

Ube2a Cas9-CKO Strategy

Designer

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Reviewer

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Design Date

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Project Overview

Project Name

Ube2a

Project type

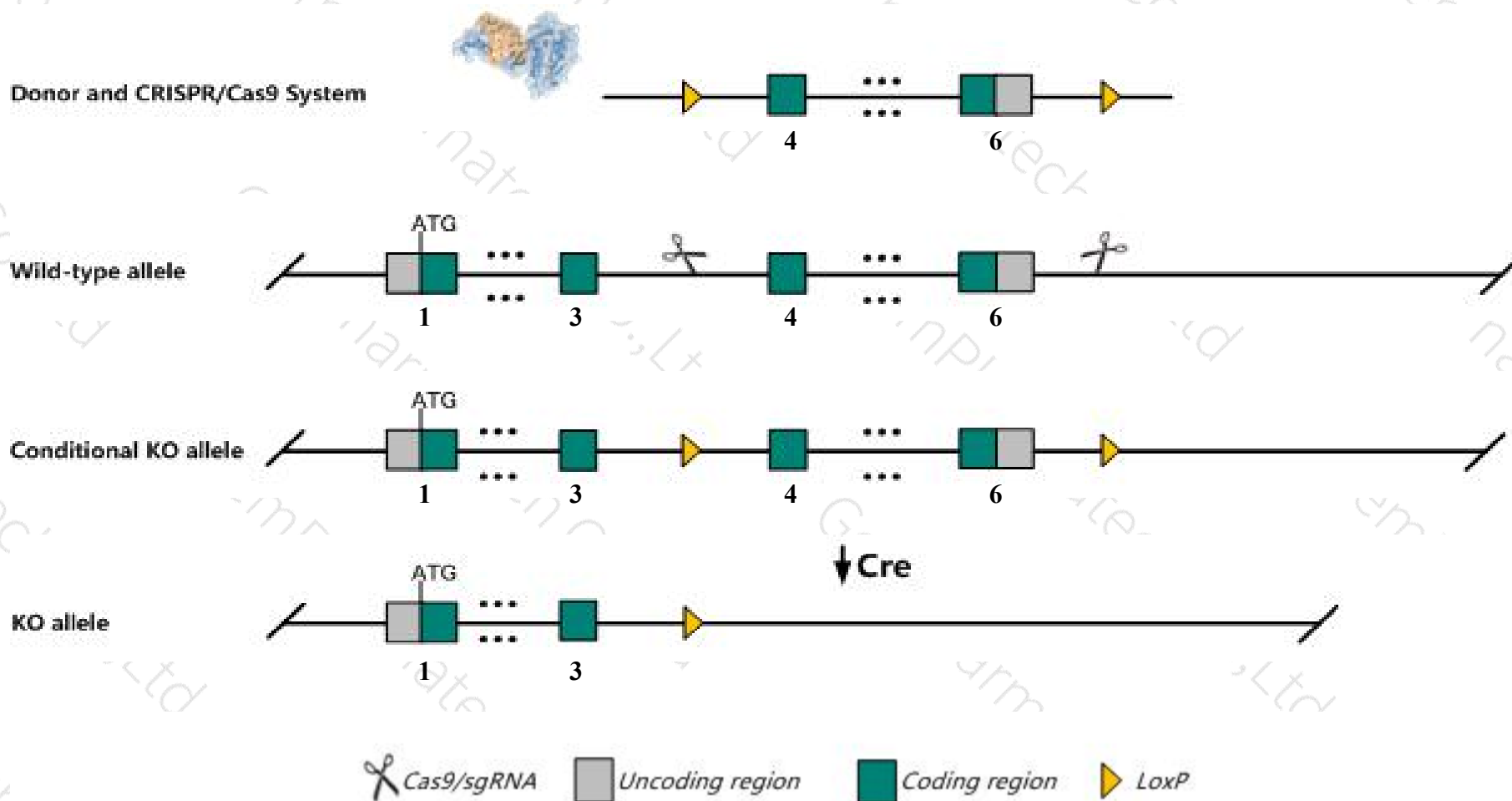
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ube2a* gene. The schematic diagram is as follows:



