

***Gnrhr* Cas9-CKO Strategy**

Designer:

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Design Date:

2019-8-23

Project Overview

Project Name

Gnrhr

Project type

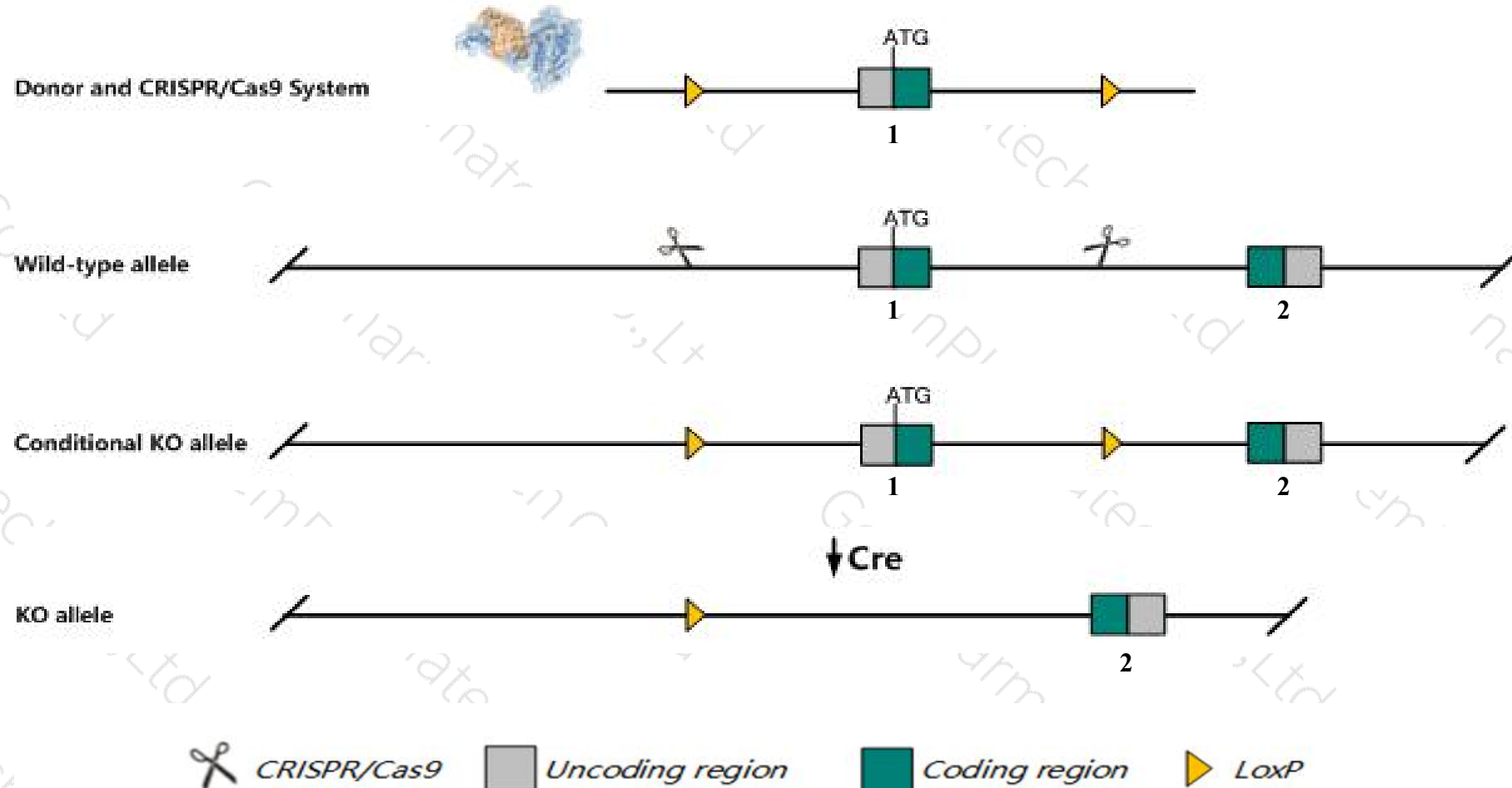
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



