G6pc3 Cas9-CKO Strategy

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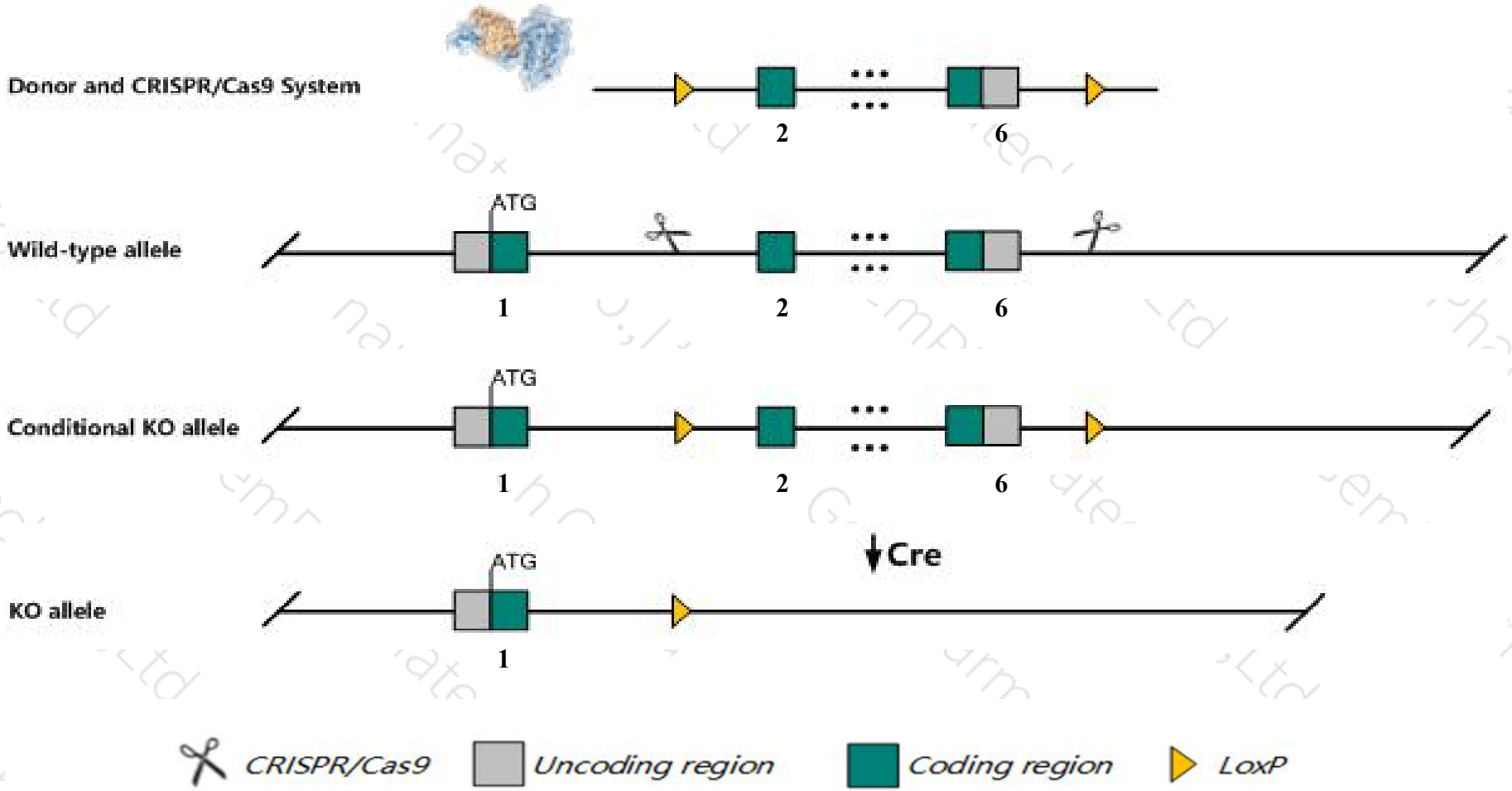
Project Overview