Technical routes

- The *Ube2a* gene has 9 transcripts. According to the structure of *Ube2a* gene, exon4-exon6 of *Ube2a-201* (ENSMUST00000016452.10) transcript is recommended as the knockout region. The region contains 308bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ube2a* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 was 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 mutation of this gene results in decreased body weight. Females are infertile while males show partial prenatal lethality.
- The *Ube2a* gene is located on the ChrX. 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)

Ube2a ubiquitin-conjugating enzyme E2A [*Mus musculus* (house mouse)]

Gene ID: 22209, updated on 31-Jan-2019

Summary

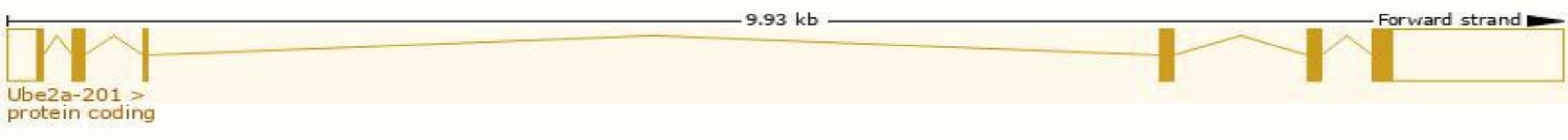
Official Symbol	Ube2a provided by MGI
Official Full Name	ubiquitin-conjugating enzyme E2A provided by MGI
Primary source	MGI:MGI:102959
See related	Ensembl:ENSMUSG00000016308
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	HR6A; HHR6A; Mhr6a
Expression	Ubiquitous expression in placenta adult (RPKM 13.3), CNS E18 (RPKM 11.1) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

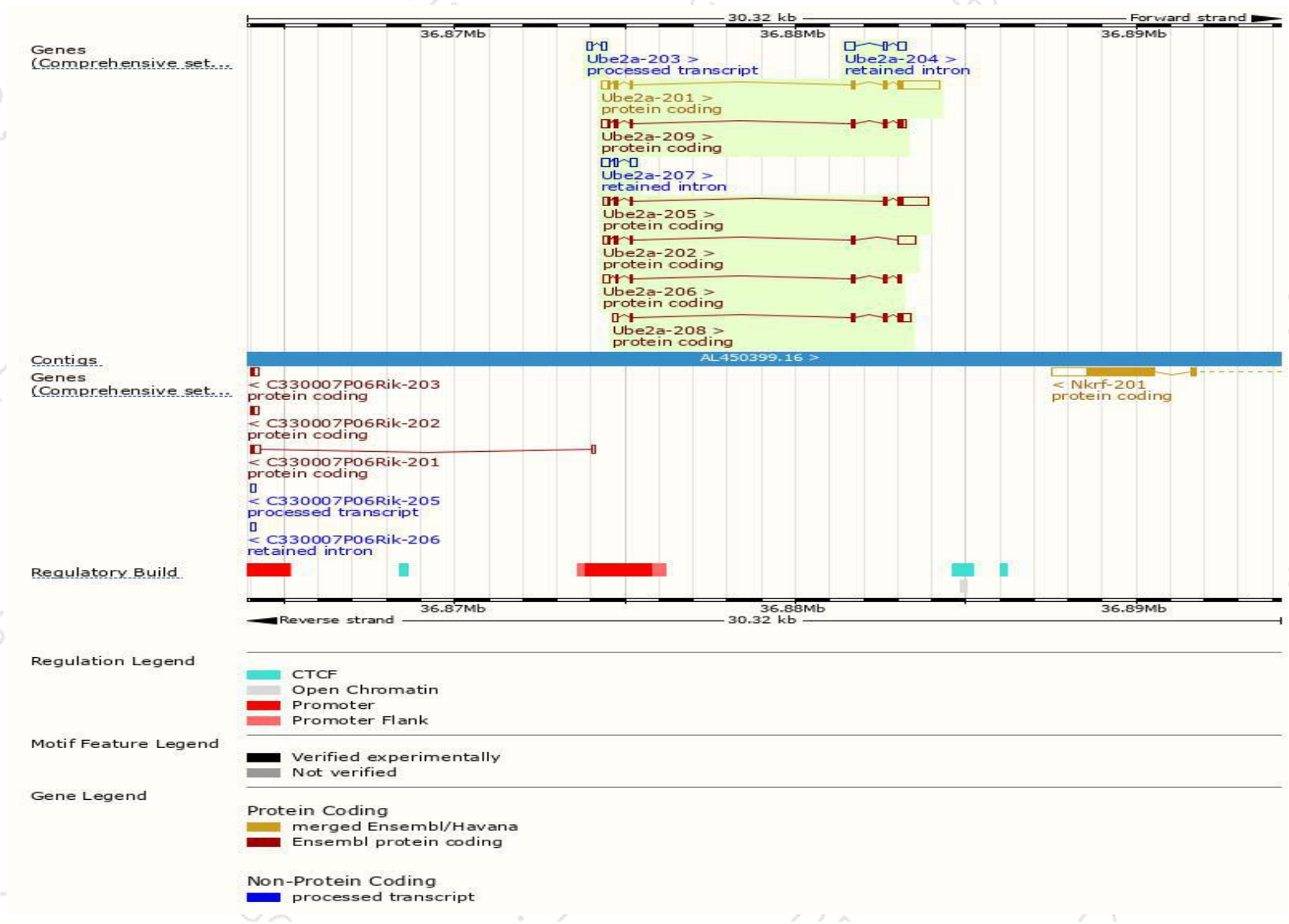
The gene has 9 transcripts,all transcripts are shown below:

