Agr2 Cas9-CKO Strategy

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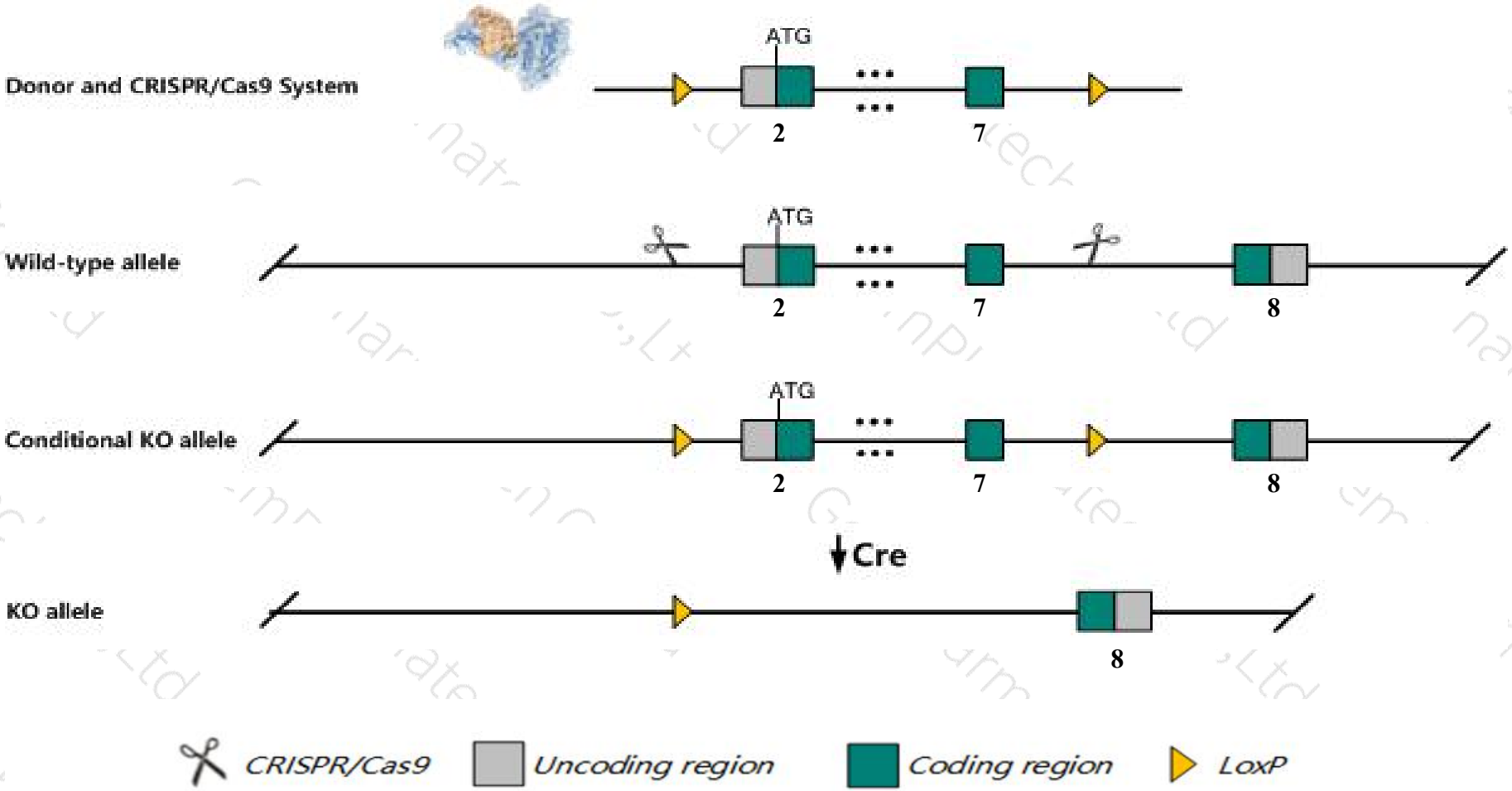
Design Date: 2019-8-6

Project Overview

Project Name	***Agr2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Agr2* gene. The schematic diagram is as follows:



Technical routes

- The *Agr2* gene has 2 transcripts. According to the structure of *Agr2* gene, exon2-exon7 of *Agr2-201* (ENSMUST00000020898.11) transcript is recommended as the knockout region.The region contains start codon ATG. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Agr2* 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 knock-out allele exhibit colitis and increased susceptibility to induced colitis. Mice homozygous for another knock-out allele exhibit hyperplasia and defective lineage maturation in the stomach that leads to intestinal obstruction and premature death.

