Pla2g6 Cas9-CKO Strategy

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Project Overview

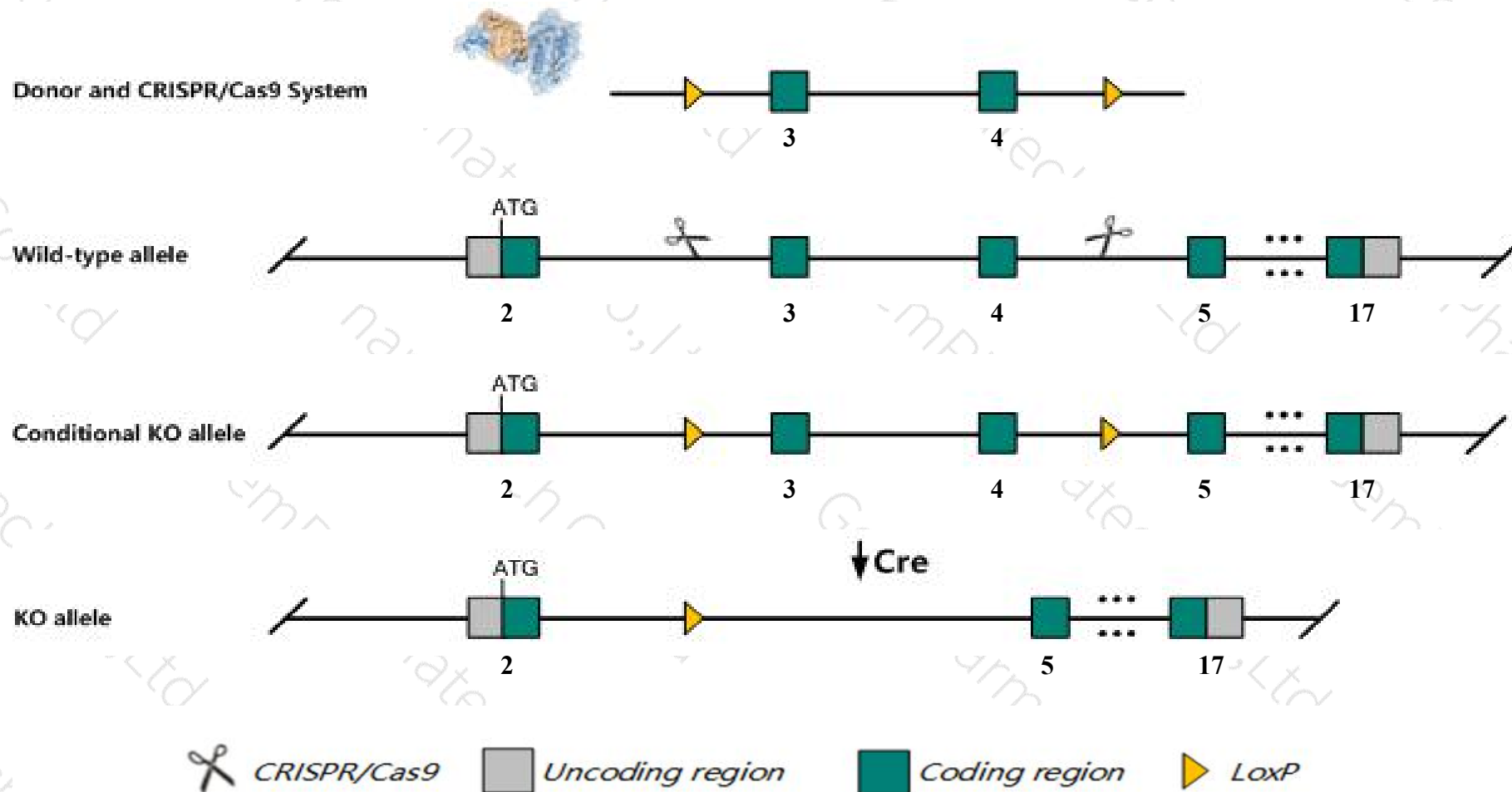|	
Project Name	*Pla2g6*
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pla2g6* gene. The schematic diagram is as follows:

Technical routes

➤ The *Pla2g6* gene has 13 transcripts. According to the structure of *Pla2g6* gene, exon3-exon4 of *Pla2g6-210* (ENSMUST00000174021.7) transcript is recommended as the knockout region.The region contains Ens 400bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Pla2g6* 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 null mice display impaired male fertility and asthenozoospermia.