Show/hide columns								Filter		
Name ▲	Transcript ID ▲	bp ▲	Protein ▲	Translation ID ▲	Biotype ▲	CCDS ▲	UniProt ▲	Flags ▲		
Ube2a-201	ENSMUST00000016452.10	1732	152aa	ENSMUSP00000016452.7	Protein coding	CCDS30064	Q3UCS1 Q9Z255	TSL:1	GENCODE basic	APPRIS P3
Ube2a-202	ENSMUST000000200835.3	911	84aa	ENSMUSP000000144306.1	Protein coding	-	A0A0J9YUR7	TSL:5	GENCODE basic	
Ube2a-203	ENSMUST000000201012.1	287	No protein	-	lncRNA	-	-	TSL:2		
Ube2a-204	ENSMUST000000201050.1	603	No protein	-	Retained intron	-	-	TSL:2		
Ube2a-205	ENSMUST000000201068.3	1298	122aa	ENSMUSP000000143836.1	Protein coding	-	A0A0J9YTT1	TSL:5	GENCODE basic	
Ube2a-206	ENSMUST000000201117.3	552	110aa	ENSMUSP000000144372.1	Protein coding	-	A0A0J9YUW6	CDS 3' incomplete	TSL:5	
Ube2a-207	ENSMUST000000202783.1	481	No protein	-	Retained intron	-	-	TSL:2		
Ube2a-208	ENSMUST000000202812.1	719	119aa	ENSMUSP000000144262.1	Protein coding	-	A0A0J9YUN2	TSL:2	GENCODE basic	
Ube2a-209	ENSMUST000000202991.3	715	149aa	ENSMUSP000000144242.1	Protein coding	CCDS85765	Q3TQ27	TSL:1	GENCODE basic	APPRIS ALT1