Project Name	***G6pc3***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *G6pc3* gene. The schematic diagram is as follows:



Technical routes

➢ The *G6pc3* gene has 7 transcripts. According to the structure of *G6pc3* gene, exon2-exon6 of *G6pc3-202*(ENSMUST00000078975.7) transcript is recommended as the knockout region.The region contains 823bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *G6pc3* 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 mutation of this gene results in females, but not males, that are growth retarded and weight less and exhibit elevated plasma glucagon levels but lowered plasma cholesterols after fasting.

➢The flox region is about 300bp away from the 3th end of the Hdac5 gene, which may affect the regulation of this gene.

➢ The *G6pc3* gene is located on the Chr11. 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）

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G6pc3 glucose 6 phosphatase, catalytic, 3 [Mus musculus (house mouse)]

Gene ID: 68401, updated on 13-Mar-2020

Summary

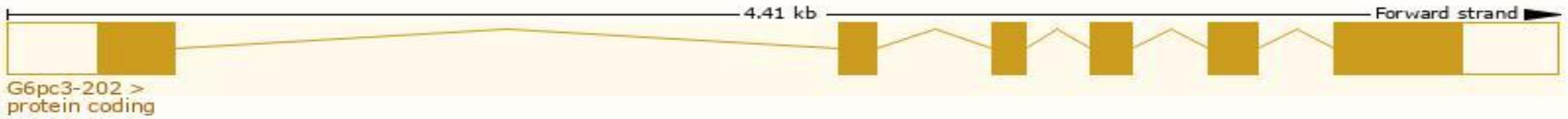
Official Symbol	G6pc3 provided by MGI
Official Full Name	glucose 6 phosphatase, catalytic, 3 provided byMGI
Primary source	MGI:MGI:1915651
See related	Ensembl:ENSMUSG00000034793
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	0710001K01Rik, AU019276, AU045429, AV128920, AW545836, UGRP
Expression	Ubiquitous expression in testis adult (RPKM 154.8), adrenal adult (RPKM 105.5) and 25 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

