Pex10 Cas9-KO Strategy

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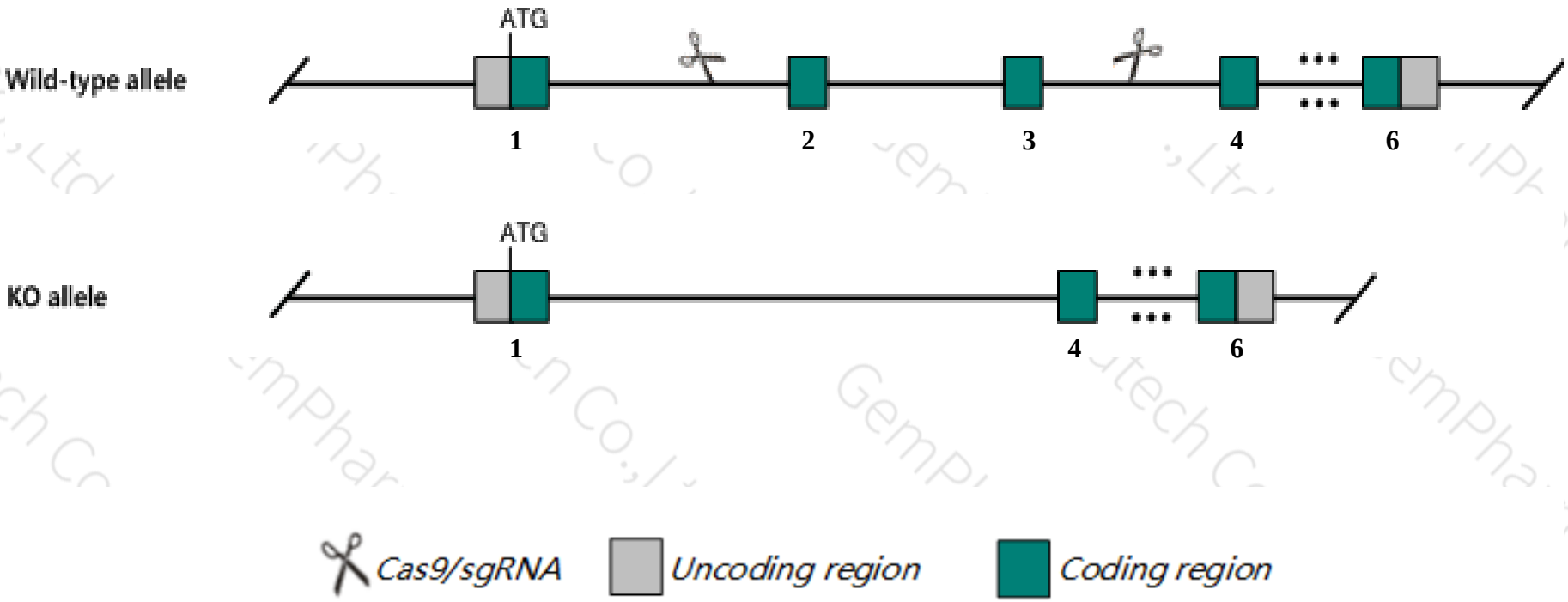
Design Date: 2020-11-23

Project Overview

Project Name	***Pex10***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Pex10* gene. The schematic diagram is as follows:



Technical routes

➢ The *Pex10* gene has 5 transcripts. According to the structure of *Pex10* gene, exon2-exon3 of *Pex10-201*(ENSMUST00000103180.3) transcript is recommended as the knockout region.The region contains 488bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Pex10* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

Notice

➢ According to the existing MGI data,mice homozygous for an ENU-induced allele exhibit partial neonatal mortality due to respiratory distress, loss of embryonic movement, and prenatal pathology including altered biochemistry, defects in axonal integrity, decreased Schwann cell number, and defects at the neuromuscular junction.

