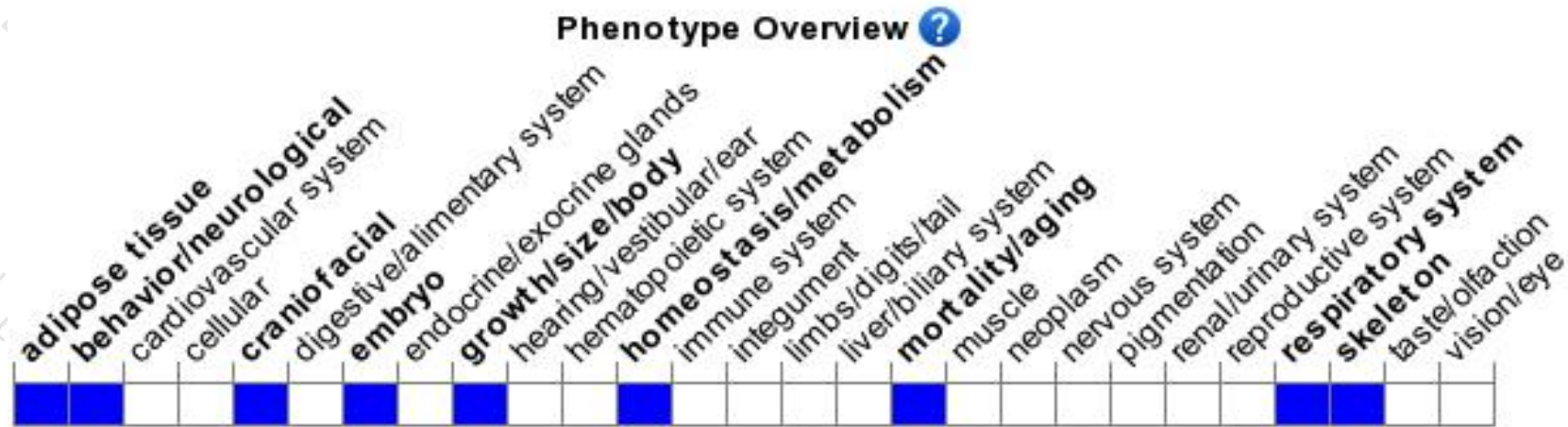All sequence SNPs/i...   Sequence variants (dbSNP and all other sources)

Variant Legend
■ missense variant
■ synonymous variant

Scale bar   0   60   120   180   240   300   360   420   480

Mouse phenotype description(MGI)

Phenotype Overview

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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