Sparc Cas9-CKO Strategy

Designer: Daohua Xu
Reviewer: Huimin Su

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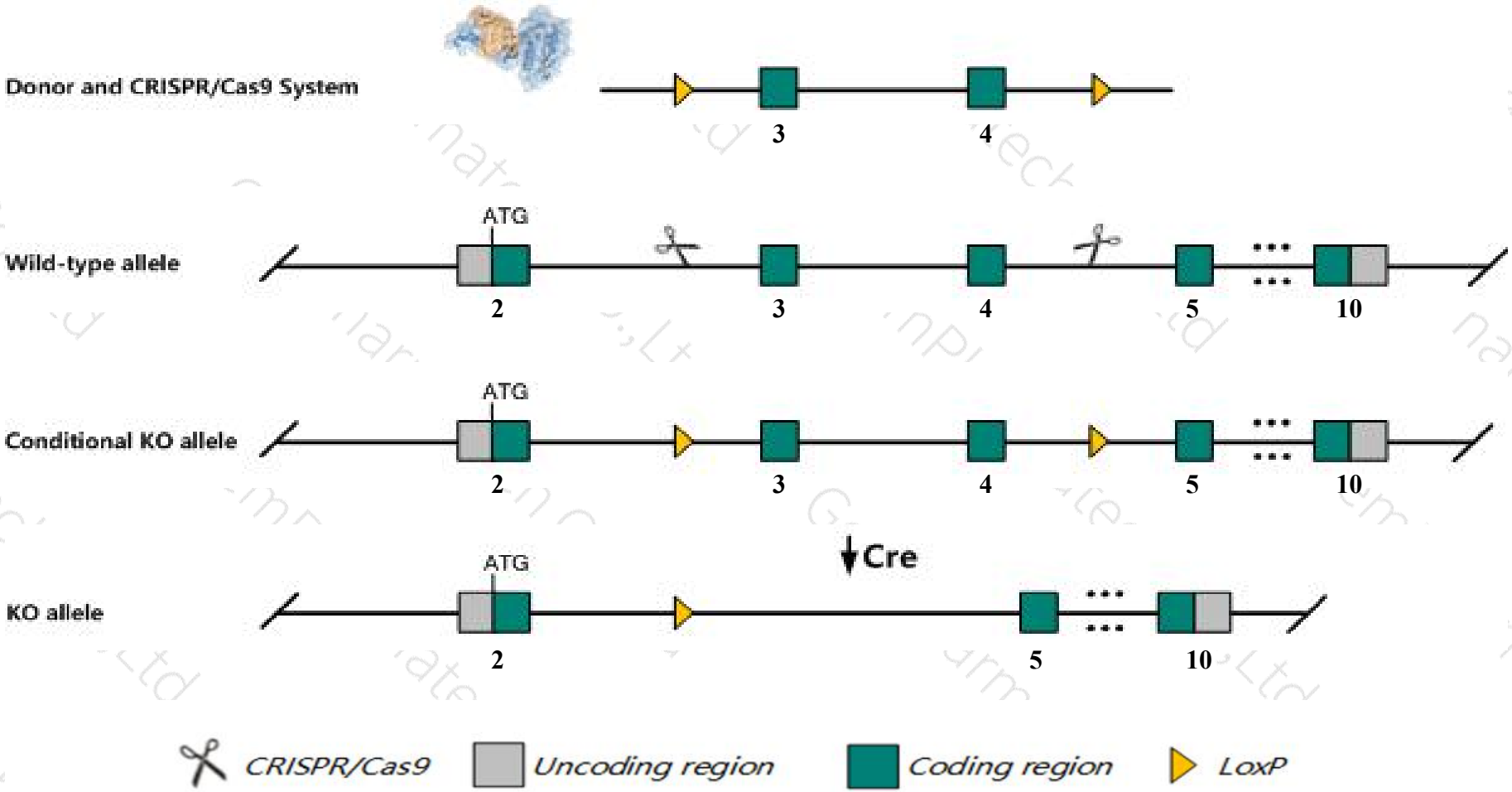
Project Overview

Project Name	***Sparc***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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



Technical routes

- The *Sparc* gene has 9 transcripts. According to the structure of *Sparc* gene, exon3-exon4 of *Sparc-201* (ENSMUST00000018737.12) transcript is recommended as the knockout region.The region contains 148bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Sparc* 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.

Notice

➢ According to the existing MGI data,Homozygotes for targeted null mutations exhibit cataracts, reduced skin collagen content, accelerated wound closure, osteopenia associated with reduced bone remodeling, and increased growth of implanted tumors.

