Myo7a Cas9-CKO Strategy

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Project Overview

Project Name	***Myo7a***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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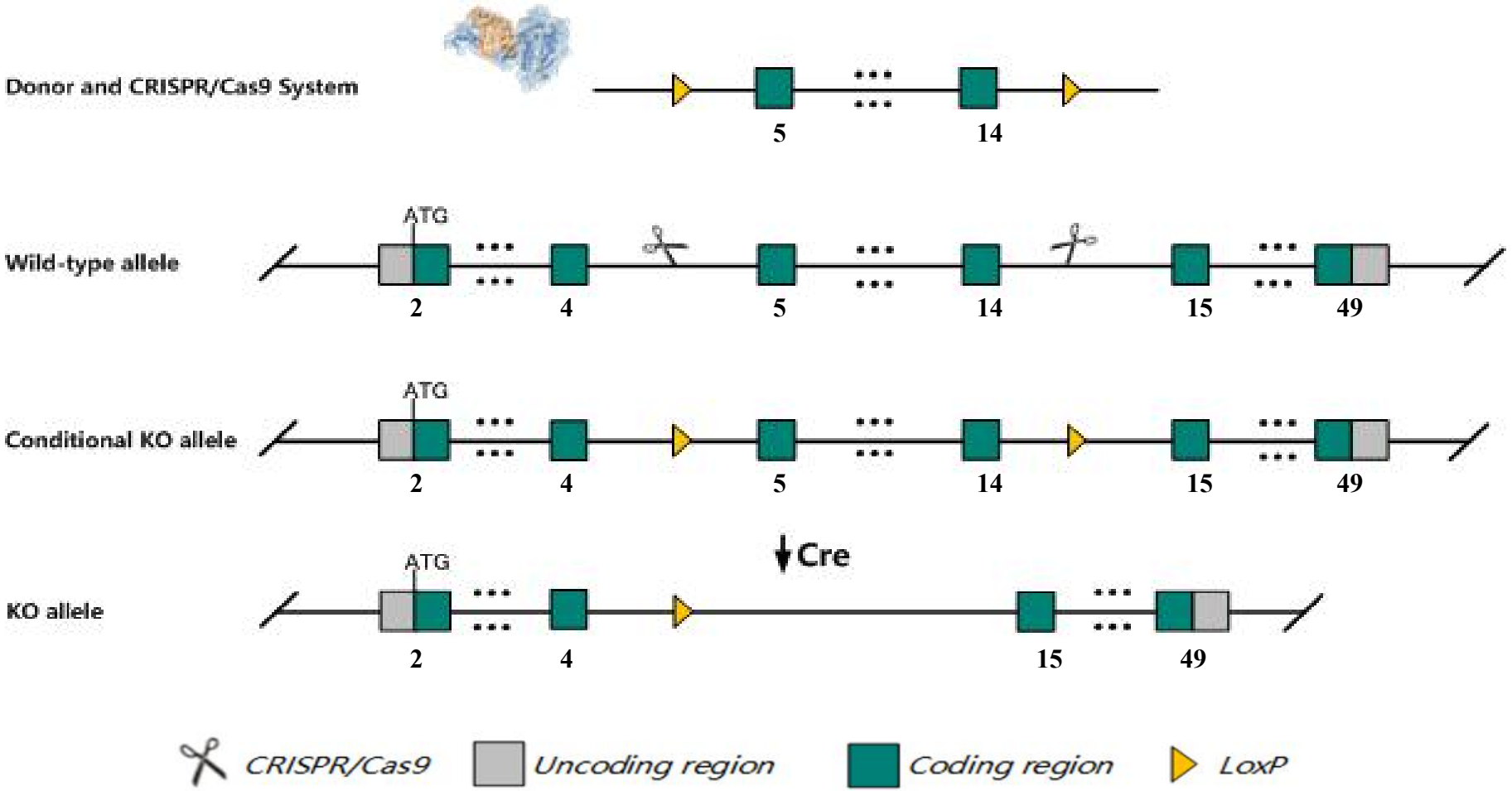
This model will use CRISPR/Cas9 technology to edit the *Myo7a* gene. The schematic diagram is as follows:



Technical routes

The *Myo7a* gene has 15 transcripts. According to the structure of *Myo7a* gene, exon5-exon14 of *Myo7a-204* (ENSMUST00000107128.7) transcript is recommended as the knockout region.The region contains 1405bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Myo7a* 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,A number of spontaneous and ENU-induced mutations cause head-shaking, circling and deafness, often associated with cochlear hair cell degeneration and stereocilia anomalies. Defects in retinal pigment epithelial cells, male infertility, and light-inducedphotoreceptor damage have also been observed.