➢ The *Agr2* gene is located on the Chr12. 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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Agr2 anterior gradient 2 [Mus musculus (house mouse)]

Gene ID: 23795, updated on 31-Jan-2019

Summary

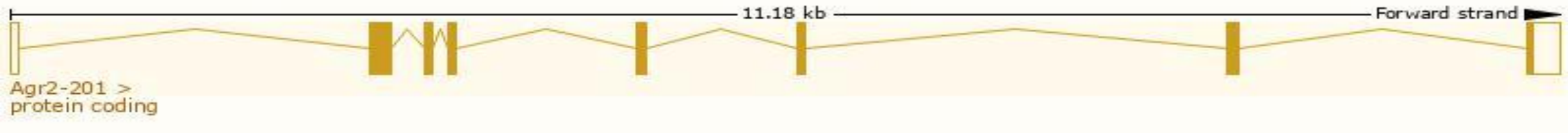
Official Symbol	Agr2 provided by MGI
Official Full Name	anterior gradient 2 provided by MGI
Primary source	MGI:MGI:1344405
See related	Ensembl:ENSMUSG00000020581
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Agr2h, Gob-4, HAG-2, XAG-2, mAG-2
Expression	Biased expression in large intestine adult (RPKM 438.3), colon adult (RPKM 350.8) and 4 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

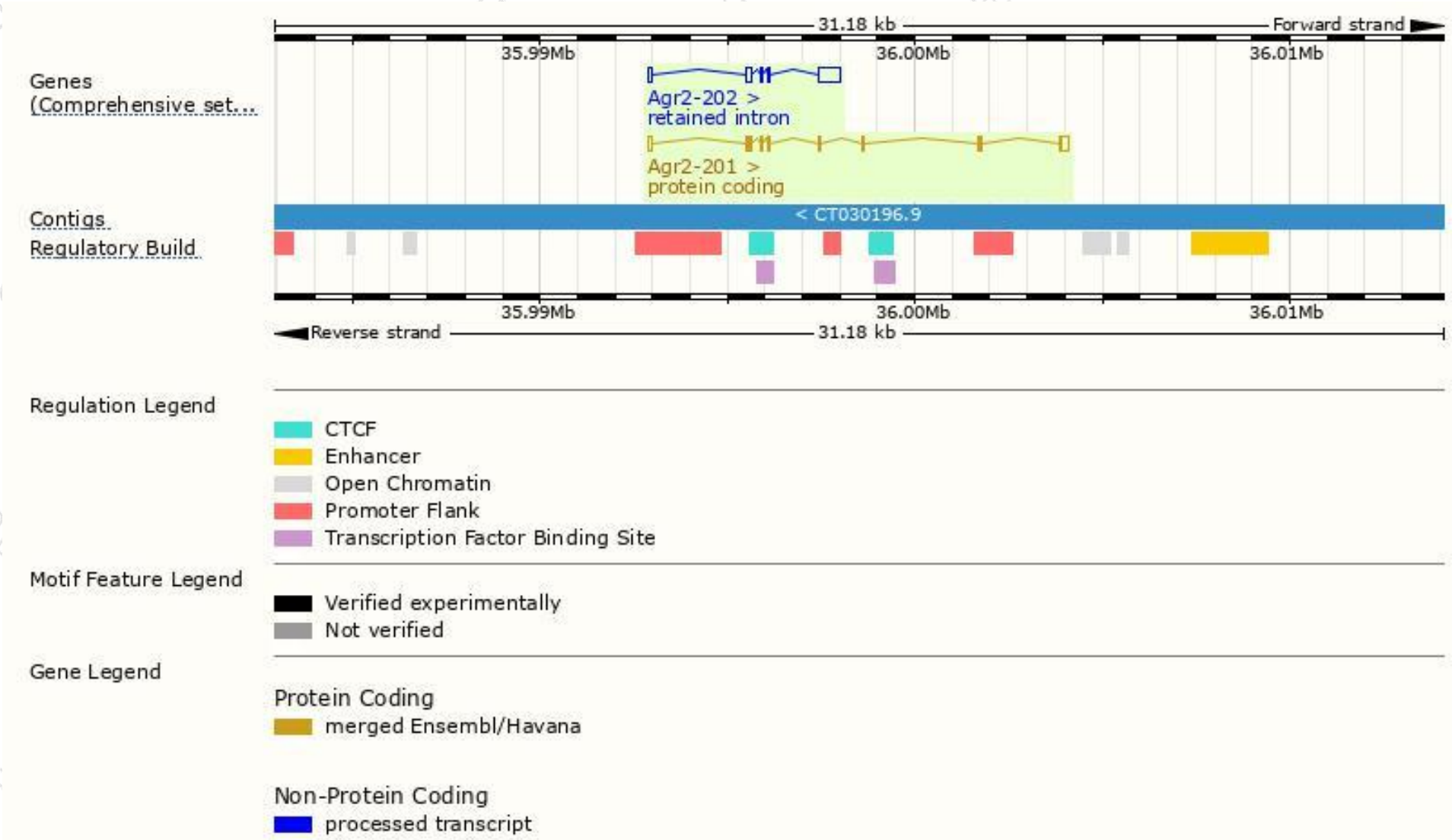
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Agr2-201	ENSMUST00000020898.11	784	175aa	Protein coding	CCDS25881	O88312	TSL:1 GENCODE basic APPRIS P1
Agr2-202	ENSMUST00000147861.1	907	No protein	Retained intron	-	-	TSL:2

