

***Rbm5* Cas9-CKO Strategy**

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Project Overview

Project Name

Rbm5

Project type

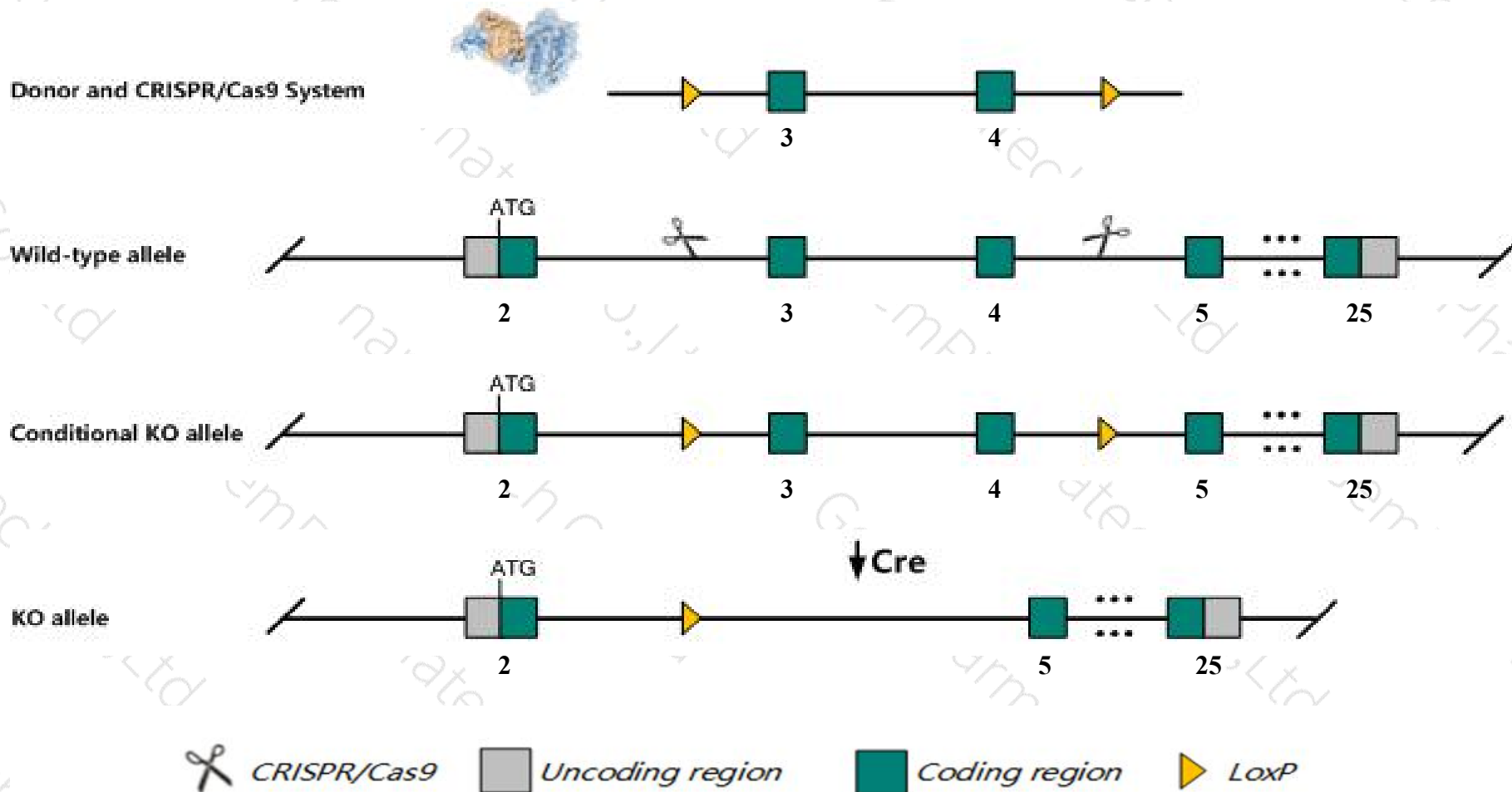
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rbm5* gene. The schematic diagram is as follows:



- The *Rbm5* gene has 24 transcripts. According to the structure of *Rbm5* gene, exon3-exon4 of *Rbm5*-214 (ENSMUST00000182659.7) transcript is recommended as the knockout region. The region contains 322bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rbm5* 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, mice homozygous for an enu-induced allele exhibit male infertility with azoospermia, male germ cell apoptosis, round spermatid arrest and spermatid differentiation arrest.
- Transcript *Rbm5-205&222* may not be affected.
- The *Rbm5* gene is located on the Chr9. 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)

Rbm5 RNA binding motif protein 5 [Mus musculus (house mouse)]

