Gns Cas9-KO Strategy

Designer: Rui Xiong

Reviewer: Yumeng Wang

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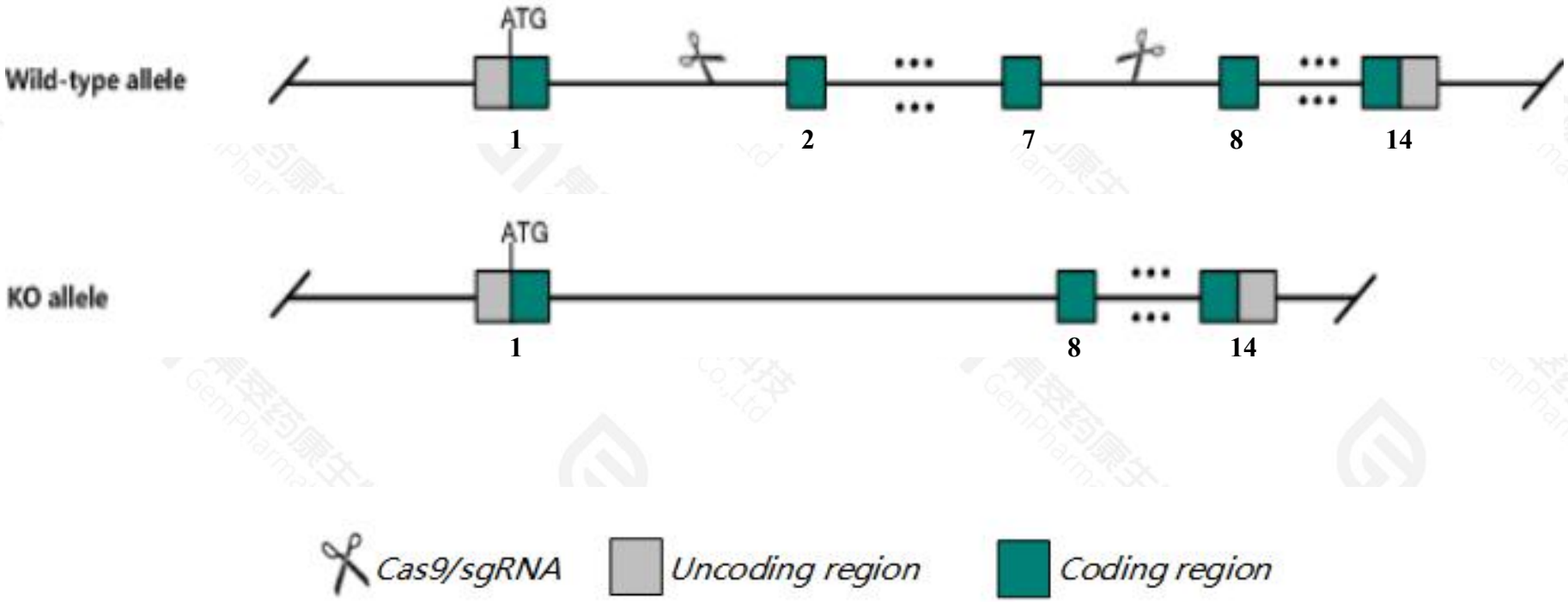
Project Overview

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

Knockout strategy

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



Technical routes

➢ The *Gns* gene has 10 transcripts. According to the structure of *Gns* gene, exon2-exon7 of *Gns-201*(ENSMUST00000040344.7) transcript is recommended as the knockout region.The region contains 683bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Gns* 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,homozygous KO results in progressive lysosomal glycosaminoglycan accumulation in the central nervous system and peripheral organs and causes hypoactivity and shortened lifespan.