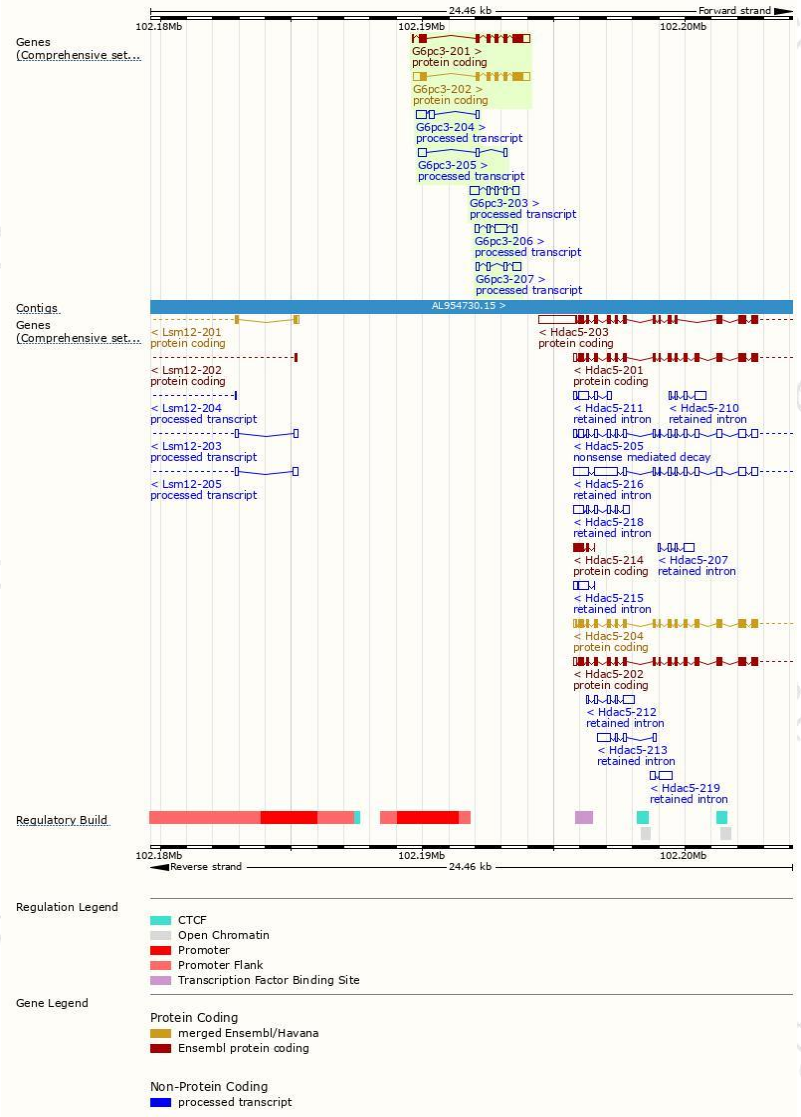
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
G6pc3-202	ENSMUST00000078975.7	1573	346aa	Protein coding	CCDS25491	Q6NSQ9	TSL:1 GENCODE basic APPRIS P1
G6pc3-201	ENSMUST00000070334.9	1404	346aa	Protein coding	CCDS25491	Q6NSQ9	TSL:1 GENCODE basic APPRIS P1
G6pc3-203	ENSMUST00000124594.7	895	No protein	Processed transcript	-	-	TSL:2
G6pc3-206	ENSMUST00000145490.7	848	No protein	Processed transcript	-	-	TSL:5
G6pc3-204	ENSMUST00000131382.7	673	No protein	Processed transcript	-	-	TSL:3
G6pc3-207	ENSMUST00000153178.1	659	No protein	Processed transcript	-	-	TSL:5
G6pc3-205	ENSMUST00000139995.1	541	No protein	Processed transcript	-	-	TSL:5

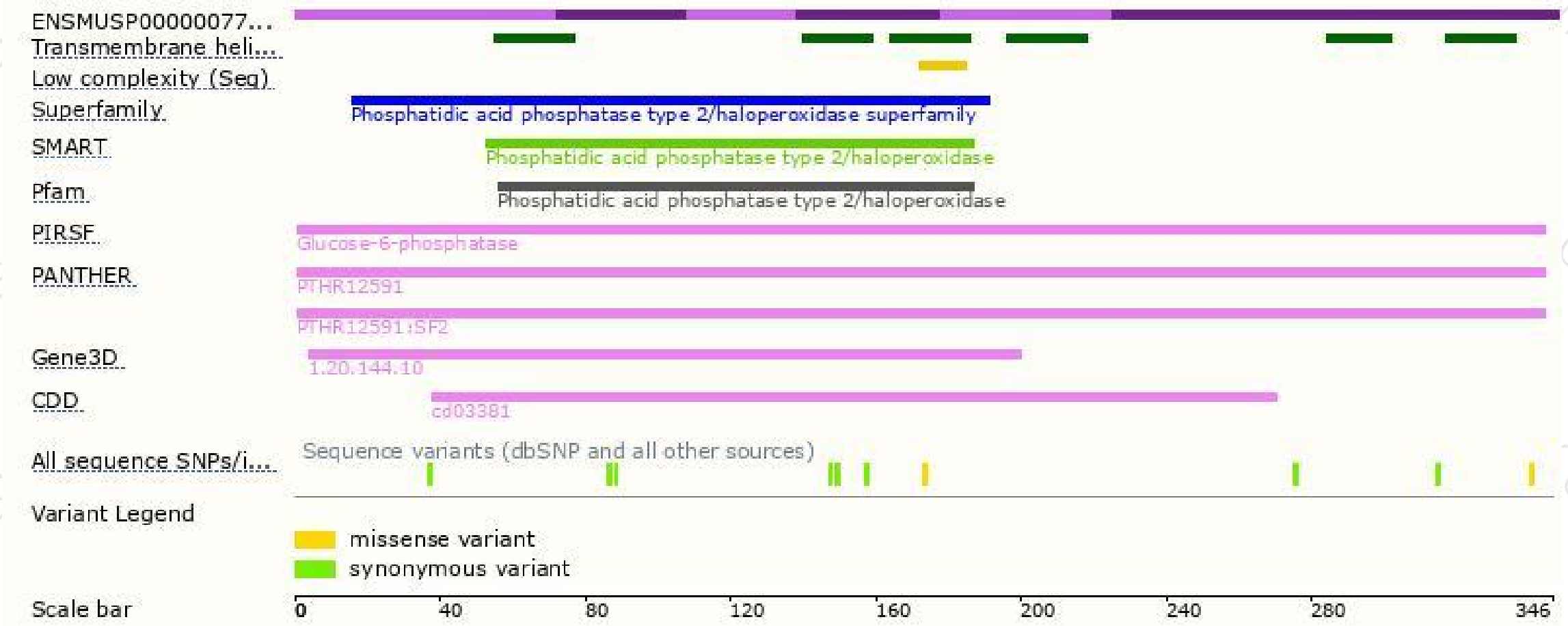
The strategy is based on the design of *G6pc3-202* transcript,the transcription is shown below:



