

P3h1 Cas9-CKO Strategy

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Project Overview

Project Name

P3h1

Project type

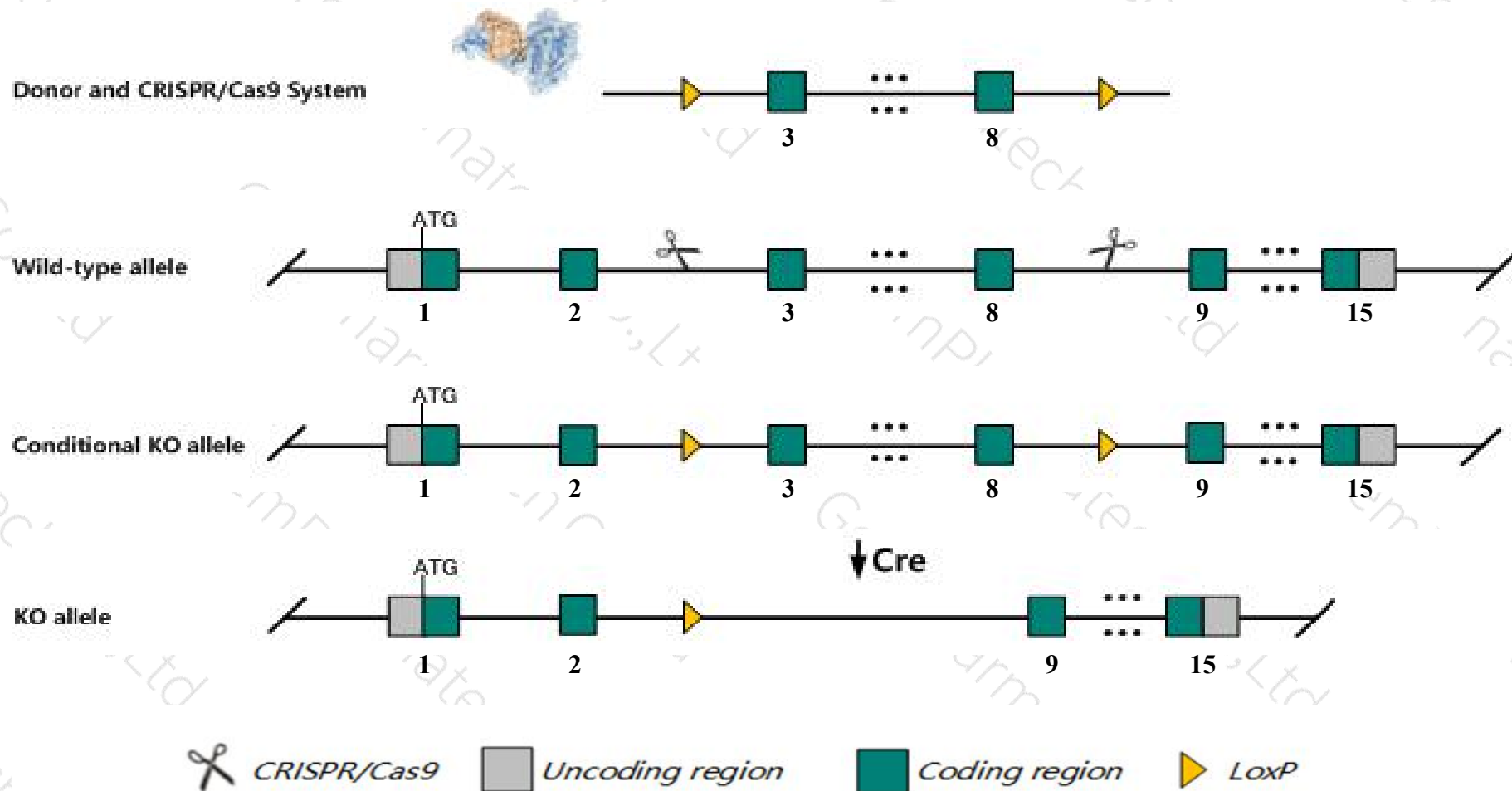
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *P3hl* gene. The schematic diagram is as follows:



- The *P3h1* gene has 12 transcripts. According to the structure of *P3h1* gene, exon3-exon8 of *P3h1-201* (ENSMUST00000030393.12) transcript is recommended as the knockout region. The region contains 748bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *P3h1* 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.