➢ The *Pla2g6* gene is located on the Chr15. 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）

Pla2g6 phospholipase A2, group VI [Mus musculus (house mouse)]
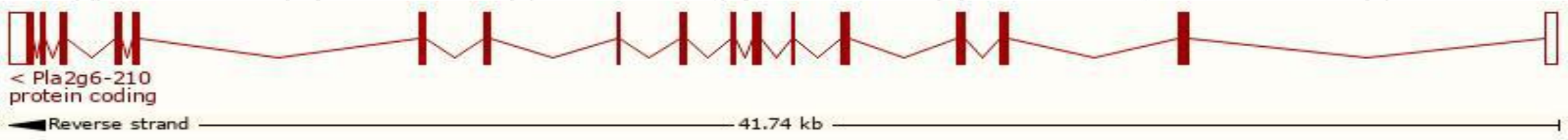
Gene ID: 53357, updated on 13-Mar-2020

Summary

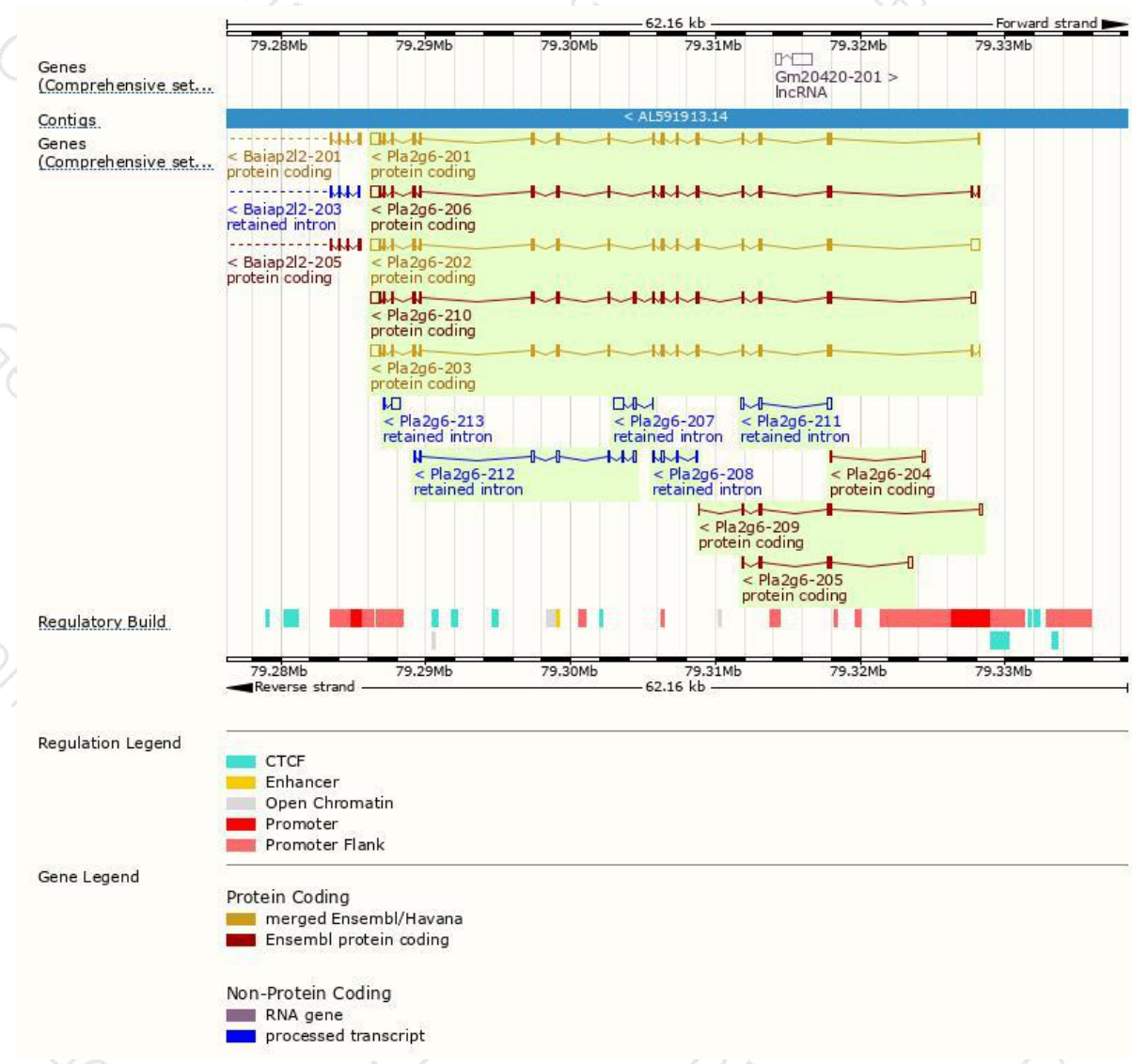|	
Official Symbol	Pla2g6 provided by MGI
Official Full Name	phospholipase A2, group VI provided by MGI
Primary source	MGI:MGI:1859152
See related	Ensembl:ENSMUSG00000042632
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	BB112799, PNPLA9, iPLA(2)beta, iPLA2, iPLA2beta
Expression	Broad expression in testis adult (RPKM 76.7), adrenal adult (RPKM 14.0) and 23 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