No damage to transcript Myo7a-212.

The *Myo7a* gene is located on the Chr7. 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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Myo7a myosin VIIA [Mus musculus (house mouse)]

Gene ID: 17921, updated on 19-Feb-2019

Summary

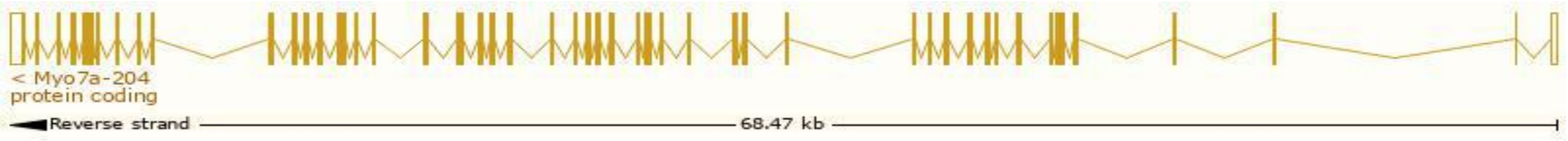
Official Symbol	Myo7a provided by MGI
Official Full Name	myosin VIIA provided by MGI
Primary source	MGI:MGI:104510
See related	Ensembl:ENSMUSG00000030761
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Hdb, Myo7, USH1B, nmf371, polka, sh-1, sh1
Expression	Broad expression in adrenal adult (RPKM 25.0), testis adult (RPKM 17.2) and 25 other tissues See more
Orthologs	human all

Transcript information Ensembl

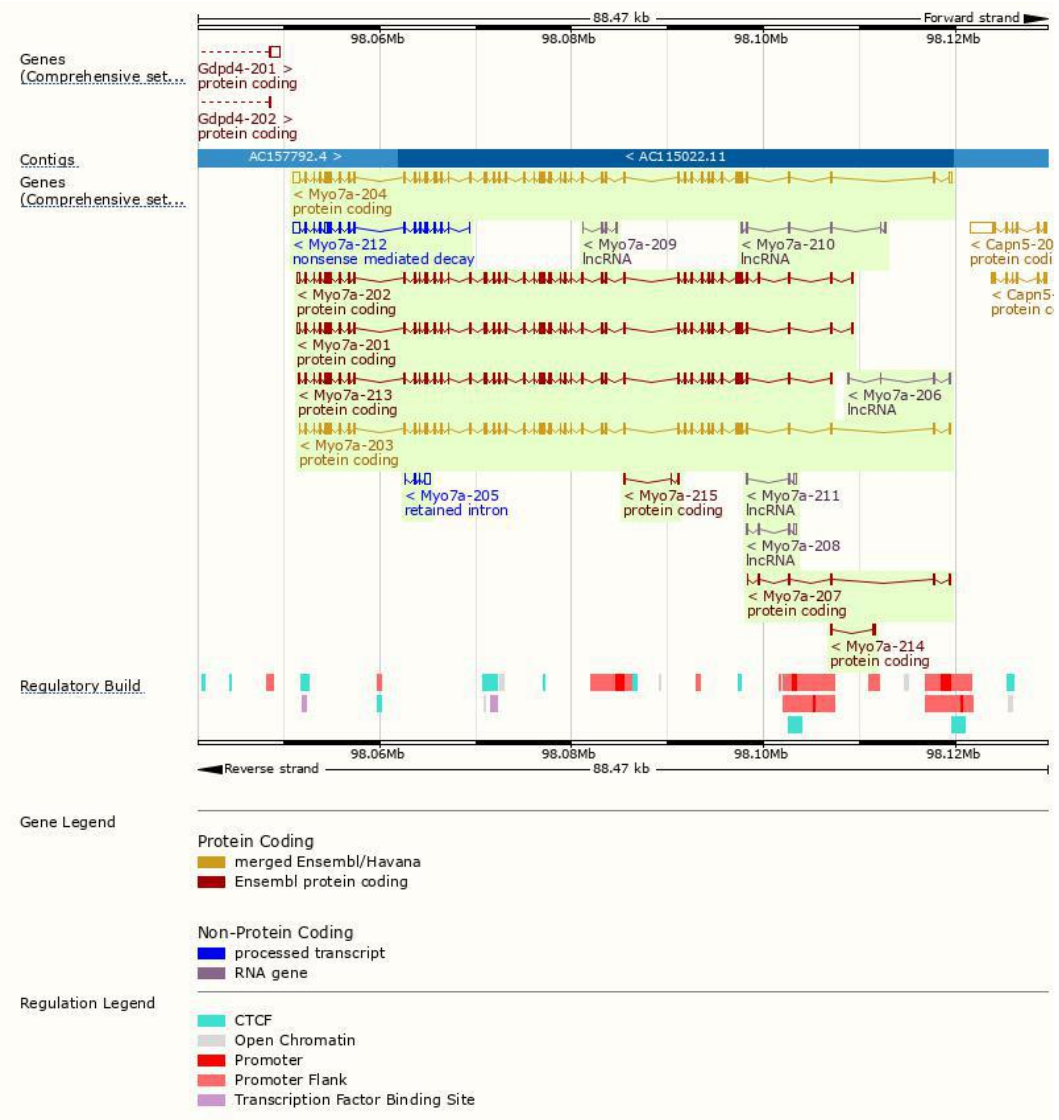
The gene has 15 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Myo7a-204	ENSMUST00000107128.7	7506	2215aa	Protein coding	CCDS57565	P97479	TSL:5 GENCODE basic
Myo7a-202	ENSMUST00000107122.7	6867	2172aa	Protein coding	CCDS57564	Q5MJ56	TSL:1 GENCODE basic
Myo7a-201	ENSMUST00000084979.10	6849	2166aa	Protein coding	CCDS57563	A0A0R4J113	TSL:1 GENCODE basic
Myo7a-203	ENSMUST00000107127.7	6806	2177aa	Protein coding	CCDS40026	P97479	TSL:5 GENCODE basic APPRIS P1
Myo7a-213	ENSMUST00000205746.1	6689	2164aa	Protein coding	-	A0A0U1RPX7	TSL:5 GENCODE basic
Myo7a-207	ENSMUST00000138627.1	679	139aa	Protein coding	-	D3YUT5	CDS 3' incomplete TSL:3
Myo7a-214	ENSMUST00000238309.1	368	76aa	Protein coding	-	-	CDS 3' incomplete
Myo7a-215	ENSMUST00000238540.1	299	100aa	Protein coding	-	-	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete
Myo7a-212	ENSMUST00000156992.7	2853	215aa	Nonsense mediated decay	-	A0A0U1RPT3	CDS 5' incomplete TSL:5
Myo7a-205	ENSMUST00000124787.1	838	No protein	Retained intron	-	-	TSL:3
Myo7a-210	ENSMUST00000153657.7	647	No protein	lncRNA	-	-	TSL:5
Myo7a-206	ENSMUST00000131632.1	498	No protein	lncRNA	-	-	TSL:3
Myo7a-211	ENSMUST00000155637.7	493	No protein	lncRNA	-	-	TSL:2
Myo7a-208	ENSMUST00000149079.1	467	No protein	lncRNA	-	-	TSL:2
Myo7a-209	ENSMUST00000152975.1	347	No protein	lncRNA	-	-	TSL:5