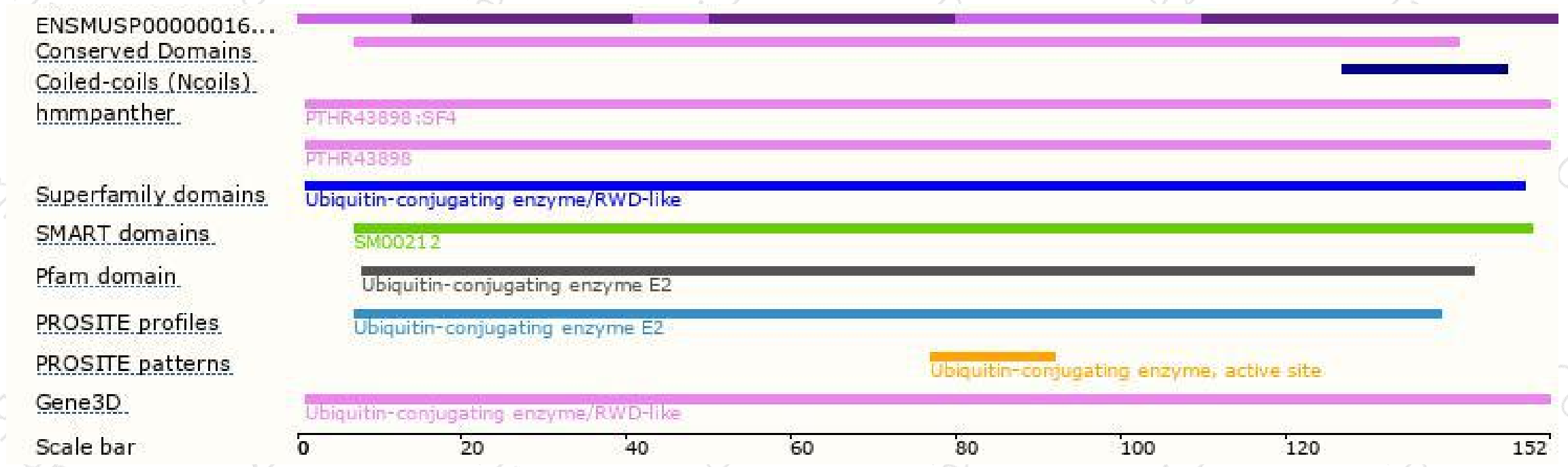
The strategy is based on the design of *Ube2a-201* transcript,The transcription is shown below



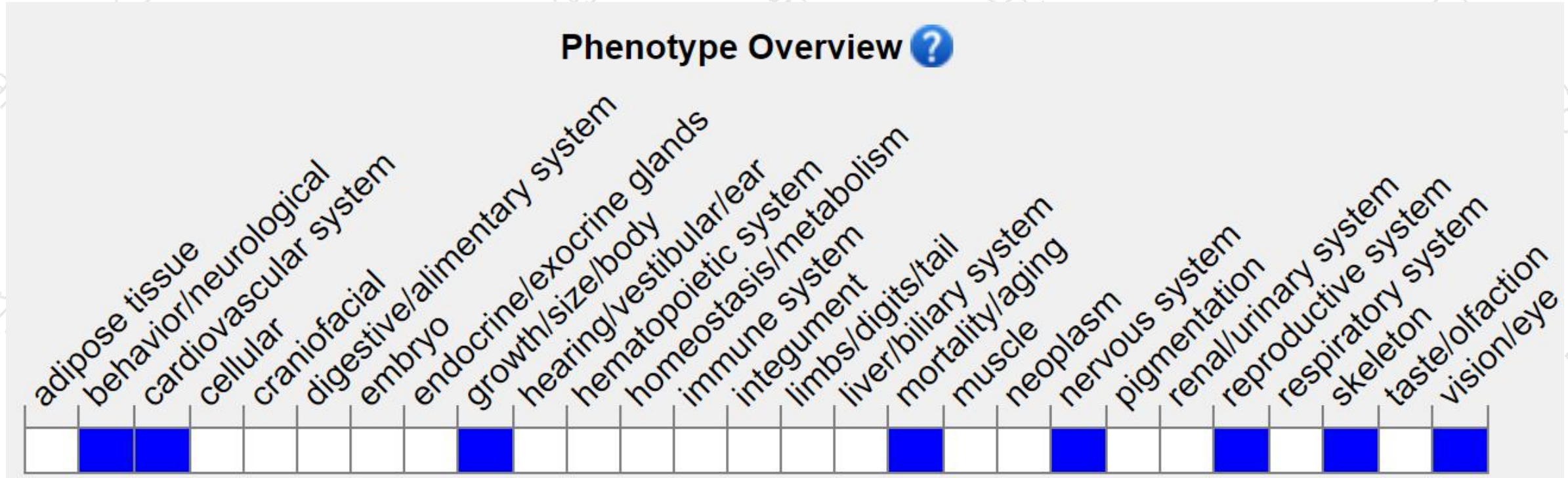
Genomic location distribution



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, Homozygous mutation of this gene results in decreased body weight. Females are infertile while males show partial prenatal lethality.

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

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