

***Phactr2* Cas9-CKO Strategy**

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Project Overview

Project Name

Phactr2

Project type

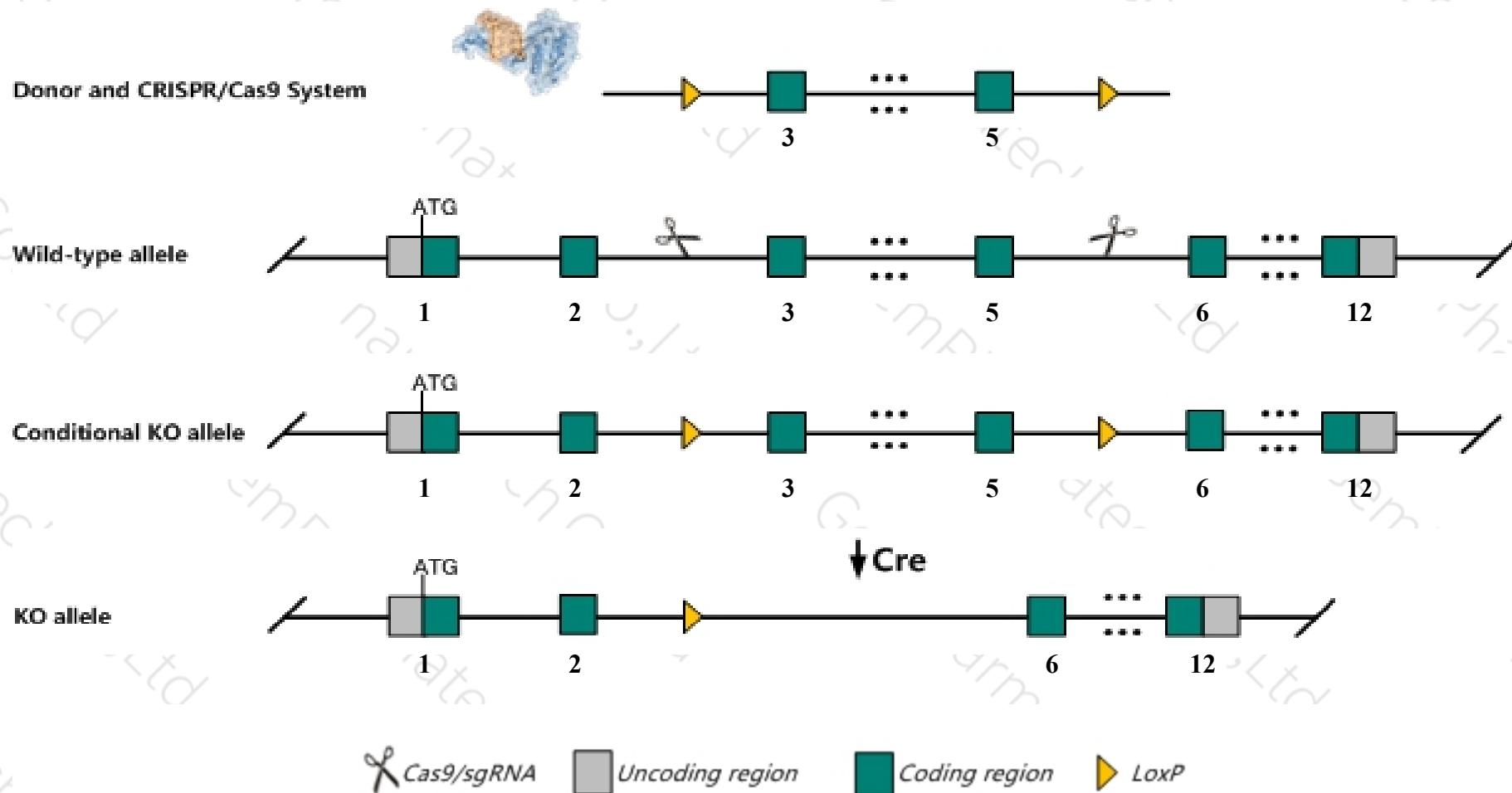
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Phactr2* gene. The schematic diagram is as follows:



- The *Phactr2* gene has 5 transcripts. According to the structure of *Phactr2* gene, exon3-exon5 of *Phactr2*-202(ENSMUST00000105543.8) transcript is recommended as the knockout region. The region contains 778bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Phactr2* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 was 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.