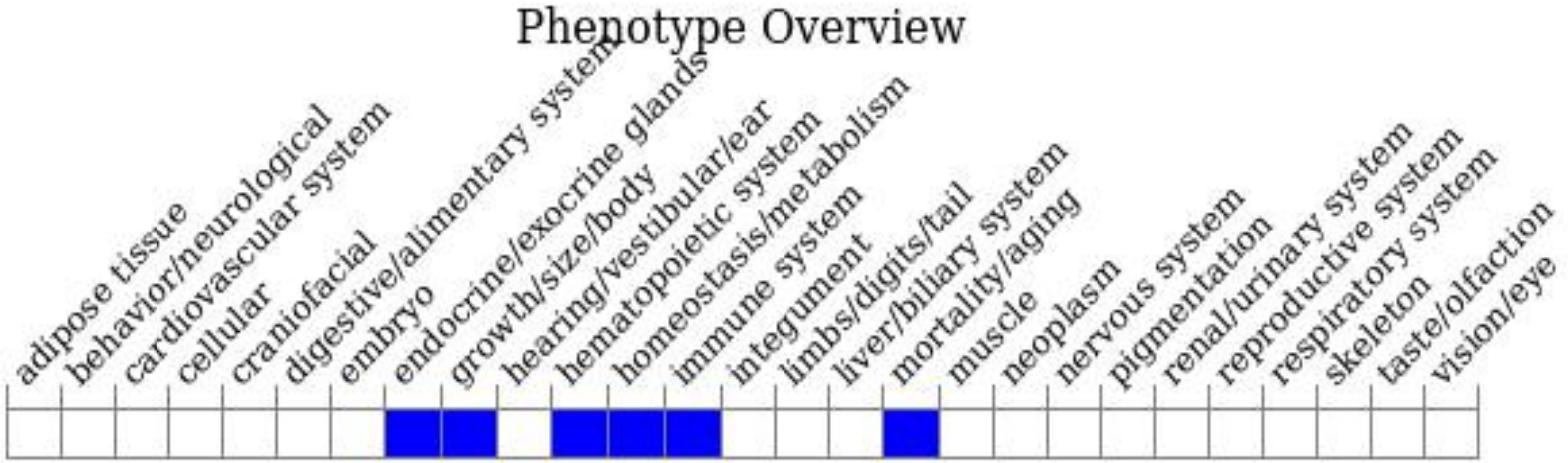
Genomic location distribution



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/).

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

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