

Pax6 Cas9-KO Strategy

Designer:

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Project Overview

Project Name

Pax6

Project type

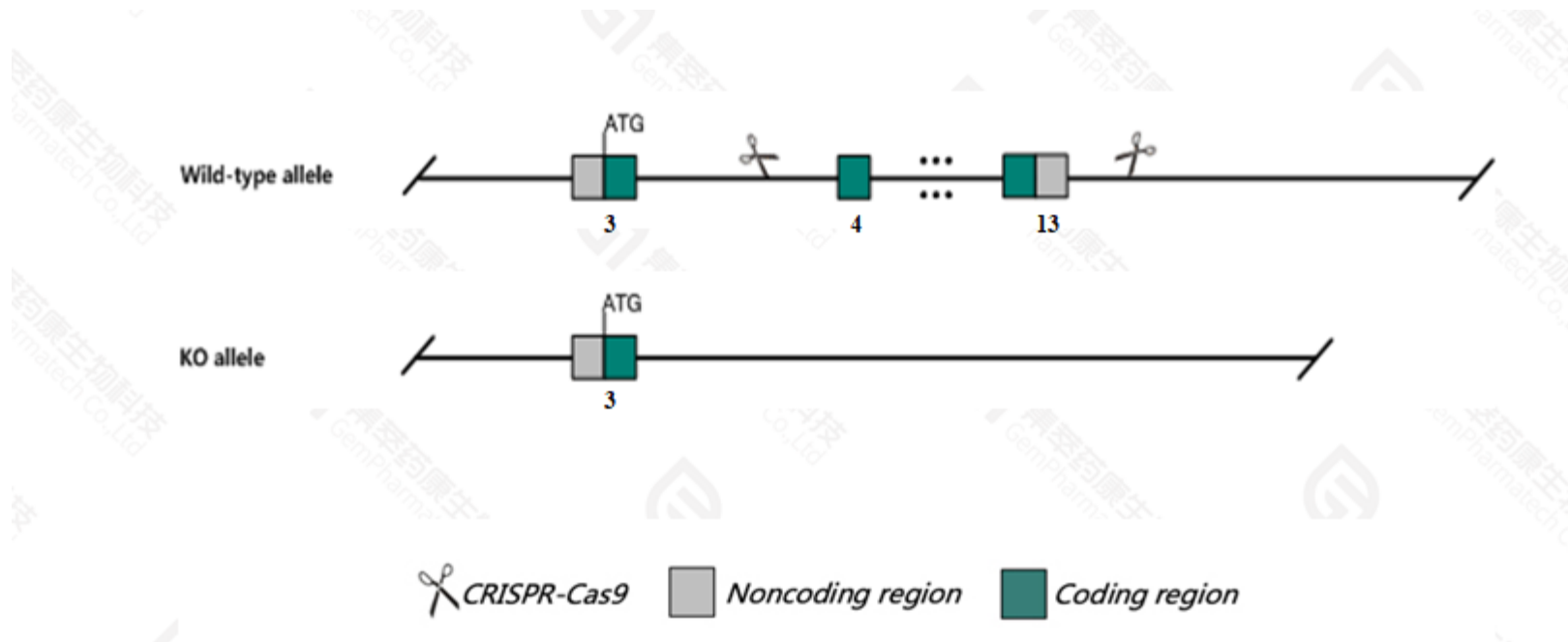
Cas9-KO

Strain background

C57BL/6J

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pax6* gene. The schematic diagram is as follows:



- The *Pax6* gene has 26 transcripts. According to the structure of *Pax6* gene, exon4-exon10 of *Pax6-201* ([ENSMUST00000090391.12](#)) transcript is recommended as the knockout region. The region contains most of coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Pax6* gene. The brief process is as follows: sgRNA was transcribed in vitro. Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

- According to the existing MGI data, Null and hypomorphic mutants show a range of phenotypes from viable with small eyes and lens/cornea fusion to microphthalmia and cataract to embryonic or perinatal lethality with anophthalmia and severe craniofacial and forebrain defects.
- The *Pax6* gene is located on the Chr2. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

Pax6 paired box 6 [Mus musculus (house mouse)]

Gene ID: 18508, updated on 2-Apr-2019

Summary



Official Symbol Pax6 provided by [MGI](#)

Official Full Name paired box 6 provided by [MGI](#)

Primary source [MGI:MGI:97490](#)

See related [Ensembl:ENSMUSG000000027168](#)

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 1500038E17Rik, AEY11, Dey, Gsfaey11, Pax-6, Sey