➢ The *Pex10* gene is located on the Chr4. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information（NCBI）

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Pex10 peroxisomal biogenesis factor 10 [Mus musculus (house mouse)]

Gene ID: 668173, updated on 13-Mar-2020

Summary

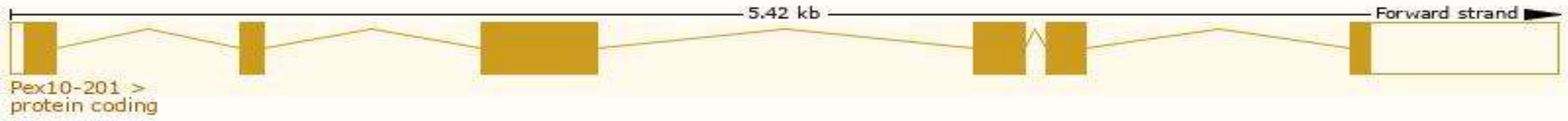
Official Symbol	Pex10 provided by MGI
Official Full Name	peroxisomal biogenesis factor 10 provided byMGI
Primary source	MGI:MGI:2684988
See related	Ensembl:ENSMUSG00000029047
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AV128229, Gm142
Expression	Ubiquitous expression in adrenal adult (RPKM 12.3), ovary adult (RPKM 11.7) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

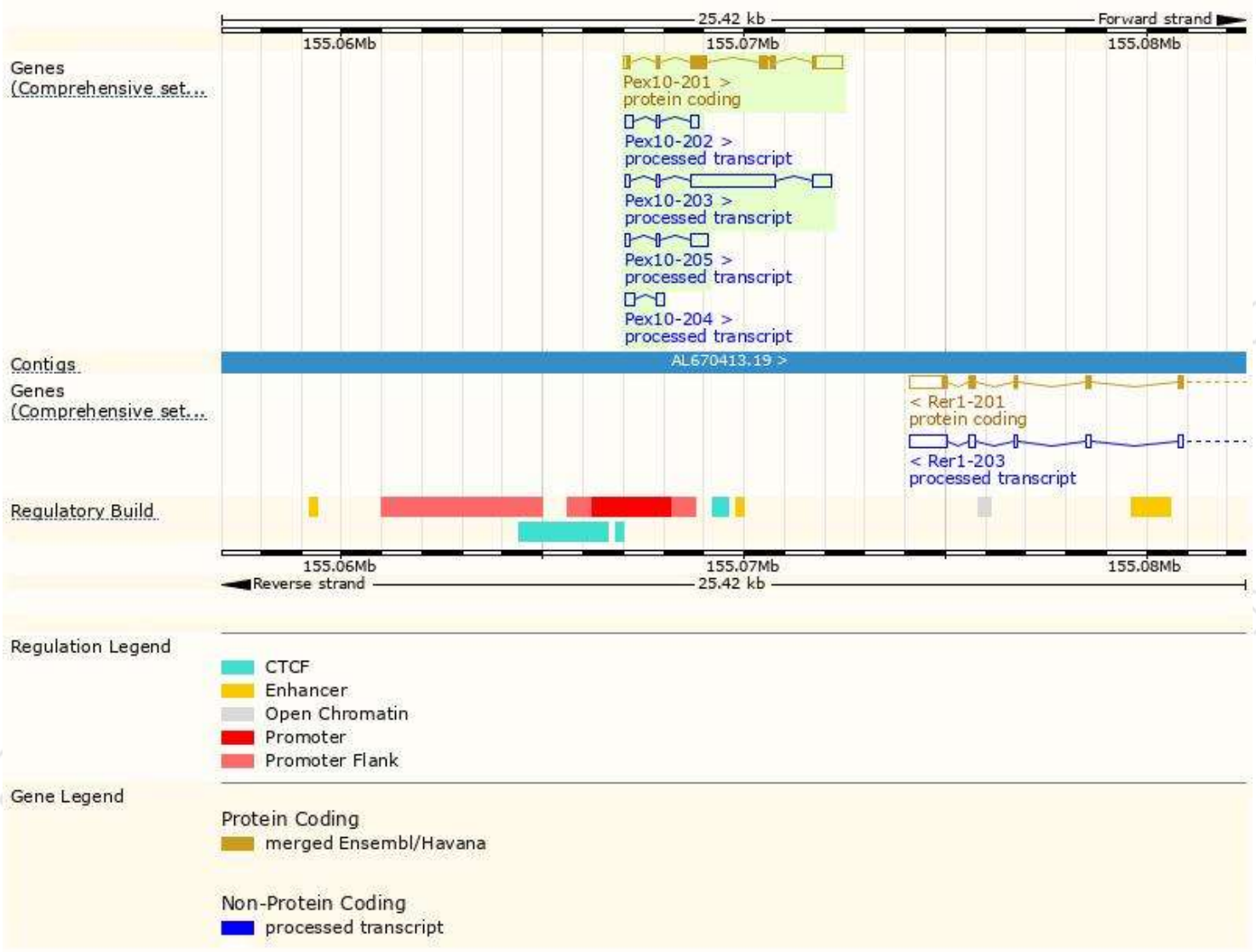
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Pex10-201	ENSMUST00000103180.3	1688	324aa	Protein coding	CCDS19021	B1AUE5	TSL:1 GENCODE basic APPRIS P1
Pex10-203	ENSMUST00000125432.7	2790	No protein	Processed transcript	-	-	TSL:1
Pex10-205	ENSMUST00000134341.1	651	No protein	Processed transcript	-	-	TSL:2
Pex10-202	ENSMUST00000123395.7	491	No protein	Processed transcript	-	-	TSL:3
Pex10-204	ENSMUST00000133116.1	412	No protein	Processed transcript	-	-	TSL:3