- According to the existing MGI data, Mice homozygous for a knock-out allele exhibit reduced size, disproportional reduction in long bone length, decreased bone density, decreased bone mineral density, reduced body fat, delayed ossification, and abnormal collagen networks in the skin and tendons.
- Transcript *P3h1*-207&209&211 may not be affected.
- The floxed region is near to the N-terminal of *AU022252* gene, this strategy may influence the regulatory function of the N-terminal of *AU022252* gene.
- The *P3h1* gene is located on the Chr4. 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)

P3h1 prolyl 3-hydroxylase 1 [*Mus musculus* (house mouse)]

Gene ID: 56401, updated on 12-Aug-2019

Summary

- Official Symbol

P3h1 provided by [MGI](#)
- Official Full Name

prolyl 3-hydroxylase 1 provided by [MGI](#)
- Primary source

[MGI:MGI:1888921](#)
- See related

[Ensembl:ENSMUSG00000028641](#)
- 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

Gros1; Lepre1; 2410024C15Rik
- Expression

Broad expression in limb E14.5 (RPKM 50.3), ovary adult (RPKM 44.1) and 24 other tissues [See more](#)
- Orthologs

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

Genomic context

Location: 4; 4 D2.1

See P3h1 in [Genome Data Viewer](#)

Exon count: 17

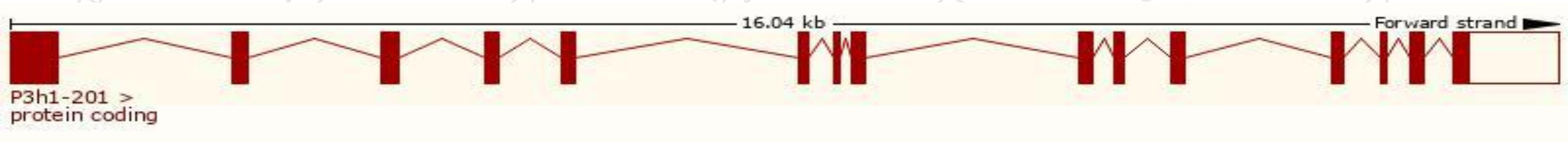
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	4	NC_000070.6 (119232915..119248977)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	4	NC_000070.5 (118905520..118921582)

Transcript information (Ensembl)

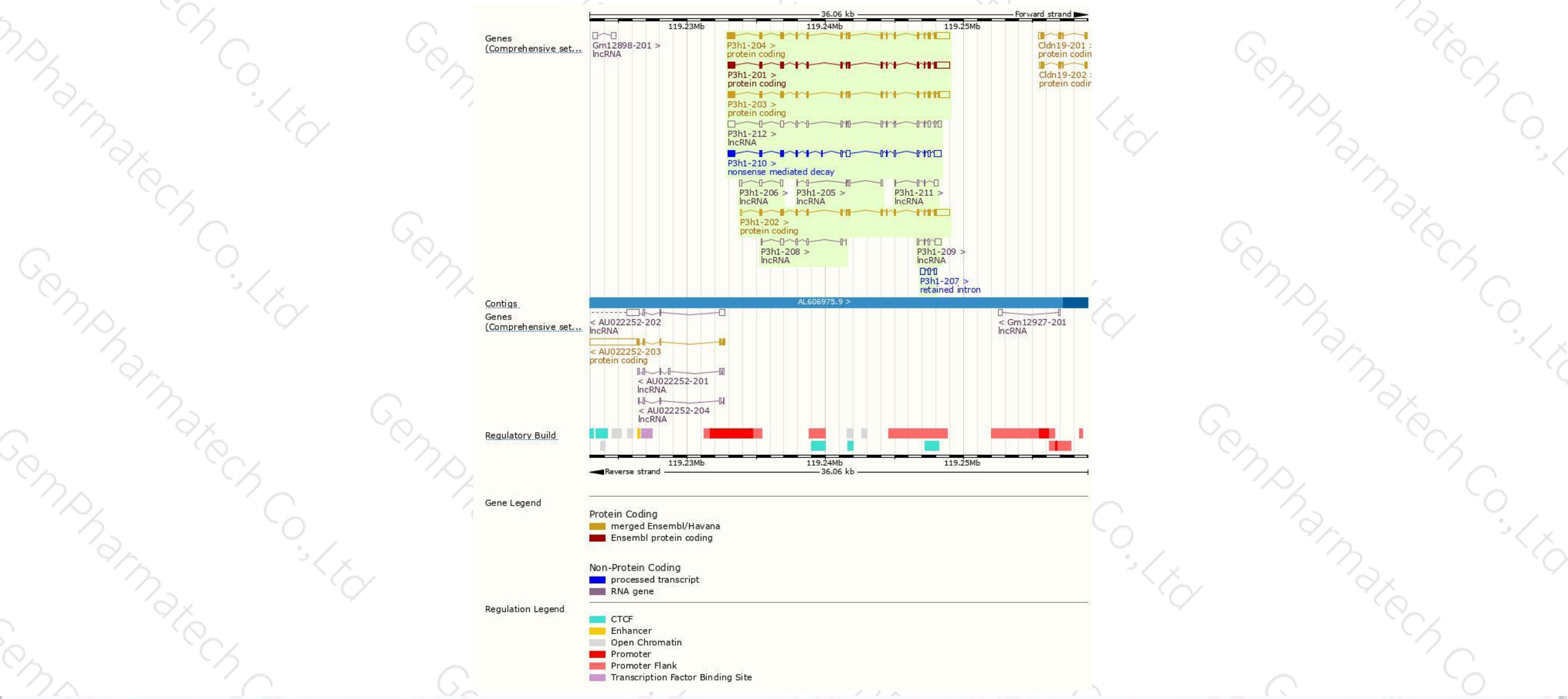
The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
P3h1-201	ENSMUST00000030393.12	3188	746aa	Protein coding	CCDS71460	A6PW84	TSL:1 GENCODE basic APPRIS ALT2
P3h1-204	ENSMUST00000121111.8	3188	739aa	Protein coding	CCDS38859	Q3V1T4	TSL:1 GENCODE basic APPRIS ALT2
P3h1-203	ENSMUST00000102662.10	2977	748aa	Protein coding	CCDS18576	A2A7Q5	TSL:5 GENCODE basic APPRIS P3
P3h1-202	ENSMUST00000081606.6	2820	560aa	Protein coding	CCDS38860	Q3V1T4	TSL:1 GENCODE basic APPRIS ALT2
P3h1-210	ENSMUST00000136278.7	2750	363aa	Nonsense mediated decay	-	F2Z499	TSL:5
P3h1-207	ENSMUST00000127087.1	650	No protein	Retained intron	-	-	TSL:2
P3h1-212	ENSMUST00000153940.7	2278	No protein	lncRNA	-	-	TSL:5
P3h1-209	ENSMUST00000134548.1	714	No protein	lncRNA	-	-	TSL:5
P3h1-208	ENSMUST00000131875.7	624	No protein	lncRNA	-	-	TSL:3
P3h1-211	ENSMUST00000142361.7	525	No protein	lncRNA	-	-	TSL:5
P3h1-205	ENSMUST00000123963.1	498	No protein	lncRNA	-	-	TSL:5
P3h1-206	ENSMUST00000126056.1	491	No protein	lncRNA	-	-	TSL:5