Summary This gene encodes a homeobox-containing protein that functions as a regulator of transcription. It plays a key role in the development of neural tissues, particularly the eye. Activity of this protein is also required for expression of glucagon in the pancreas. This gene is regulated by multiple enhancers located up to tens or hundreds of kilobases upstream and downstream of the transcription start sites. Mutations in this gene or deletion of these regulatory elements results in severe defects in eye development. Alternative splicing and the use of alternative promoters results in multiple transcript variants, some of which encode proteins that lack the N-terminal paired domain. [provided by RefSeq, Jul 2015]

Expression Biased expression in cerebellum adult (RPKM 9.1), whole brain E14.5 (RPKM 7.7) and 6 other tissues [See more](#)

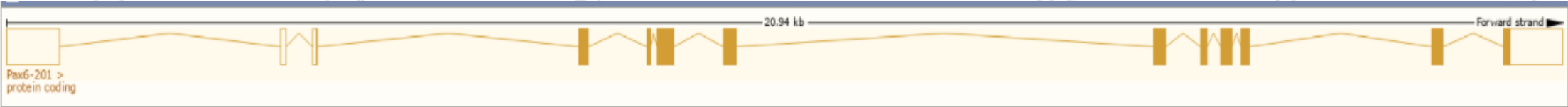
Orthologs [human](#) [all](#)

Transcript information（Ensembl）

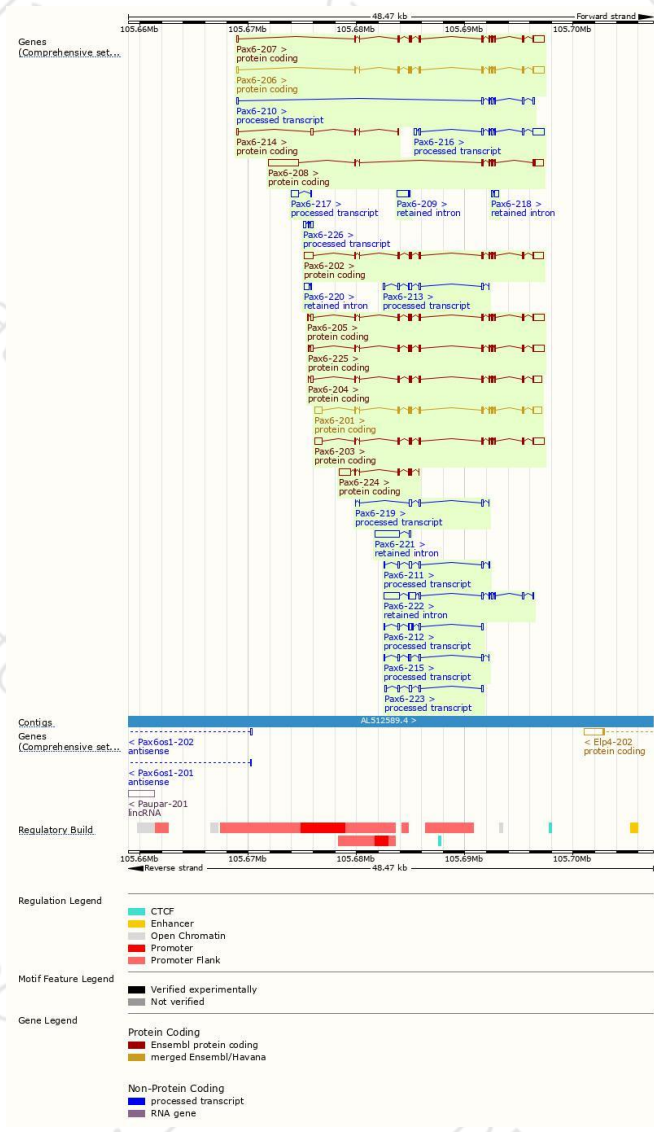
The gene has 26 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pax6-203	ENSMUST00000111082.7	3116	422aa	Protein coding	CCDS57181	P63015	TSL:1 GENCODE basic APPRIS ALT:1
Pax6-202	ENSMUST00000090397.12	3069	422aa	Protein coding	CCDS57181	P63015	TSL:5 GENCODE basic APPRIS ALT:1
Pax6-201	ENSMUST00000090391.12	2669	436aa	Protein coding	CCDS16499	P63015	TSL:1 GENCODE basic APPRIS P:3
Pax6-225	ENSMUST00000167211.8	2814	436aa	Protein coding	CCDS16499	P63015	TSL:5 GENCODE basic APPRIS P:3
Pax6-205	ENSMUST00000111085.7	2779	436aa	Protein coding	CCDS16499	P63015	TSL:5 GENCODE basic APPRIS P:3
Pax6-208	ENSMUST00000111086.10	2601	436aa	Protein coding	CCDS16499	P63015	TSL:1 GENCODE basic APPRIS P:3
Pax6-207	ENSMUST00000111087.9	2559	422aa	Protein coding	CCDS57181	P63015	TSL:1 GENCODE basic APPRIS ALT:1
Pax6-204	ENSMUST00000111083.9	2370	422aa	Protein coding	CCDS57181	P63015	TSL:5 GENCODE basic APPRIS ALT:1
Pax6-208	ENSMUST00000111088.8	4427	254aa	Protein coding	-	A2A412	TSL:1 GENCODE basic
Pax6-224	ENSMUST00000156216.2	1635	139aa	Protein coding	-	A2A409	CDS 3' incomplete TSL:5
Pax6-214	ENSMUST00000142772.7	528	17aa	Protein coding	-	V9GX16	CDS 3' incomplete TSL:3
Pax6-216	ENSMUST00000146803.3	2148	No protein	Processed transcript	-	-	TSL:1
Pax6-210	ENSMUST00000123063.7	983	No protein	Processed transcript	-	-	TSL:1
Pax6-213	ENSMUST00000140173.7	925	No protein	Processed transcript	-	-	TSL:5
Pax6-212	ENSMUST00000138365.7	874	No protein	Processed transcript	-	-	TSL:2
Pax6-211	ENSMUST00000135412.7	870	No protein	Processed transcript	-	-	TSL:3
Pax6-217	ENSMUST00000147923.2	834	No protein	Processed transcript	-	-	TSL:1
Pax6-223	ENSMUST00000155081.7	778	No protein	Processed transcript	-	-	TSL:3
Pax6-215	ENSMUST00000145744.7	743	No protein	Processed transcript	-	-	TSL:3
Pax6-219	ENSMUST00000153011.7	654	No protein	Processed transcript	-	-	TSL:5
Pax6-226	ENSMUST00000172990.7	608	No protein	Processed transcript	-	-	TSL:1
Pax6-222	ENSMUST00000154643.7	2919	No protein	Retained intron	-	-	TSL:5
Pax6-221	ENSMUST00000154302.2	2343	No protein	Retained intron	-	-	TSL:3
Pax6-209	ENSMUST00000123000.2	1096	No protein	Retained intron	-	-	TSL:5
Pax6-220	ENSMUST00000153639.1	538	No protein	Retained intron	-	-	TSL:2
Pax6-218	ENSMUST00000147946.1	527	No protein	Retained intron	-	-	TSL:3