➢ The *Gns* gene is located on the Chr10. 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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Gns glucosamine (N-acetyl)-6-sulfatase [Mus musculus (house mouse)]

Gene ID: 75612, updated on 25-Sep-2020

Summary

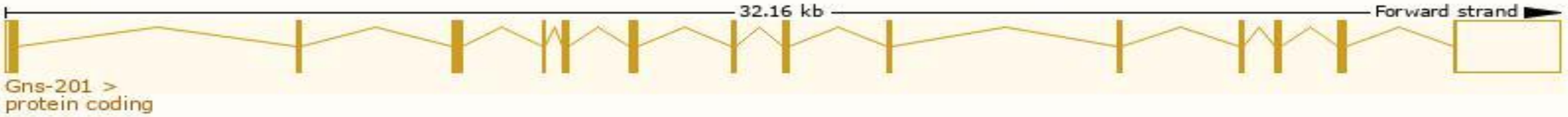
Official Symbol	Gns provided by MGI
Official Full Name	glucosamine (N-acetyl)-6-sulfatase provided byMGI
Primary source	MGI:MGI:1922862
See related	Ensembl:ENSMUSG00000034707
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	2610016K11Rik, AU042285, C87209, G6, G6S, N28088
Expression	Ubiquitous expression in adrenal adult (RPKM 66.2), kidney adult (RPKM 51.5) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

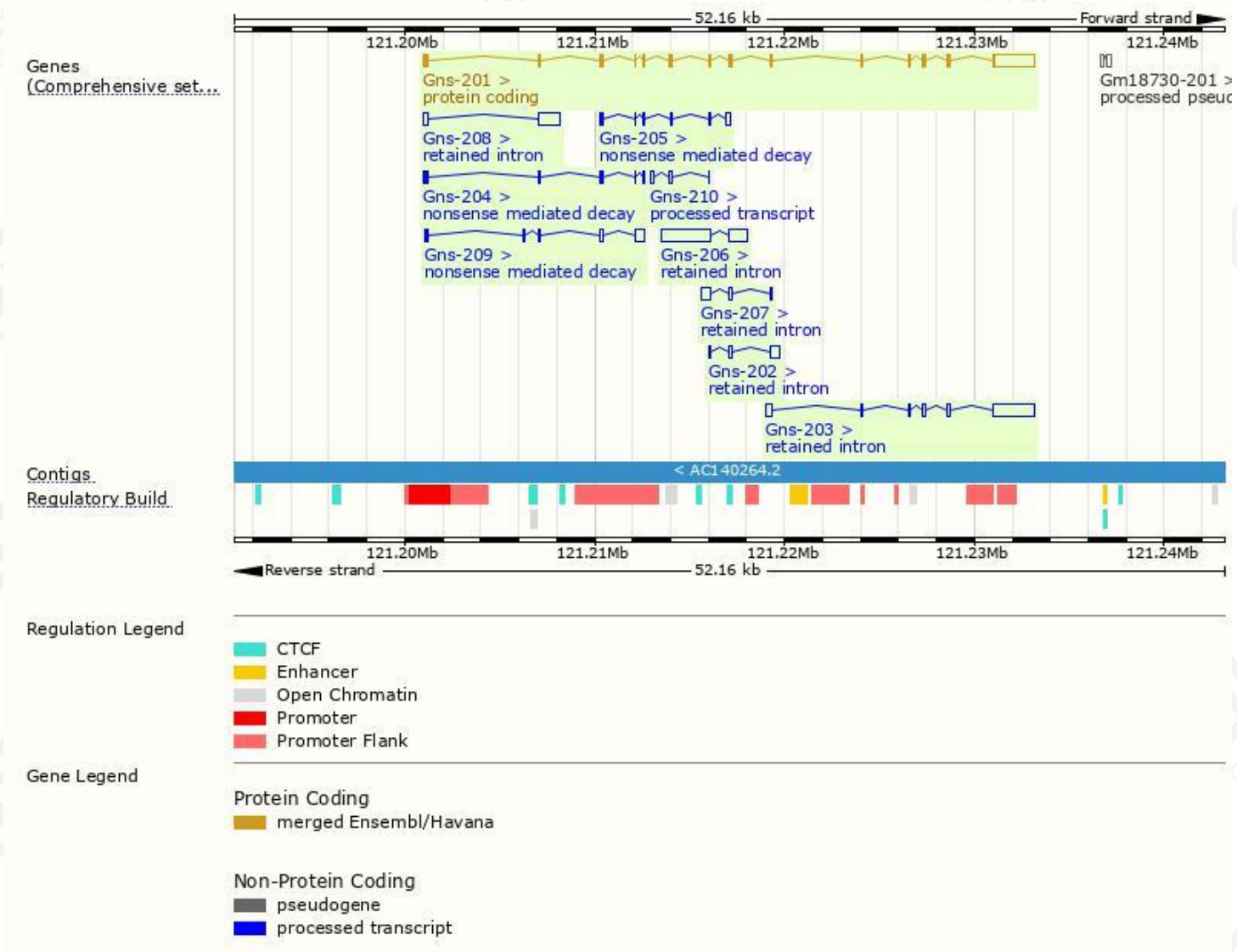
The gene has 10 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Gns-201	ENSMUST00000040344.7	3855	544aa	Protein coding	CCDS24210		TSL:1 , GENCODE basic , APPRIS P1 ,
Gns-209	ENSMUST00000219851.2	1056	99aa	Nonsense mediated decay	-		TSL:5 ,
Gns-205	ENSMUST00000219505.2	802	165aa	Nonsense mediated decay	-		CDS 5' incomplete , TSL:3 ,
Gns-204	ENSMUST00000219249.2	651	91aa	Nonsense mediated decay	-		TSL:3 ,
Gns-210	ENSMUST00000220350.2	373	No protein	Processed transcript	-		TSL:3 ,
Gns-206	ENSMUST00000219531.2	3501	No protein	Retained intron	-		TSL:1 ,
Gns-203	ENSMUST00000219216.2	3013	No protein	Retained intron	-		TSL:1 ,
Gns-208	ENSMUST00000219847.2	1358	No protein	Retained intron	-		TSL:1 ,
Gns-207	ENSMUST00000219580.2	698	No protein	Retained intron	-		TSL:3 ,
Gns-202	ENSMUST00000217712.2	672	No protein	Retained intron	-		TSL:3 ,

