

Pax9 Cas9-CKO Strategy

Designer:Xiaojing Li

Reviewer:JiaYu

Design Date:2020-3-12

Project Overview



Project Name

Pax9

Project type

Cas9-CKO

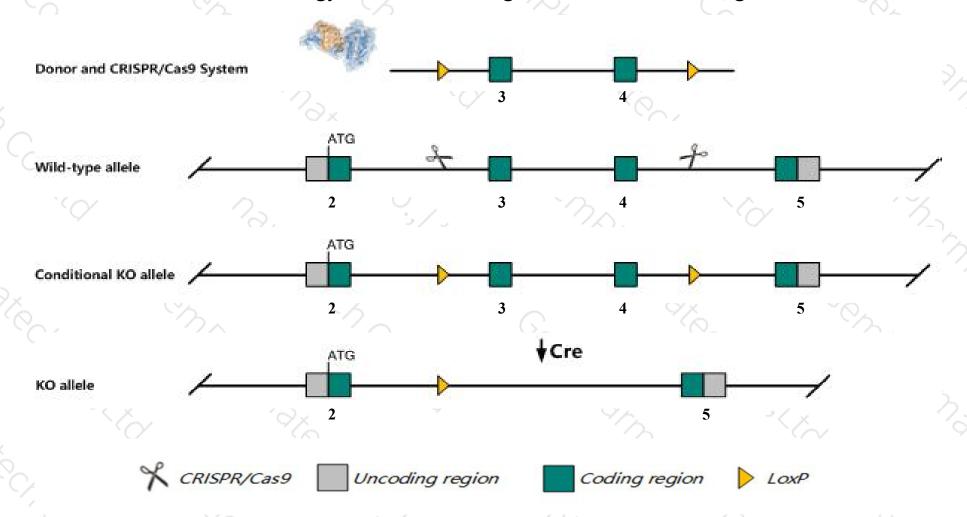
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Pax9* gene. The schematic diagram is as follows:



Technical routes



- The *Pax9* gene has 3 transcripts. According to the structure of *Pax9* gene, exon3-exon4 of *Pax9-203*(ENSMUST00000153250.8) transcript is recommended as the knockout region. The region contains 770bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Pax9* 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, Homozygous inactivation of this gene results in abnormal tooth development and absent teeth and may lead to lack of pharyngeal pouch derivatives, athymia, craniofacial and limb anomalies, a small tympanic ring, various defects of the skeleton and musculature, and neonatal lethality.
- > Insertion of 5-terminal loxP destroys noncoding transcripts Pax9-202.
- > The knockout region is about 3KB from the 5-terminal of Gm15524 gene, which may affect its 5-terminal regulatory function.
- The *Pax9* gene is located on the Chr12. 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)



Pax9 paired box 9 [Mus musculus (house mouse)]

Gene ID: 18511, updated on 10-Mar-2020

Summary

^

Official Symbol Pax9 provided by MGI

Official Full Name paired box 9 provided by MGI

Primary source MGI:MGI:97493

See related Ensembl: ENSMUSG00000001497

Gene type protein coding
RefSeq status REVIEWED
Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;

Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as Pax-9

Summary This gene is a member of the paired box (PAX) family of transcription factors. Members of this gene family typically contain a paired box

domain, an octapeptide, and a paired-type homeodomain. These genes play critical roles during fetal development and cancer growth.

Mice lacking this gene exhibit impaired development of organs, musculature and the skeleton, including absent and abnormally developed teeth, and neonatal lethality. Mutations in the human gene are associated with selective tooth agenesis-3. [provided by

RefSeq, Sep 2015]

Expression Biased expression in thymus adult (RPKM 3.2), lung adult (RPKM 2.8) and 5 other tissues See more

Orthologs human all

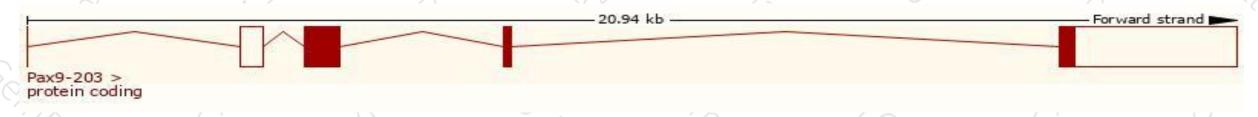
Transcript information (Ensembl)