The gene has 13 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pla2g6-202	ENSMUST00000166977.8	3365	752aa	Protein coding	CCDS27637	P97819 Q3UN31	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Pla2g6-210	ENSMUST00000174021.7	3318	807aa	Protein coding	CCDS56991	P97819	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT1
Pla2g6-206	ENSMUST00000173163.7	3018	752aa	Protein coding	CCDS27637	P97819 Q3UN31	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Pla2g6-201	ENSMUST00000047816.14	2963	752aa	Protein coding	CCDS27637	P97819 Q3UN31	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Pla2g6-203	ENSMUST00000172403.8	2943	752aa	Protein coding	CCDS27637	P97819 Q3UN31	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Pla2g6-205	ENSMUST00000173109.1	930	199aa	Protein coding	-	G3UX52	CDS 3' incomplete TSL:3
Pla2g6-209	ENSMUST00000173632.7	834	204aa	Protein coding	-	G3UY97	CDS 3' incomplete TSL:5
Pla2g6-204	ENSMUST00000172936.1	377	33aa	Protein coding	-	G3UX73	CDS 3' incomplete TSL:3
Pla2g6-212	ENSMUST00000174375.1	863	No protein	Retained intron	-	-	TSL:3
Pla2g6-207	ENSMUST00000173412.1	772	No protein	Retained intron	-	-	TSL:5
Pla2g6-211	ENSMUST00000174089.1	708	No protein	Retained intron	-	-	TSL:2
Pla2g6-213	ENSMUST00000229011.1	696	No protein	Retained intron	-	-	
Pla2g6-208	ENSMUST00000173601.1	550	No protein	Retained intron	-	-	TSL:3

The strategy is based on the design of *Pla2g6-210* transcript,the transcription is shown below

< Pla2g6-210
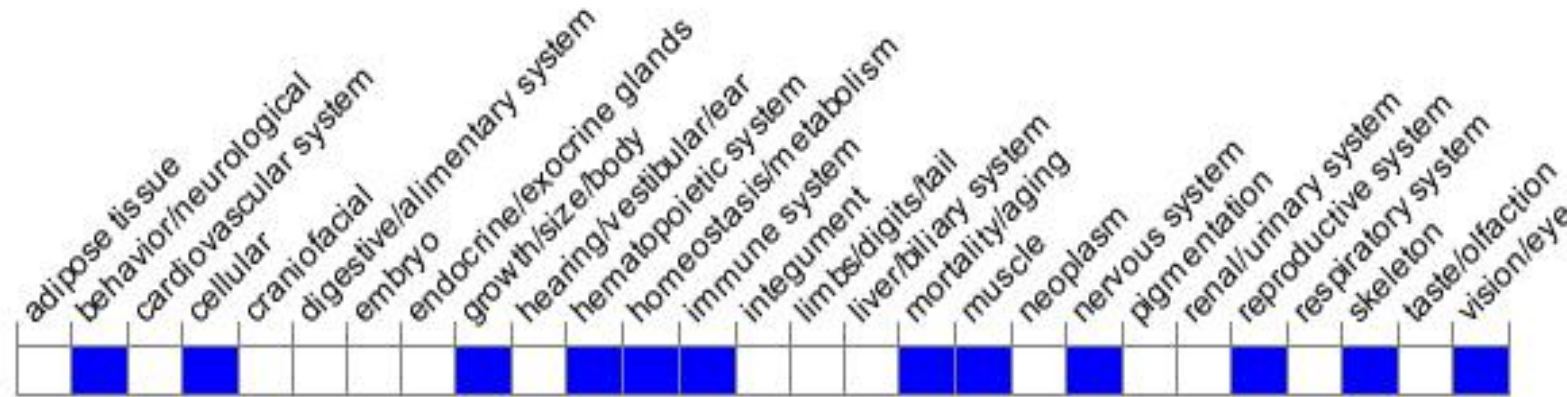protein coding

Reverse strand

41.74 kb

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 null mice display impaired male fertility and asthenozoospermia.

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