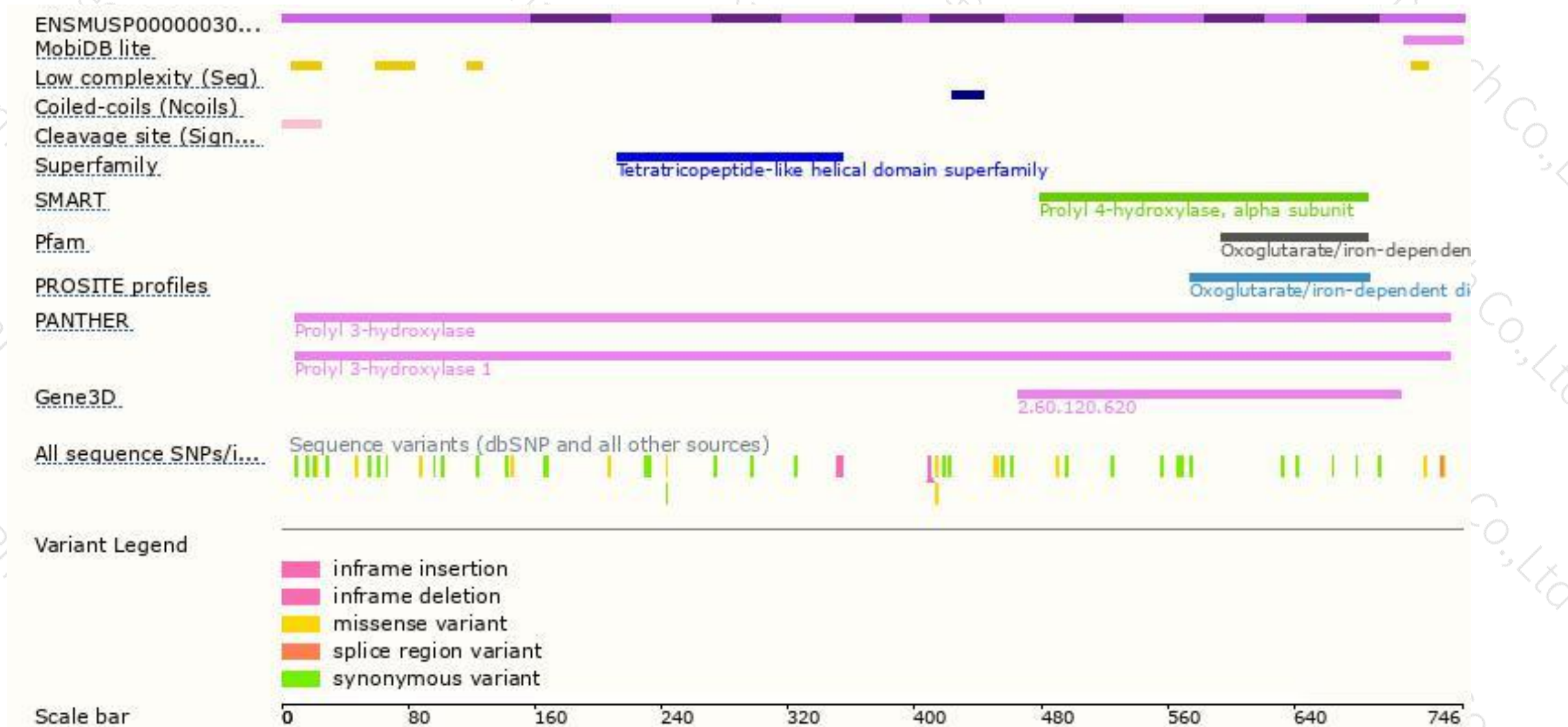
The strategy is based on the design of *P3h1-201* transcript,The transcription is shown below



