Clic5 Cas9-CKO Strategy

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Design Date: 2019-9-10

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Project Overview

Project Name	***Clic5***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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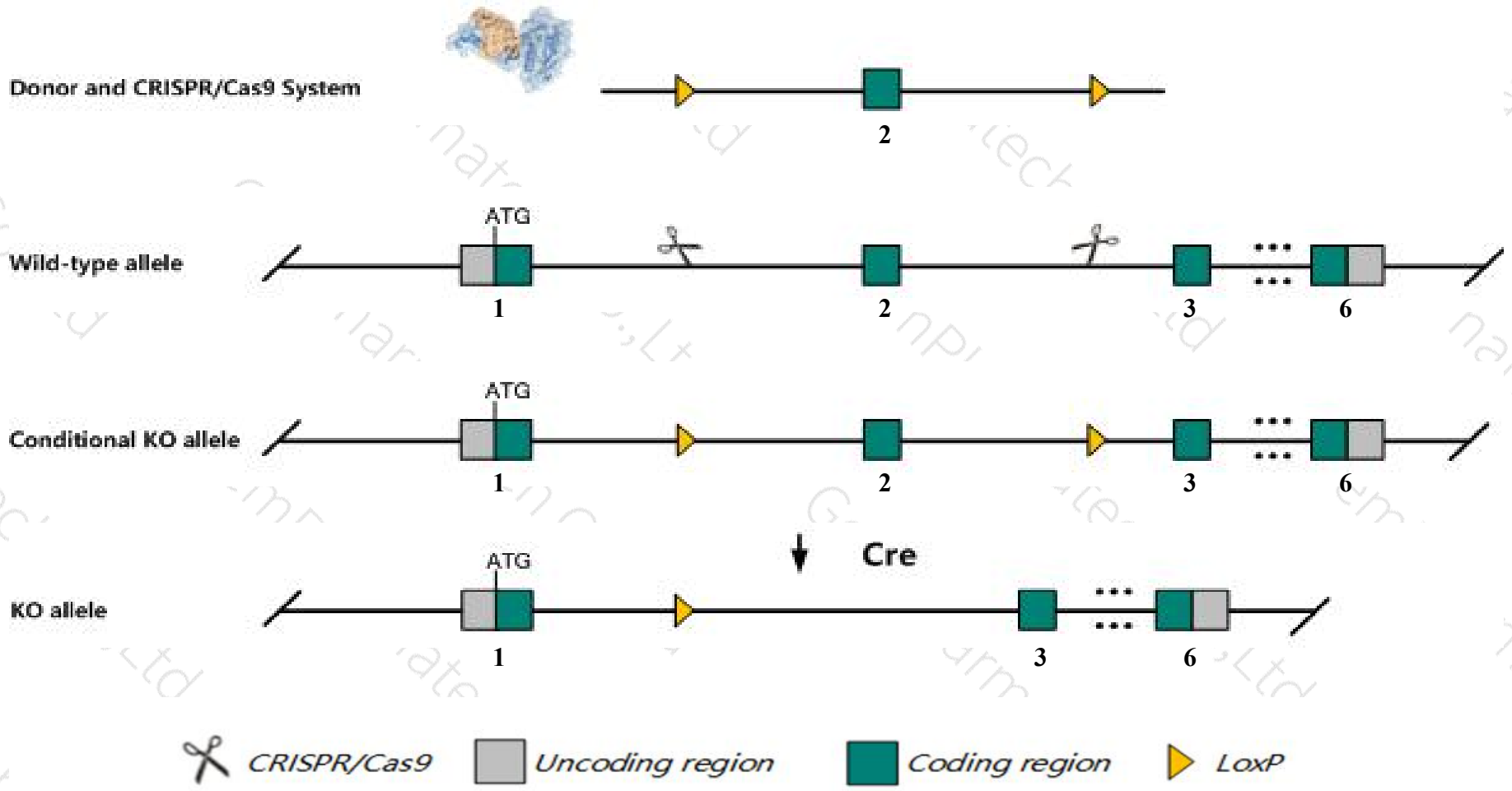
This model will use CRISPR/Cas9 technology to edit the *Clic5* gene. The schematic diagram is as follows:



Technical routes

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- The *Clic5* gene has 2 transcripts. According to the structure of *Clic5* gene, exon2 of *Clic5-201* (ENSMUST00000024755.6) transcript is recommended as the knockout region.The region contains 110bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Clic5* 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,Mice homozygous for a spontaneous mutation exhibit head bobbing and circling behavior and complete deafness by 7-8 months of age caused by dysmorphic stereocilia and progressive hair cell degeneration. Mice are resistant to diet-induced obesity and are unable to maintain energy reserves. Cataracts develop with age. Renal glomerulus defects are present.

- The *Clic5* gene is located on the Chr17. 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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Clic5 chloride intracellular channel 5 [Mus musculus (house mouse)]

Gene ID: 224796, updated on 31-Jan-2019

Summary

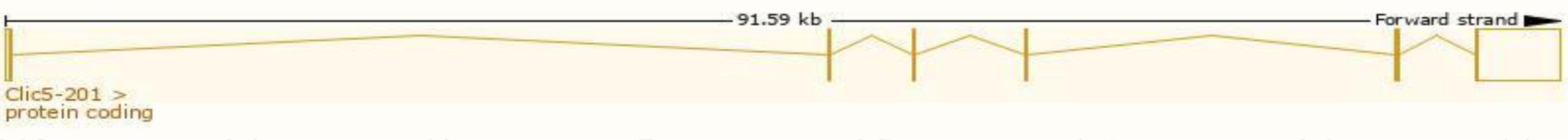
Official Symbol	Clic5 provided by MGI
Official Full Name	chloride intracellular channel 5 provided byMGI
Primary source	MGI:MGI:1917912
See related	Ensembl:ENSMUSG00000023959
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	5730531E12Rik, B330005L24, Gm322, jbg, nmf318
Expression	Biased expression in lung adult (RPKM 40.1), large intestine adult (RPKM 14.4) and 10 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