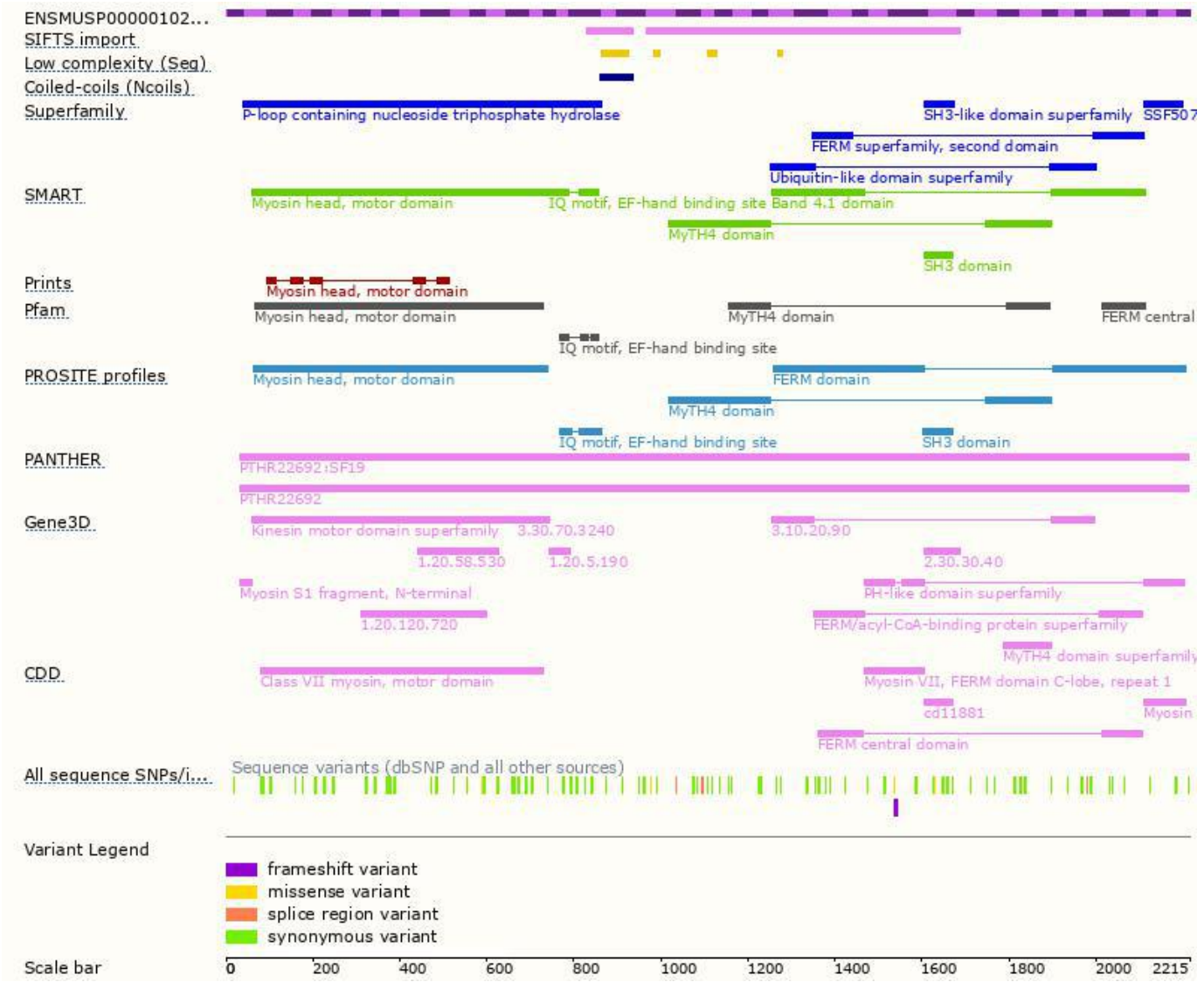
The strategy is based on the design of *Myo7a-204* transcript,The transcription is shown below