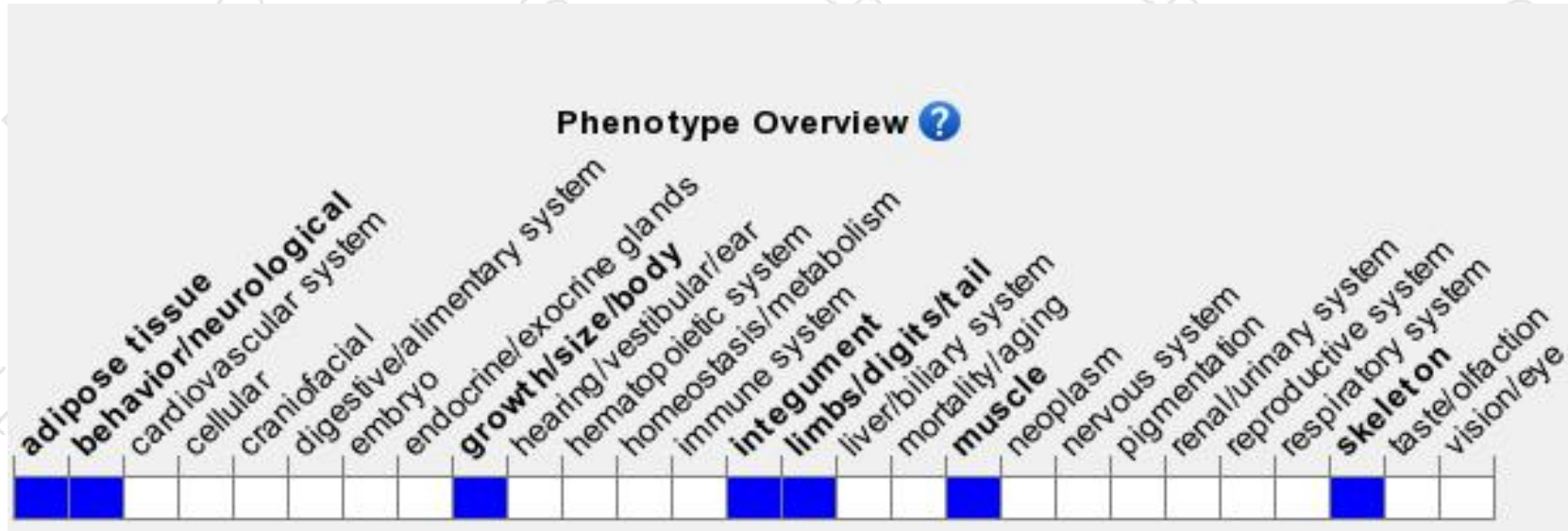
Genomic location distribution



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, Mice homozygous for a knock-out allele exhibit reduced size, disproportional reduction in long bone length, decreased bone density, decreased bone mineral density, reduced body fat, delayed ossification, and abnormal collagen networks in the skin and tendons.

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

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