The strategy is based on the design of *Agr2-201* transcript,The transcription is shown below

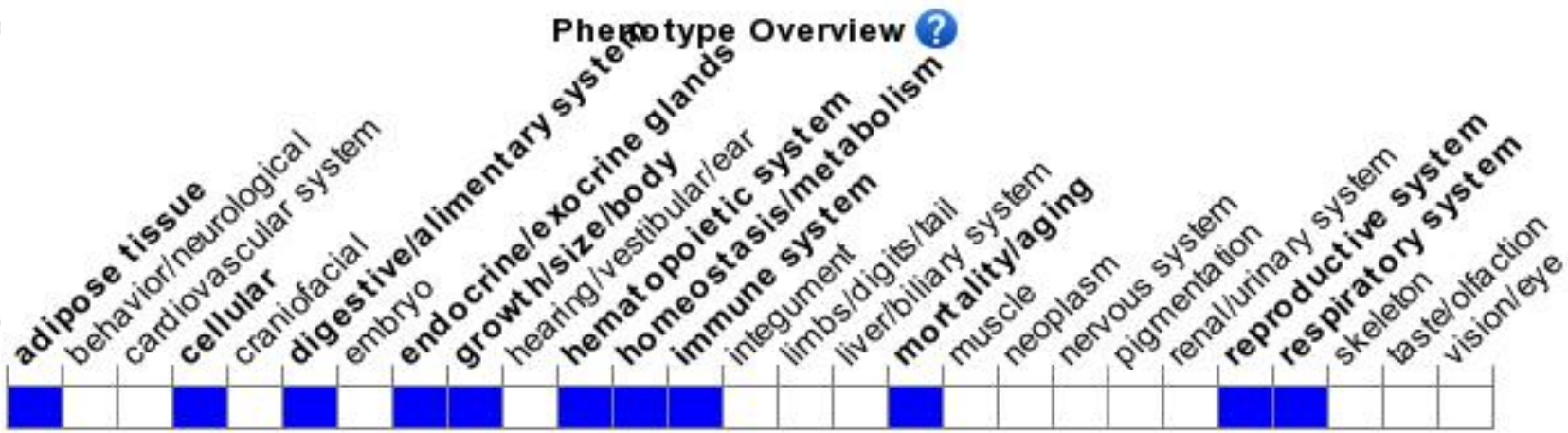


Genomic location distribution



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/).

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

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