➢ The *Sparc* gene is located on the Chr11. 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）

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Sparc secreted acidic cysteine rich glycoprotein [Mus musculus (house mouse)]

Gene ID: 20692, updated on 9-Apr-2019

Summary

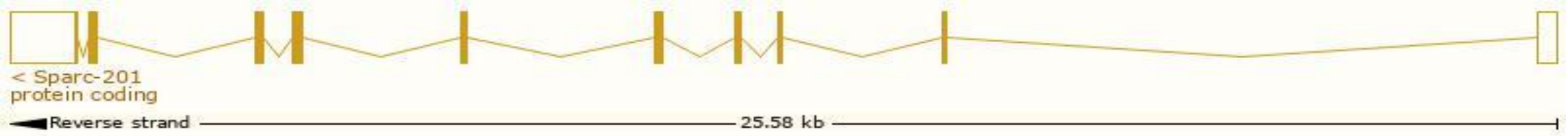
Official Symbol	Sparc provided by MGI
Official Full Name	secreted acidic cysteine rich glycoprotein provided by MGI
Primary source	MGI:MGI:98373
See related	Ensembl:ENSMUSG00000018593
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	BM-40, ON
Expression	Broad expression in subcutaneous fat pad adult (RPKM 1043.9), bladder adult (RPKM 893.4) and 22 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

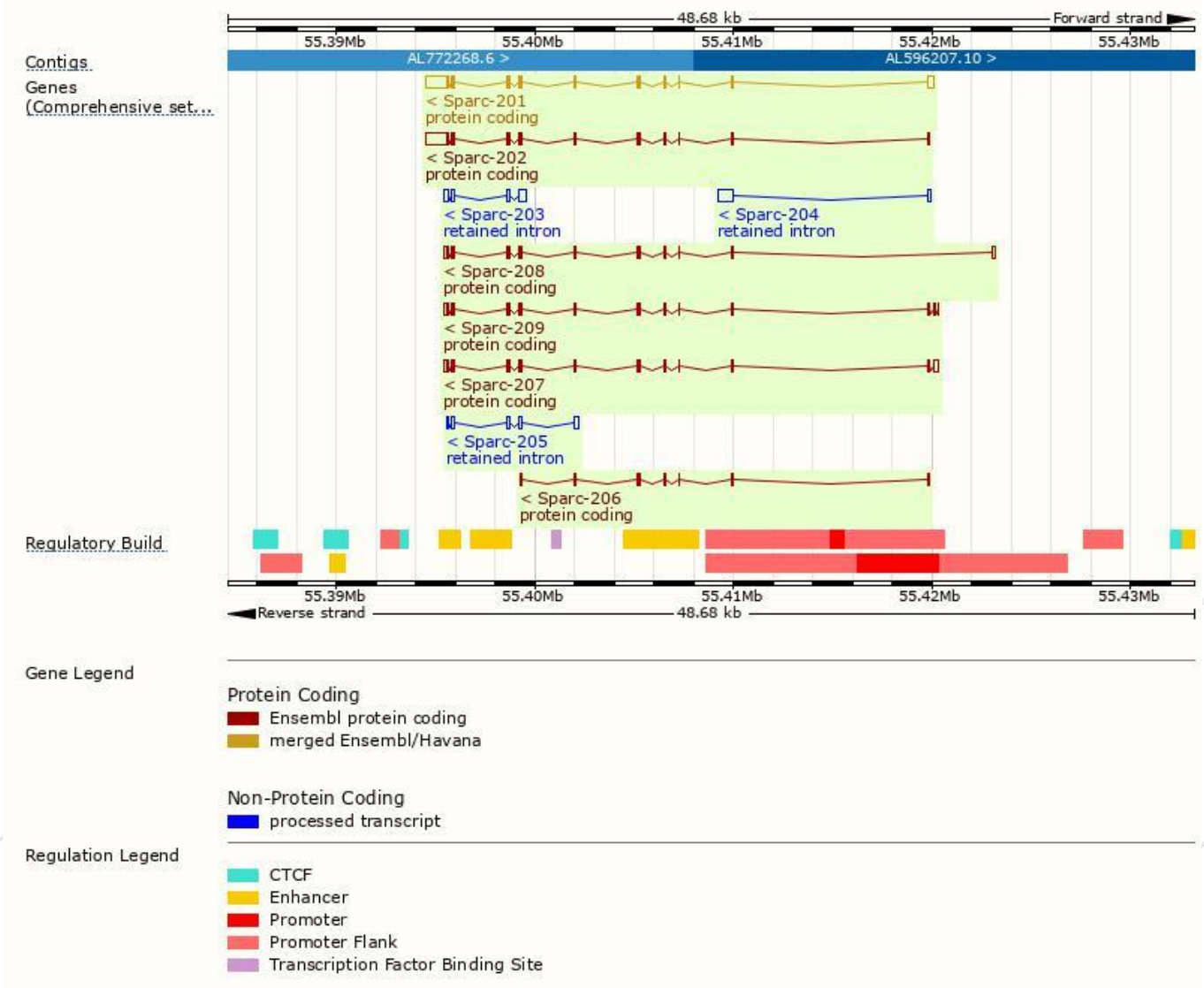
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sparc-201	ENSMUST00000018737.12	2300	302aa	Protein coding	CCDS24712	P07214 Q5NCU5	TSL:1 GENCODE basic APPRIS P3
Sparc-202	ENSMUST00000108858.7	2115	301aa	Protein coding	CCDS70189	Q5NCU4	TSL:1 GENCODE basic APPRIS ALT2
Sparc-208	ENSMUST00000214685.1	1269	302aa	Protein coding	CCDS24712	P07214 Q5NCU5	TSL:5 GENCODE basic APPRIS P3
Sparc-207	ENSMUST00000213866.1	1366	332aa	Protein coding	-	A0A1L1SSH9	TSL:5 GENCODE basic APPRIS ALT2
Sparc-209	ENSMUST00000216313.1	1304	332aa	Protein coding	-	A0A1L1SSH9	TSL:5 GENCODE basic APPRIS ALT2
Sparc-206	ENSMUST00000141530.1	640	172aa	Protein coding	-	Q5NCU3	CDS 3' incomplete TSL:5
Sparc-203	ENSMUST00000123775.7	909	No protein	Retained intron	-	-	TSL:2
Sparc-204	ENSMUST00000125787.1	907	No protein	Retained intron	-	-	TSL:2
Sparc-205	ENSMUST00000130642.1	741	No protein	Retained intron	-	-	TSL:1