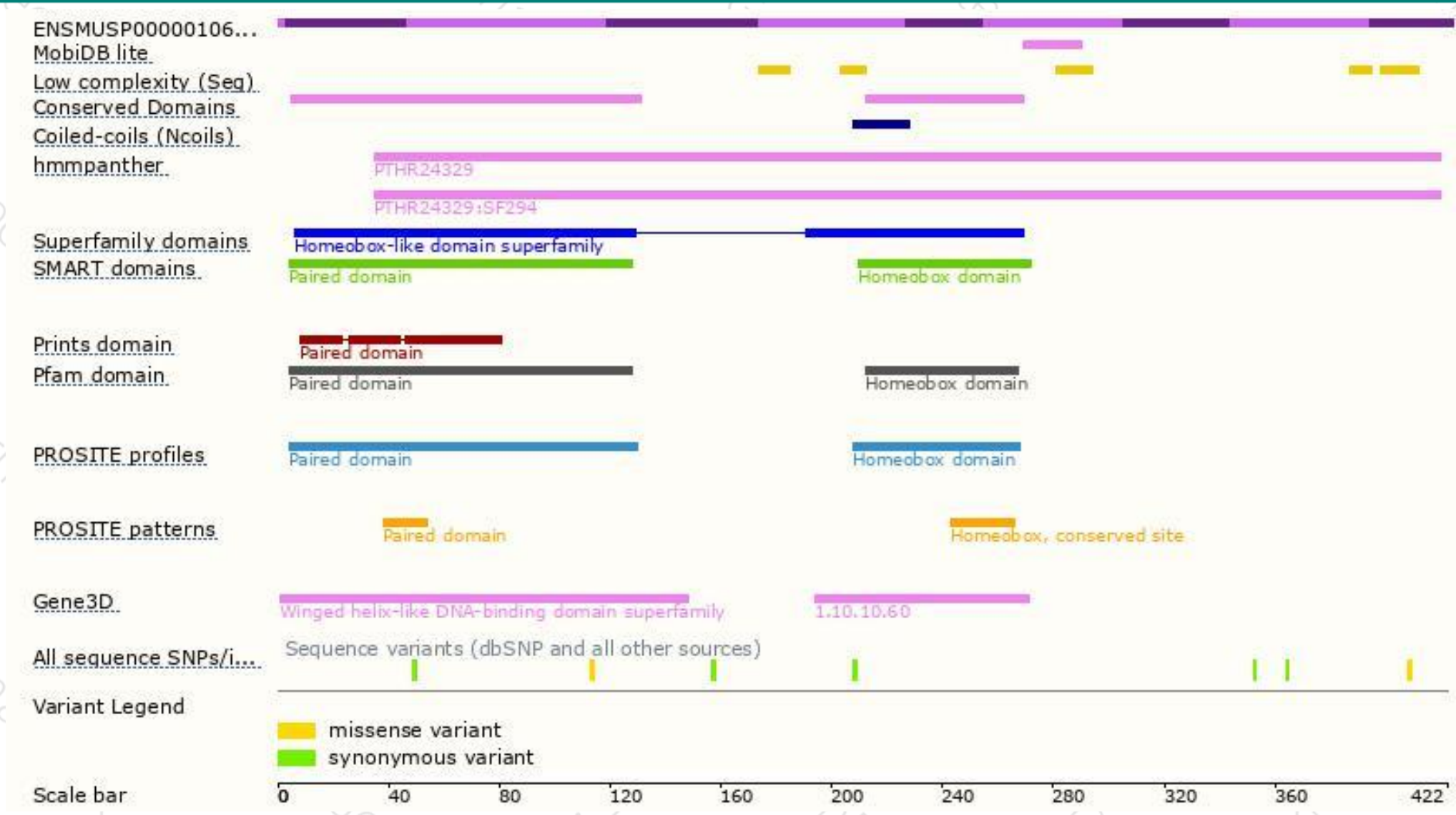
The strategy is based on the design of *Pax6-201* transcript,The transcription is shown below