- The *Gnrhr* gene has 3 transcripts. According to the structure of *Gnrhr* gene, exon1 of *Gnrhr-203* (ENSMUST00000113372.1) 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 *Gnrhr* 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, Homozygous mutant mice display prepubescent internal reproductive tracts with hypogonadism and hypogonadotrophy.
- The *Gnrhr* gene is located on the Chr5. 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)

Gnrhr gonadotropin releasing hormone receptor [Mus musculus (house mouse)]

Gene ID: 14715, updated on 2-Apr-2019

Summary



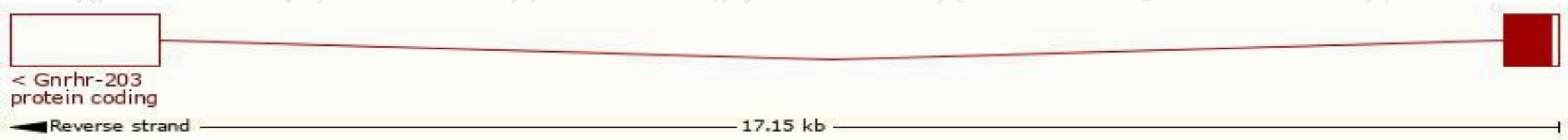
Official Symbol	Gnrhr provided by MGI
Official Full Name	gonadotropin releasing hormone receptor provided by MGI
Primary source	MGI:MGI:95790
See related	Ensembl:ENSMUSG00000029255
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Expression	Low expression observed in reference dataset See more
Orthologs	human all

Transcript information (Ensembl)

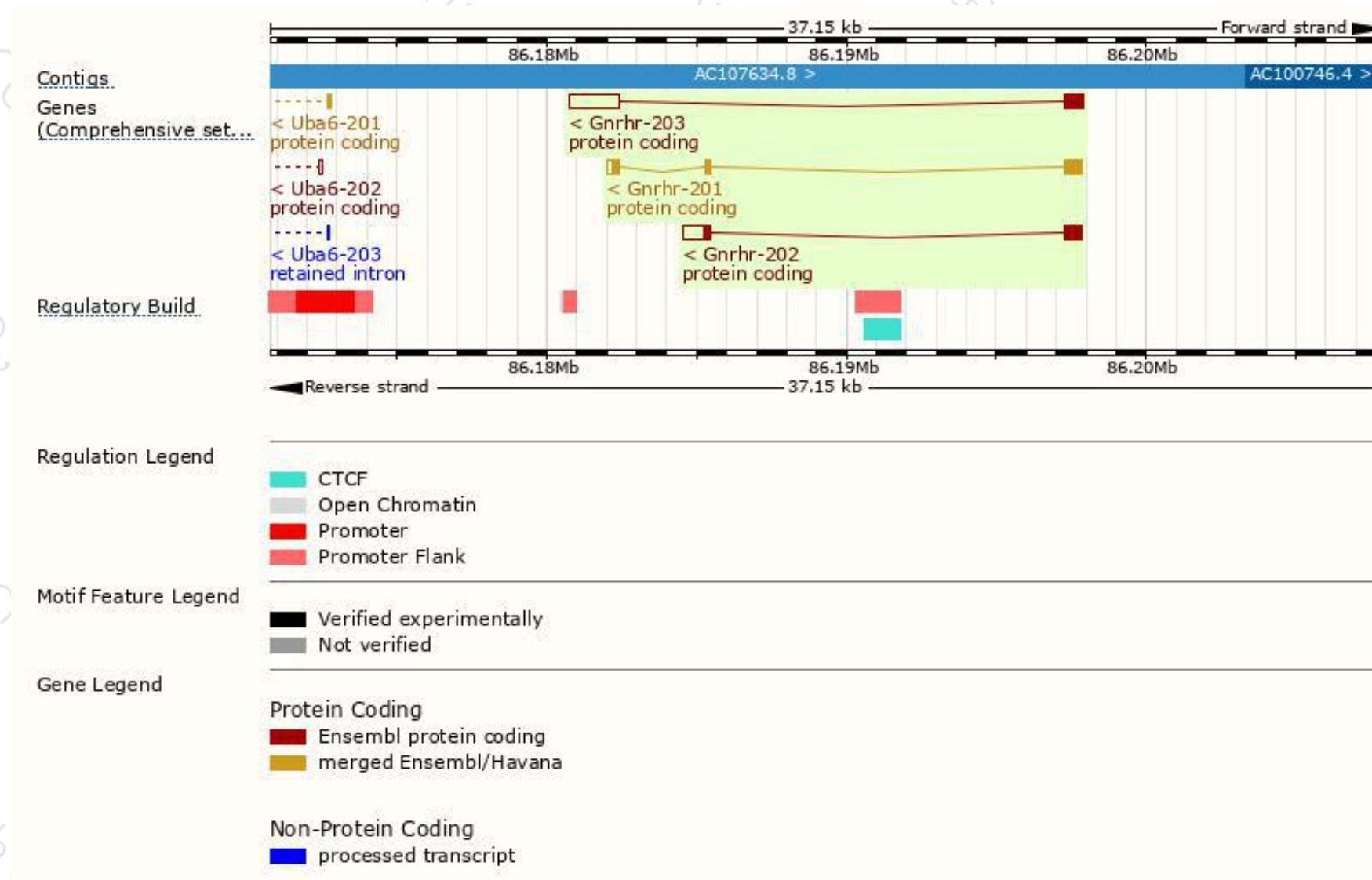
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gnrhr-203	ENSMUST00000113372.1	2265	177aa	Protein coding	CCDS80313	D3Z6P7	TSL:1 GENCODE basic
Gnrhr-202	ENSMUST00000094654.2	1475	264aa	Protein coding	CCDS84897	Q6P8H4	TSL:1 GENCODE basic
Gnrhr-201	ENSMUST00000031172.8	1211	327aa	Protein coding	CCDS19379	Q01776	TSL:1 GENCODE basic APPRIS P1