- The *Phactr2* gene is located on the Chr10. 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)

Phactr2 phosphatase and actin regulator 2 [Mus musculus (house mouse)]

Gene ID: 215789, updated on 13-Mar-2020

Summary



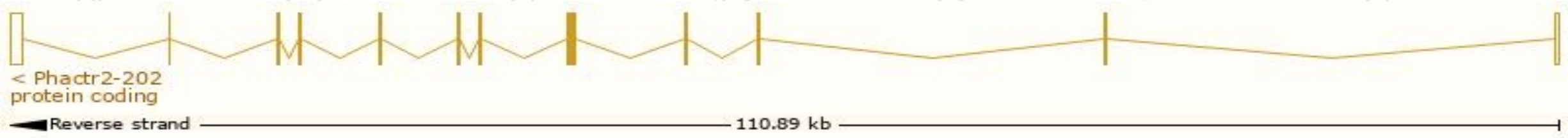
Official Symbol	Phactr2 provided by MGI
Official Full Name	phosphatase and actin regulator 2 provided by MGI
Primary source	MGI:MGI:2446138
See related	Ensembl:ENSMUSG00000062866
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AV158170, BC012871
Expression	Ubiquitous expression in lung adult (RPKM 5.6), limb E14.5 (RPKM 4.9) and 27 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

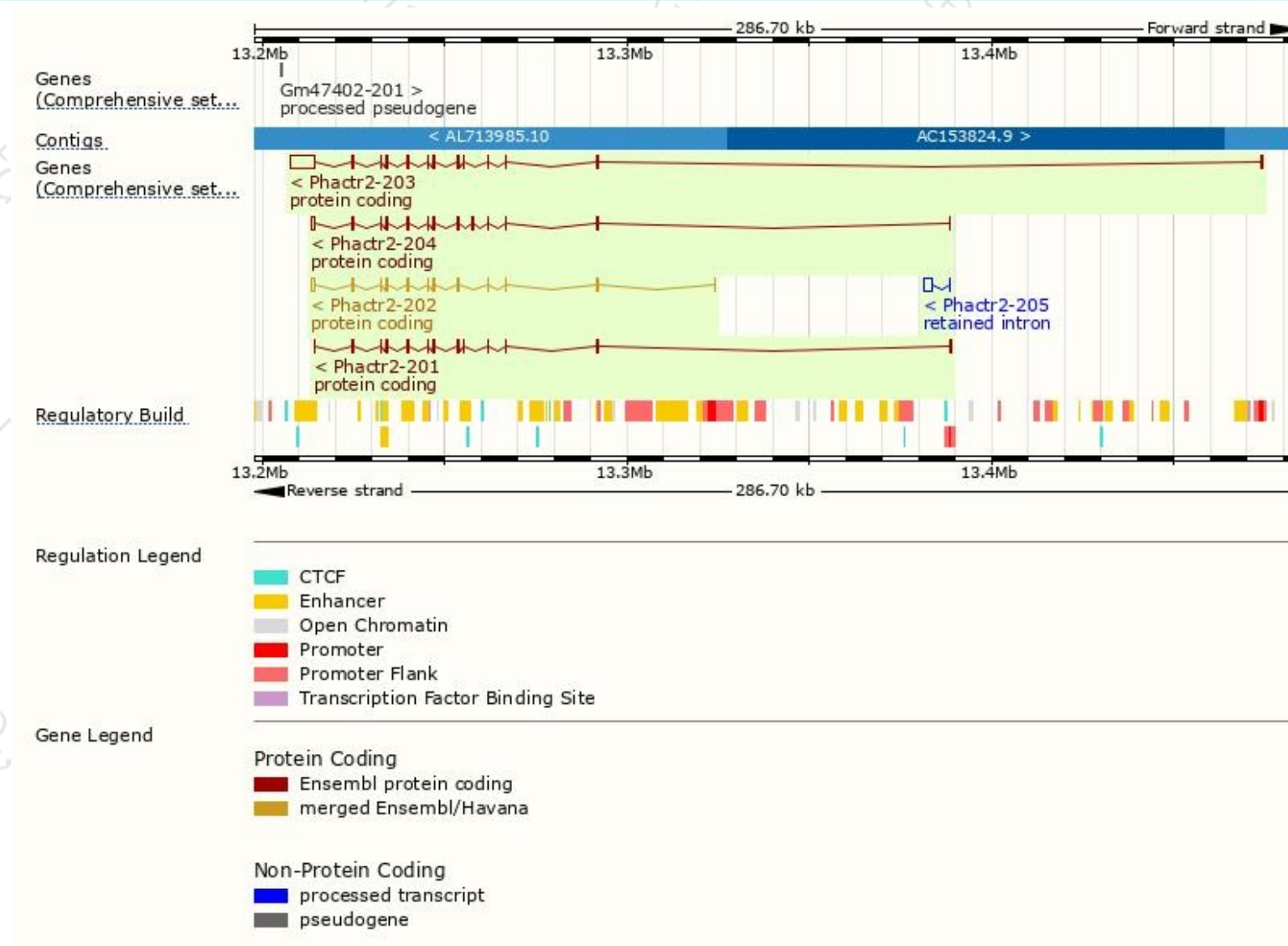
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Phactr2-202	ENSMUST00000105543.8	2735	563aa	Protein coding	CCDS35846	Q3UQ19	TSL:1 GENCODE basic APPRIS P2
Phactr2-203	ENSMUST00000105545.11	8556	626aa	Protein coding	-	F7D4H5	TSL:1 GENCODE basic APPRIS ALT2
Phactr2-204	ENSMUST00000105546.7	2808	632aa	Protein coding	-	B1AVP0	TSL:5 GENCODE basic APPRIS ALT2
Phactr2-201	ENSMUST00000079698.6	1934	556aa	Protein coding	-	B1AVN9	TSL:5 GENCODE basic APPRIS ALT2
Phactr2-205	ENSMUST00000154265.1	2431	No protein	Retained intron	-	-	TSL:1