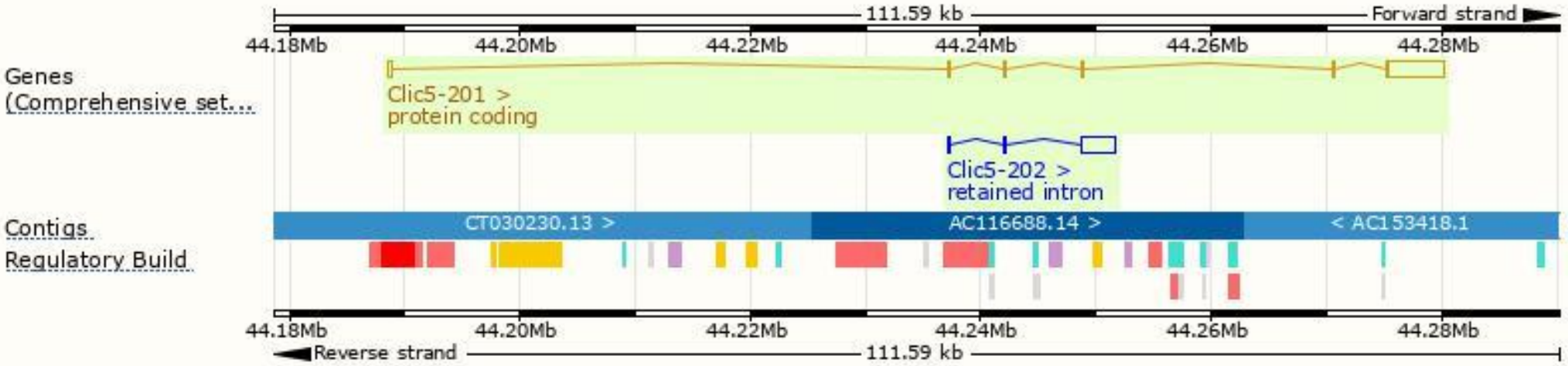
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Clic5-201	ENSMUST00000024755.6	5862	251aa	Protein coding	CCDS28804	Q8BXK9	TSL:1 GENCODE basic APPRIS P1
Clic5-202	ENSMUST00000233193.1	3263	No protein	Retained intron	-	-	

The strategy is based on the design of *Clic5-201* transcript,The transcription is shown below



Genomic location distribution



Gene Legend

Protein Coding
- merged Ensembl/Havana

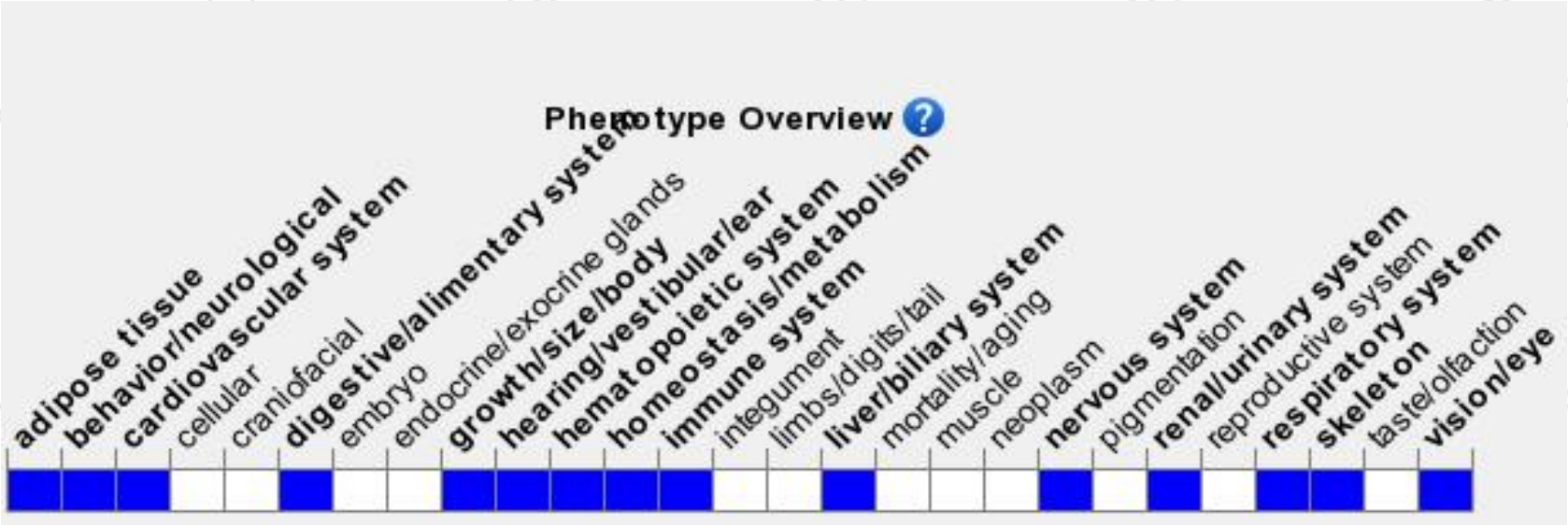
Non-Protein Coding
- processed transcript

Regulation Legend

- CTCF
- Enhancer
- Open Chromatin
- Promoter
- Promoter Flank
- Transcription Factor Binding Site

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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