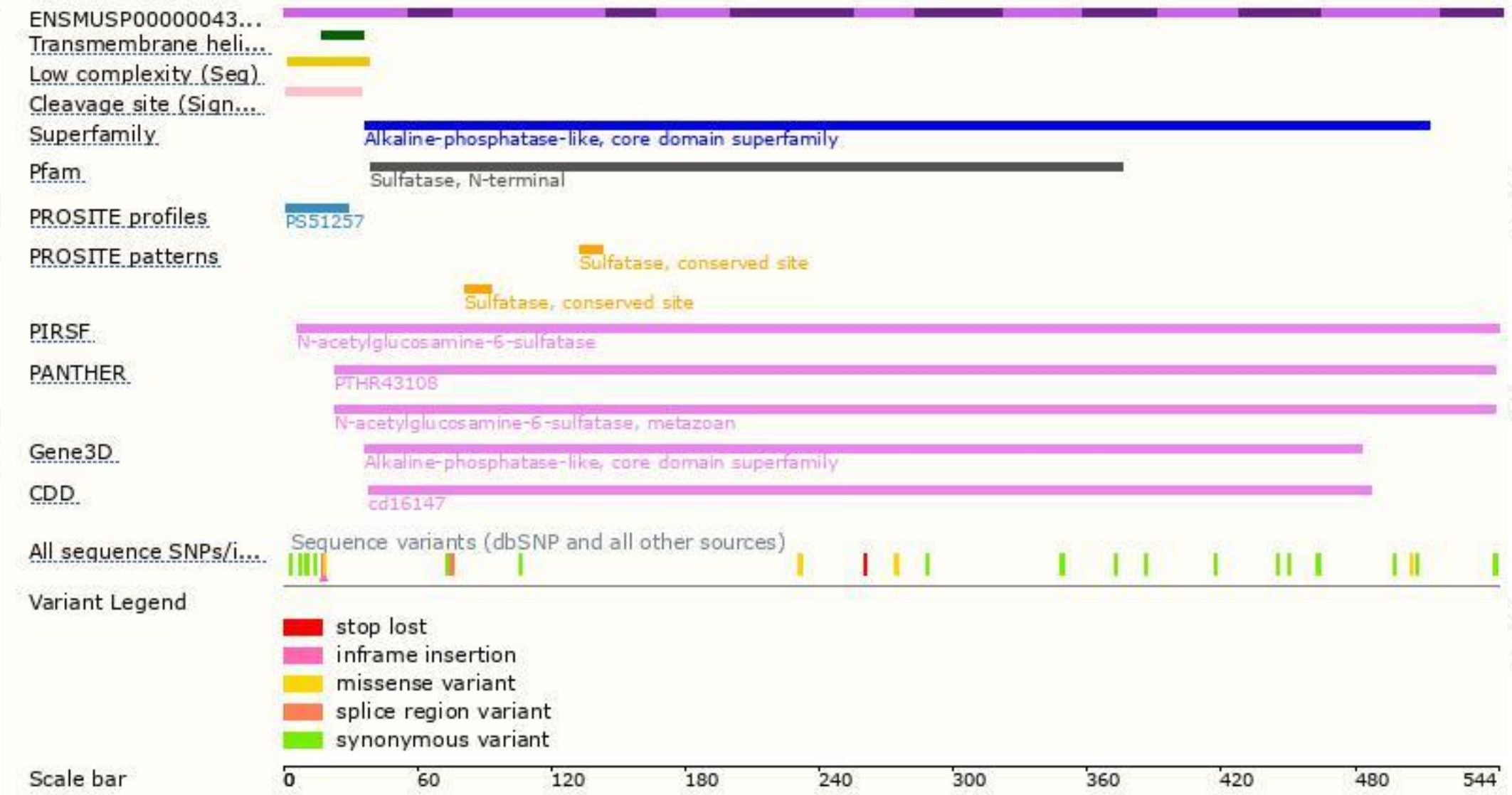
The strategy is based on the design of *Gns-201* transcript,the transcription is shown below:



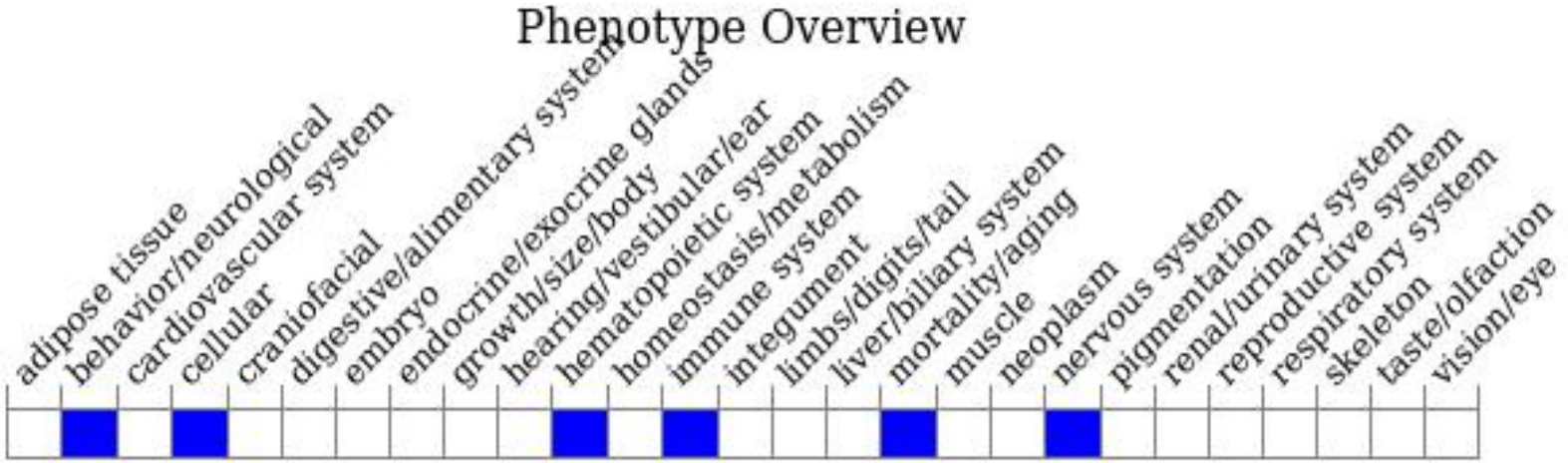
Genomic location distribution



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 KO results in progressive lysosomal glycosaminoglycan accumulation in the central nervous system and peripheral organs and causes hypoactivity and shortened lifespan.

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GemPharmatech

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

Tel: 025-5864 1534

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GemPharmatech Co.,Ltd