Gene ID: 83486, updated on 20-Mar-2020

Summary



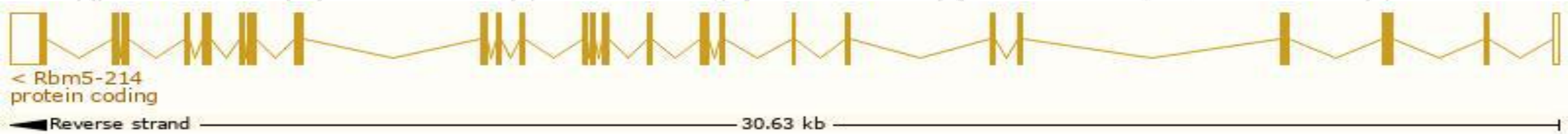
Official Symbol	Rbm5 provided by MGI
Official Full Name	RNA binding motif protein 5 provided by MGI
Primary source	MGI:MGI:1933204
See related	Ensembl:ENSMUSG00000032580
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	D030069N10Rik
Expression	Ubiquitous expression in CNS E14 (RPKM 44.1), CNS E11.5 (RPKM 42.8) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

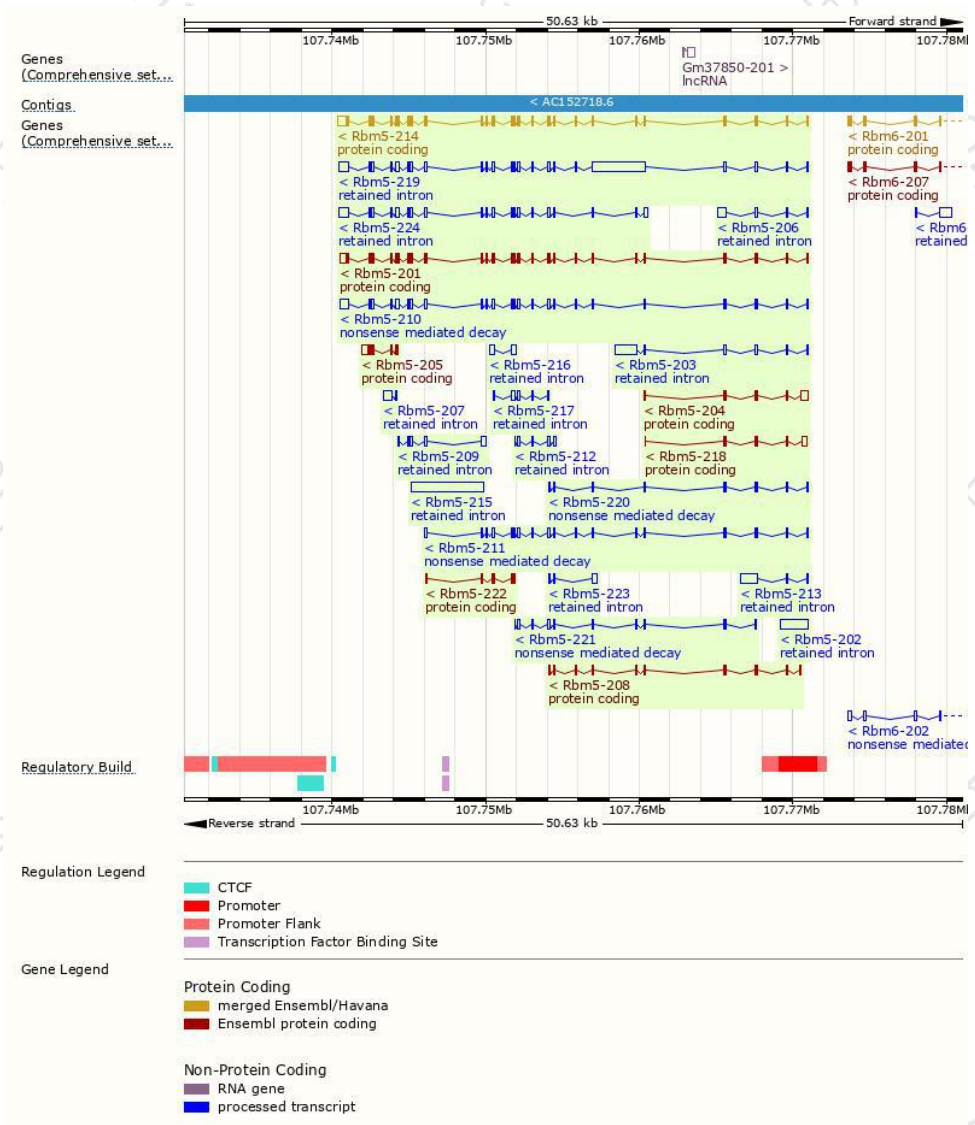
The gene has 24 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rbm5-214	ENSMUST00000182659.7	3205	815aa	Protein coding	CCDS23506	Q91YE7	TSL:1 GENCODE basic APPRIS P2
Rbm5-201	ENSMUST00000035199.12	3064	814aa	Protein coding	-	Q91YE7	TSL:5 GENCODE basic APPRIS ALT1
Rbm5-204	ENSMUST00000182022.7	869	137aa	Protein coding	-	S4R1V5	CDS 3' incomplete TSL:5
Rbm5-208	ENSMUST00000182304.7	850	251aa	Protein coding	-	S4R1U7	CDS 3' incomplete TSL:3
Rbm5-205	ENSMUST00000182026.1	835	154aa	Protein coding	-	S4R273	CDS 5' incomplete TSL:2
Rbm5-218	ENSMUST00000183248.2	781	115aa	Protein coding	-	S4R1N4	CDS 3' incomplete TSL:5
Rbm5-222	ENSMUST00000194400.1	322	101aa	Protein coding	-	A0A0A6YY86	CDS 5' incomplete TSL:3
Rbm5-210	ENSMUST00000182371.7	2986	150aa	Nonsense mediated decay	-	S4R184	TSL:1
Rbm5-211	ENSMUST00000182421.7	1704	115aa	Nonsense mediated decay	-	S4R2T4	TSL:1
Rbm5-221	ENSMUST00000193342.5	896	175aa	Nonsense mediated decay	-	A0A0A6YX44	CDS 5' incomplete TSL:3
Rbm5-220	ENSMUST00000192130.5	784	150aa	Nonsense mediated decay	-	S4R184	TSL:3
Rbm5-219	ENSMUST00000183307.7	6321	No protein	Retained intron	-	-	TSL:1
Rbm5-215	ENSMUST00000182792.1	4718	No protein	Retained intron	-	-	TSL:NA
Rbm5-224	ENSMUST00000194801.5	2691	No protein	Retained intron	-	-	TSL:1
Rbm5-203	ENSMUST00000182007.7	1939	No protein	Retained intron	-	-	TSL:1
Rbm5-202	ENSMUST00000181993.1	1775	No protein	Retained intron	-	-	TSL:NA
Rbm5-213	ENSMUST00000182518.1	1272	No protein	Retained intron	-	-	TSL:1
Rbm5-206	ENSMUST00000182073.7	897	No protein	Retained intron	-	-	TSL:2
Rbm5-209	ENSMUST00000182332.1	793	No protein	Retained intron	-	-	TSL:2
Rbm5-207	ENSMUST00000182215.1	631	No protein	Retained intron	-	-	TSL:3
Rbm5-212	ENSMUST00000182422.6	600	No protein	Retained intron	-	-	TSL:2
Rbm5-217	ENSMUST00000182856.7	539	No protein	Retained intron	-	-	TSL:3
Rbm5-223	ENSMUST00000194682.1	502	No protein	Retained intron	-	-	TSL:3
Rbm5-216	ENSMUST00000182836.1	500	No protein	Retained intron	-	-	TSL:3