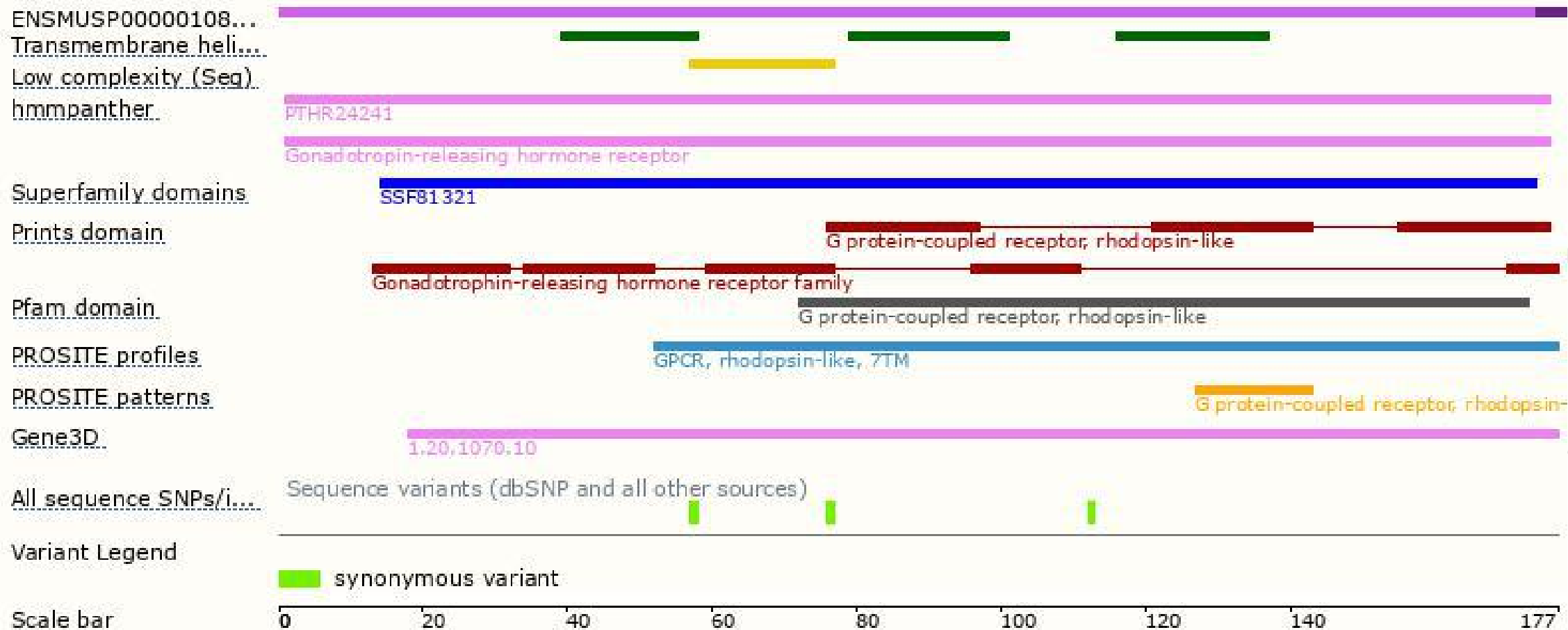
The strategy is based on the design of *Gnrhr-203* transcript,The transcription is shown below