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

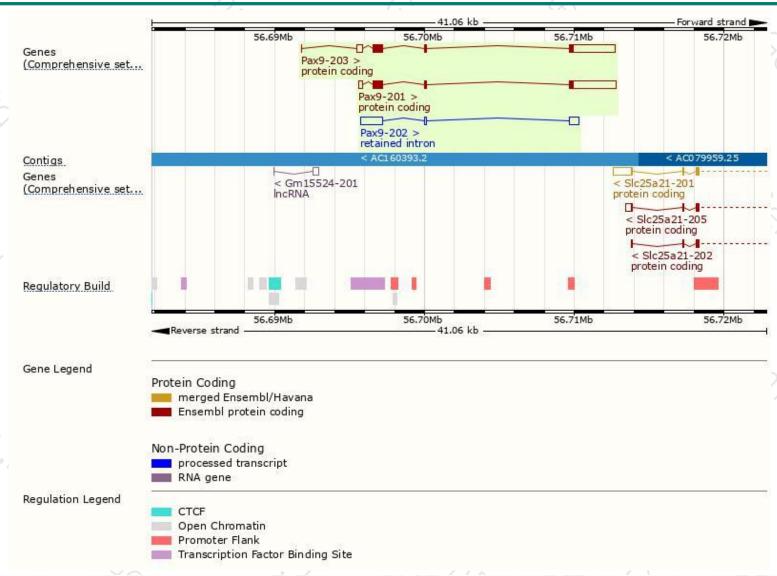
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pax9-203	ENSMUST00000153250.8	4232	342aa	Protein coding	CCDS25924	P47242 Q3V1K1	TSL:5 GENCODE basic APPRIS P1
Pax9-201	ENSMUST00000001538.9	4166	342aa	Protein coding	CCDS25924	P47242 Q3V1K1	TSL:1 GENCODE basic APPRIS P1
Pax9-202	ENSMUST00000152848.1	2309	No protein	Retained intron	-	2	TSL:1

The strategy is based on the design of *Pax9-203* transcript, The transcription is shown below



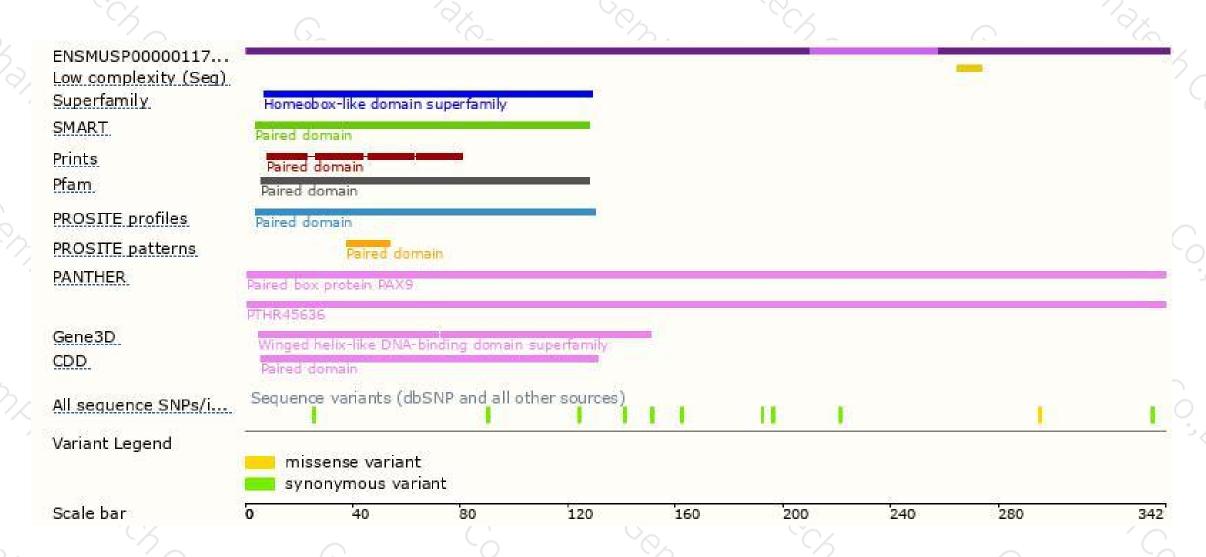
Genomic location distribution





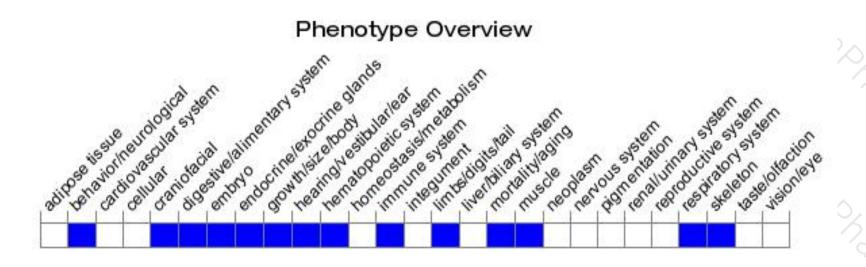
Protein domain





Mouse phenotype description(MGI)





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

According to the existing MGI data, Homozygous inactivation of this gene results in abnormal tooth development and absent teeth and may lead to lack of pharyngeal pouch derivatives, athymia, craniofacial and limb anomalies, a small tympanic ring, various defects of the skeleton and musculature, and neonatal lethality.



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