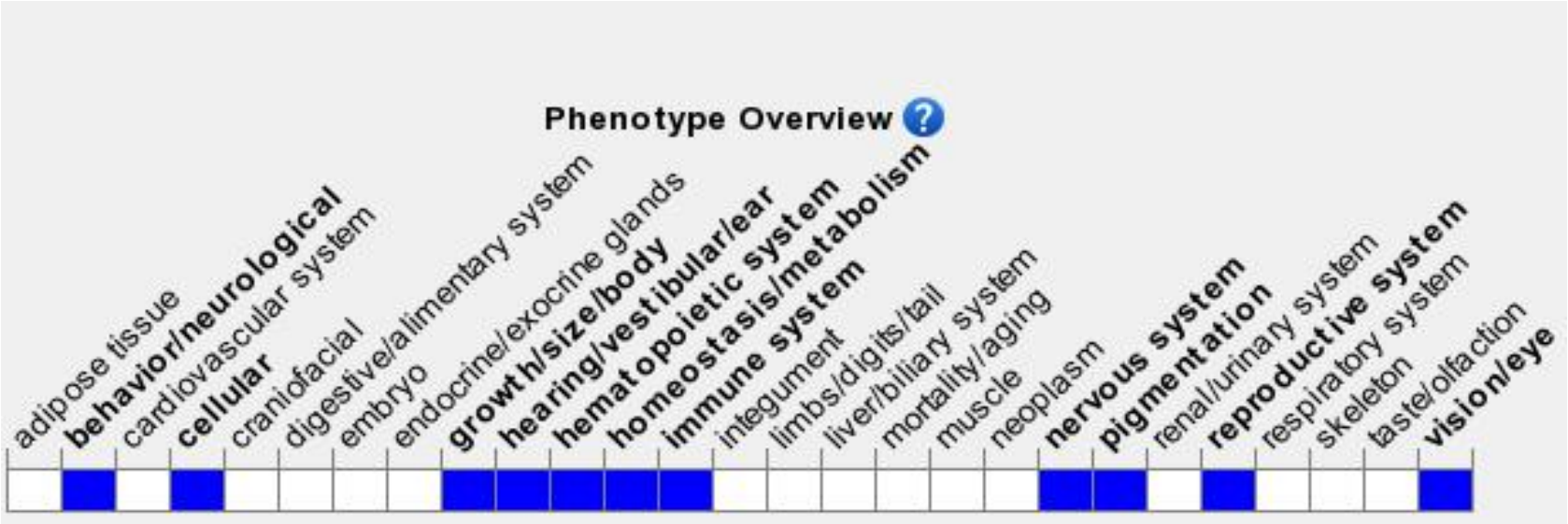
Genomic location distribution



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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