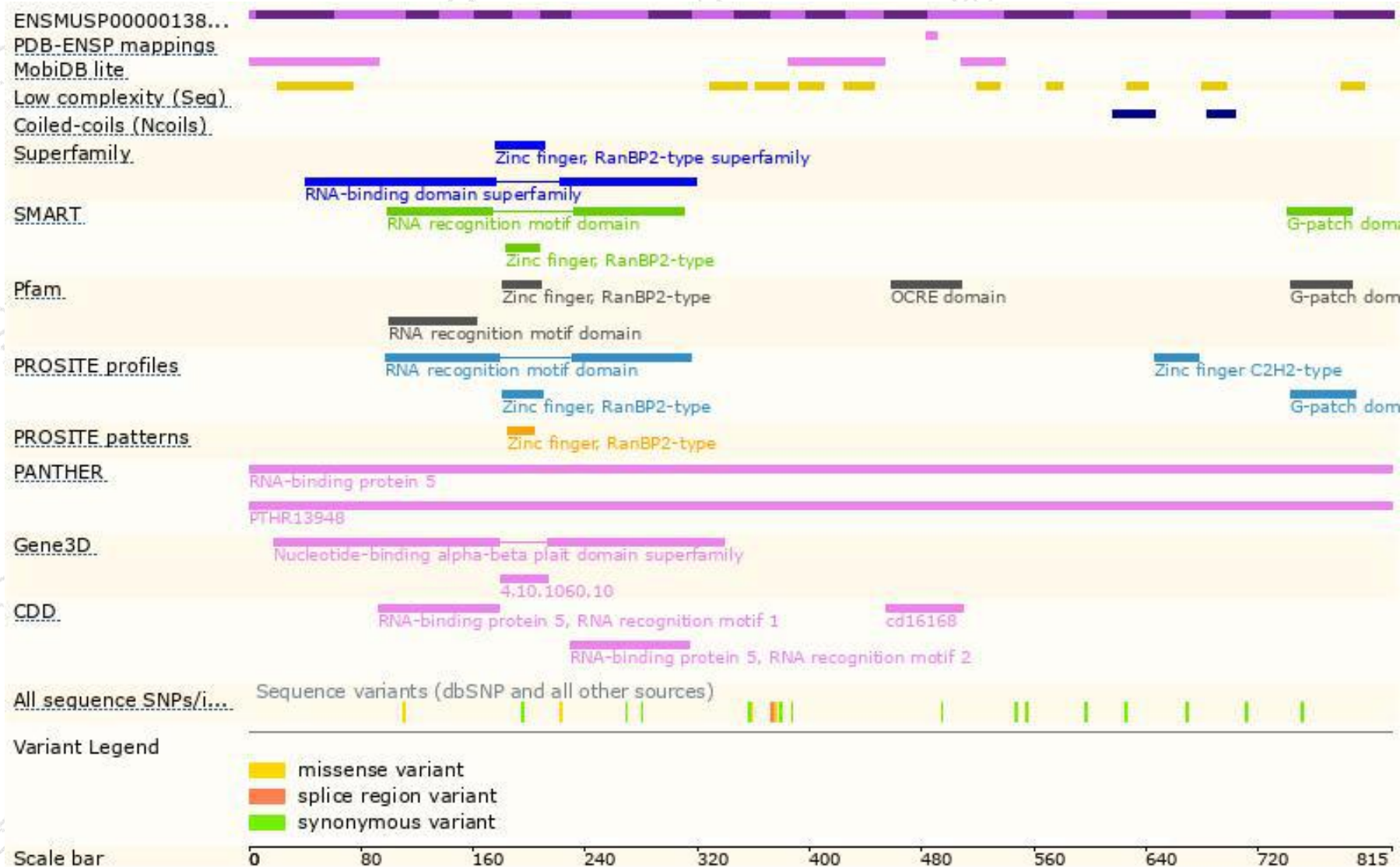
The strategy is based on the design of *Rbm5-214* transcript,the transcription is shown below:



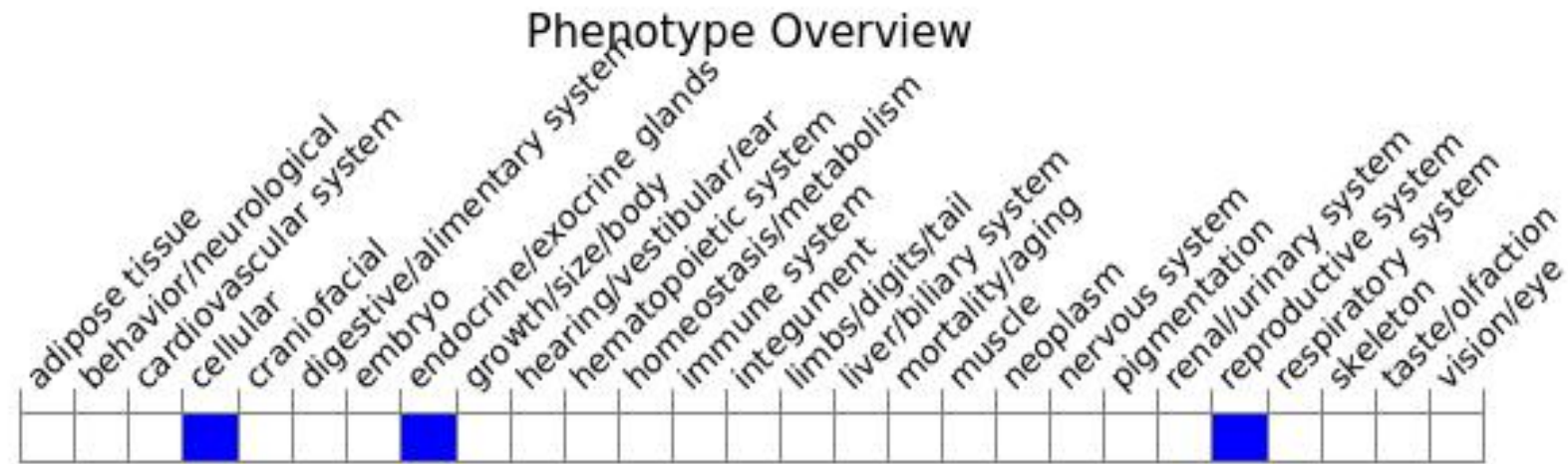
Genomic location distribution



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, mice homozygous for an ENU-induced allele exhibit male infertility with azoospermia, male germ cell apoptosis, round spermatid arrest and spermatid differentiation arrest.

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

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