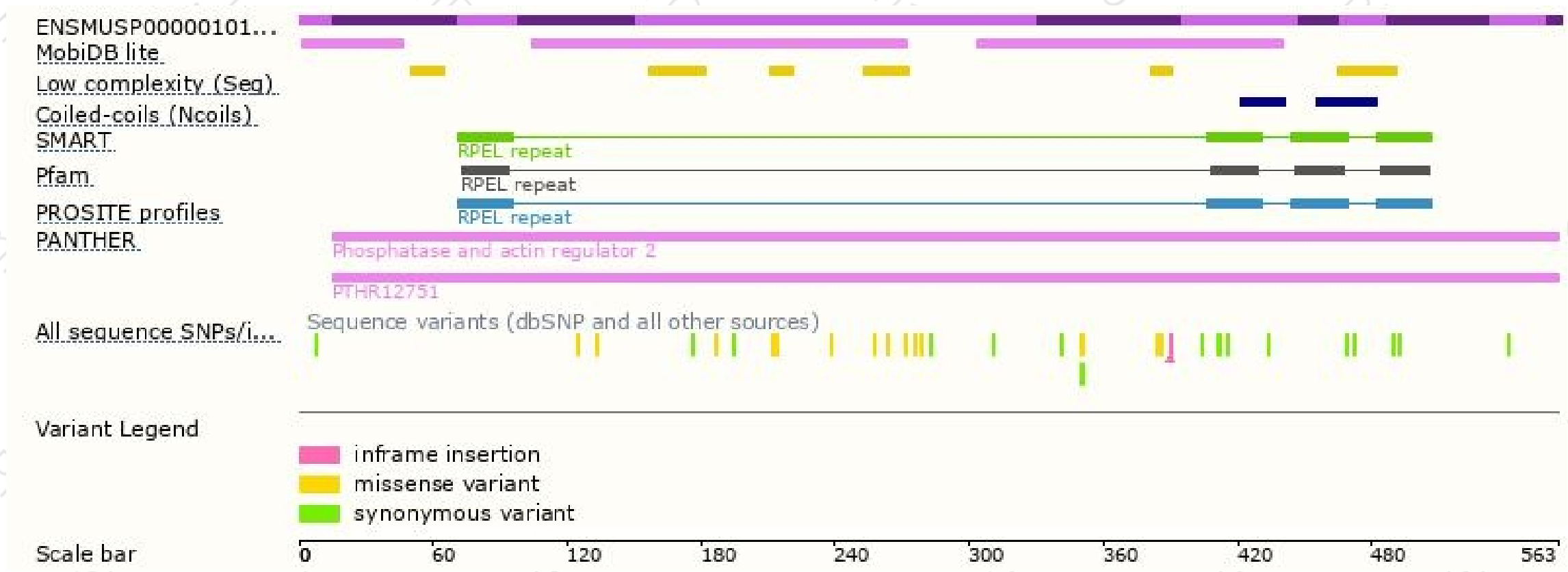
The strategy is based on the design of *Phactr2-202* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

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