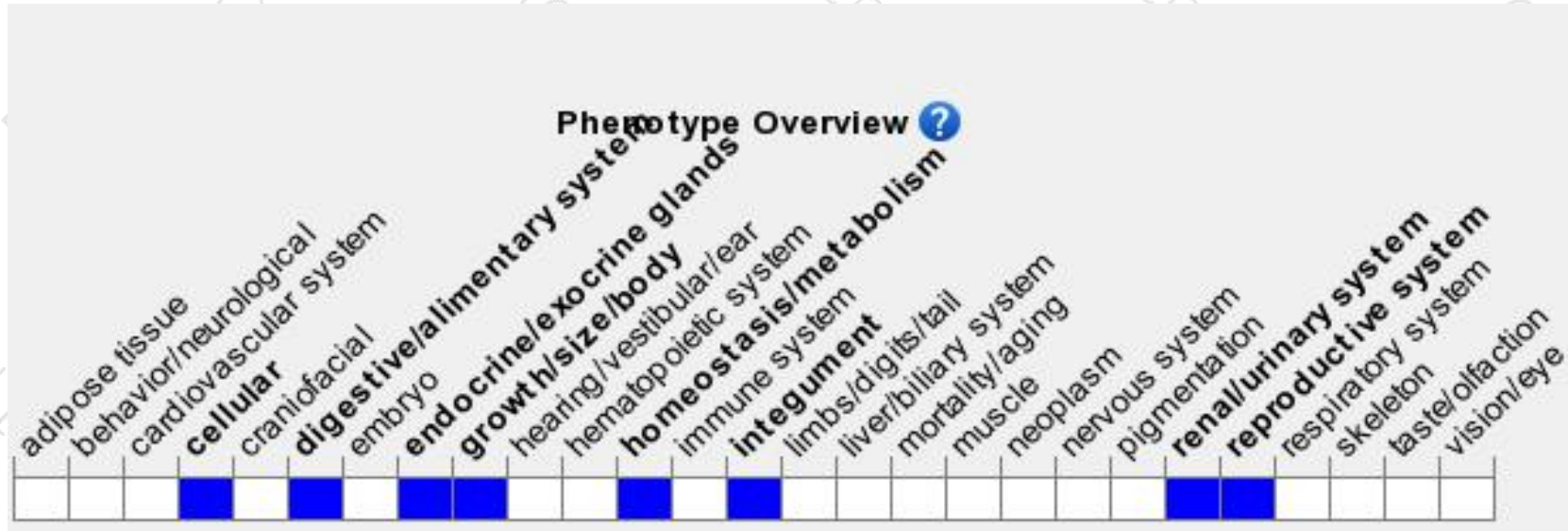
Genomic location distribution



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, Homozygous mutant mice display prepubescent internal reproductive tracts with hypogonadism and hypogonadotrophy.

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

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