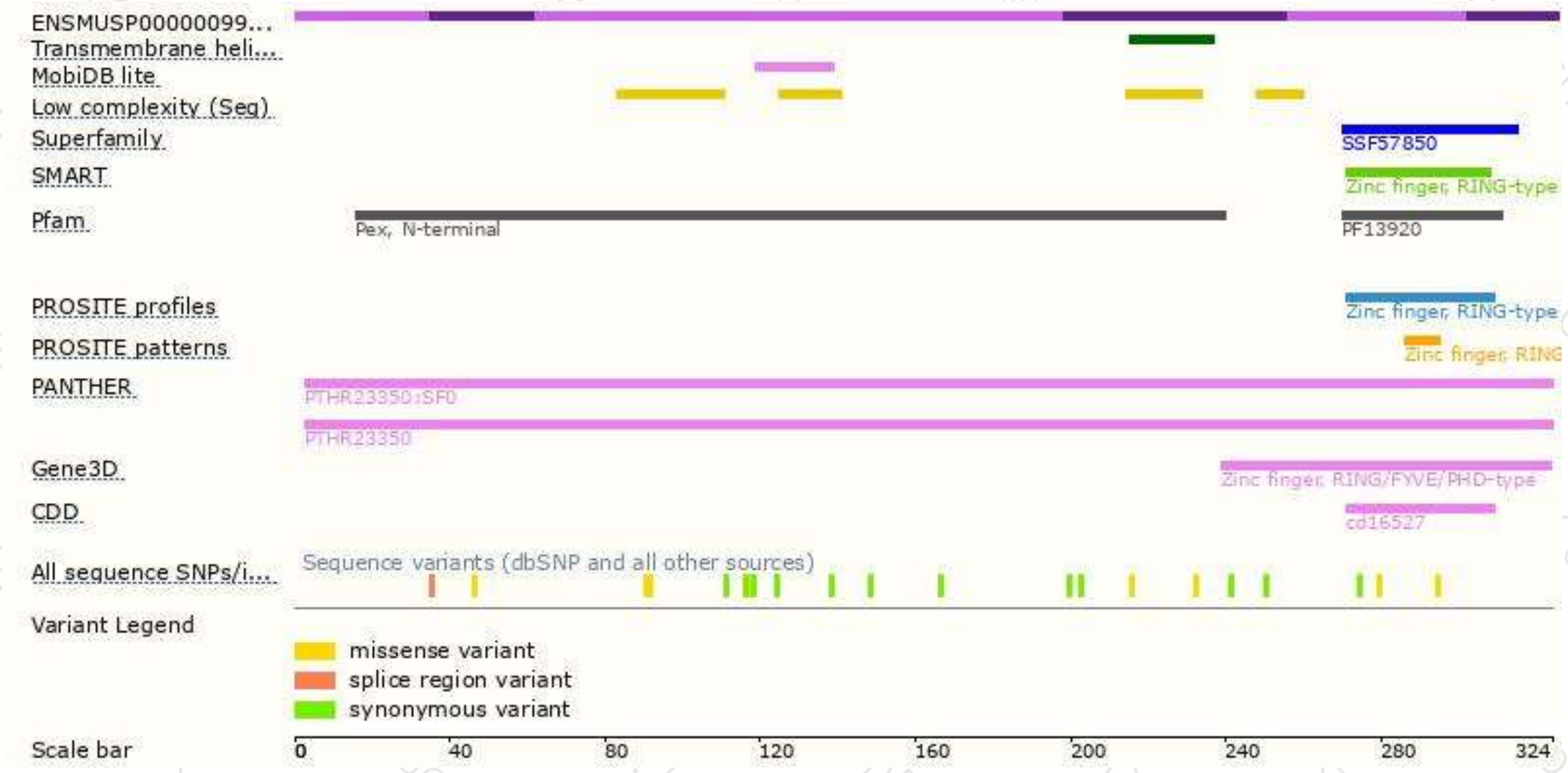
The strategy is based on the design of *Pex10-201* transcript,the transcription is shown below:



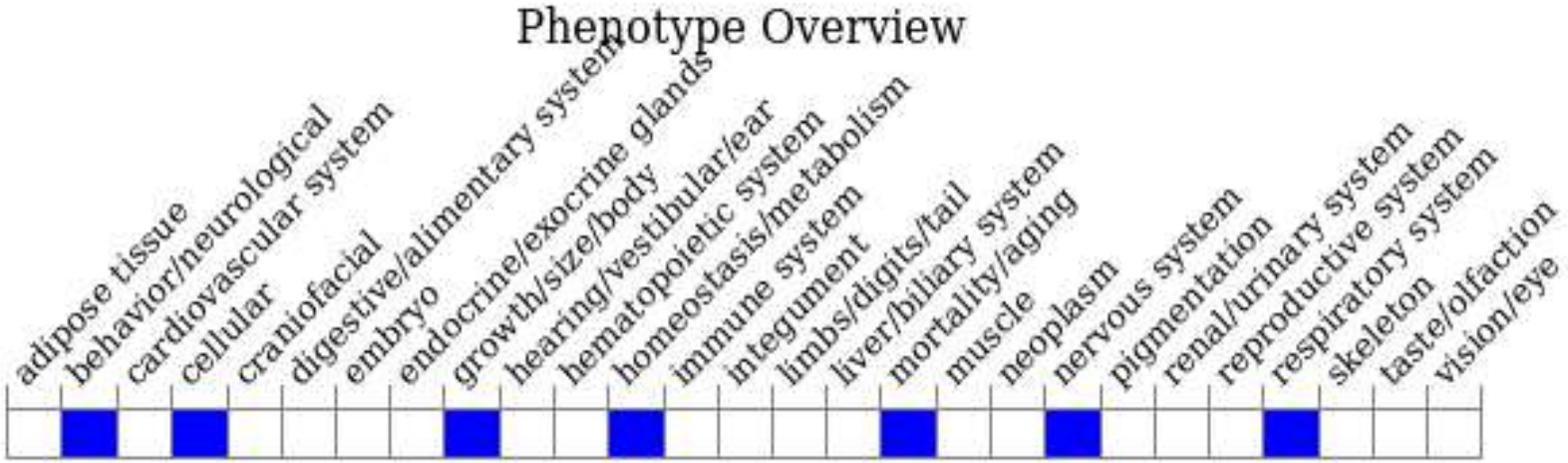
Genomic location distribution



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,mice homozygous for an ENU-induced allele exhibit partial neonatal mortality due to respiratory distress, loss of embryonic movement, and prenatal pathology including altered biochemistry, defects in axonal integrity, decreased Schwann cell number, and defects at the neuromuscular junction.

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

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