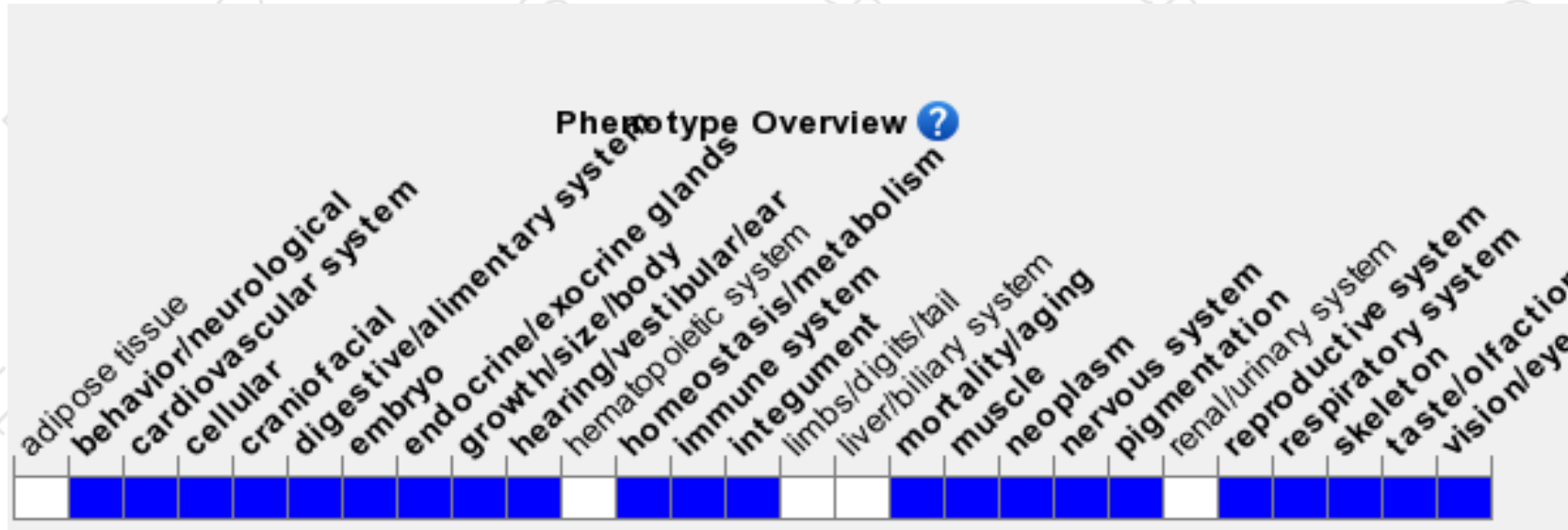
Genomic location distribution



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/>).

According to the existing MGI data, Null and hypomorphic mutants show a range of phenotypes from viable with small eyes and lens/cornea fusion to microphthalmia and cataract to embryonic or perinatal lethality with anophthalmia and severe craniofacial and forebrain defects.

If you have any questions, you are welcome to inquire.

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