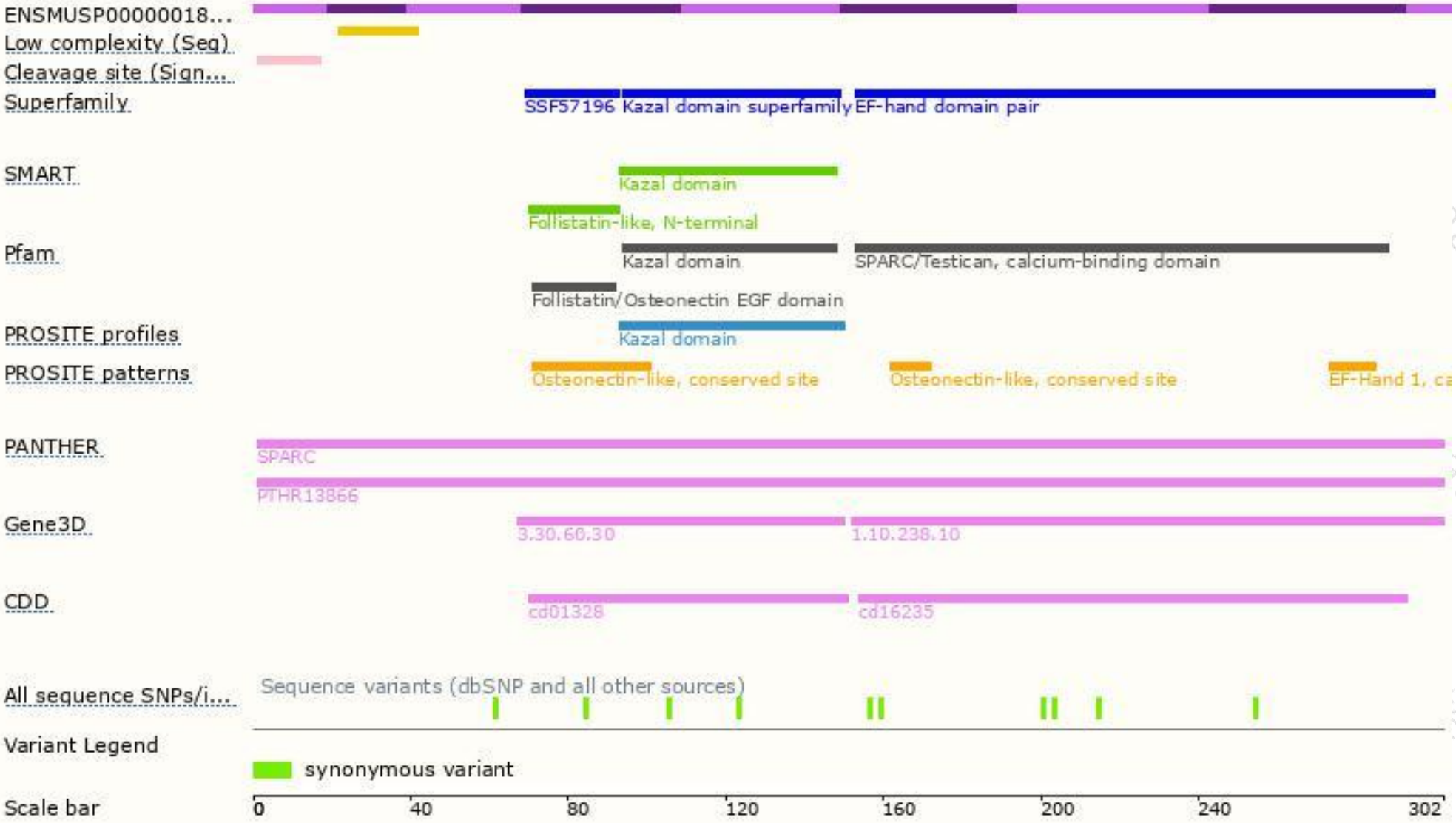
The strategy is based on the design of *Sparc-201* transcript,The transcription is shown below



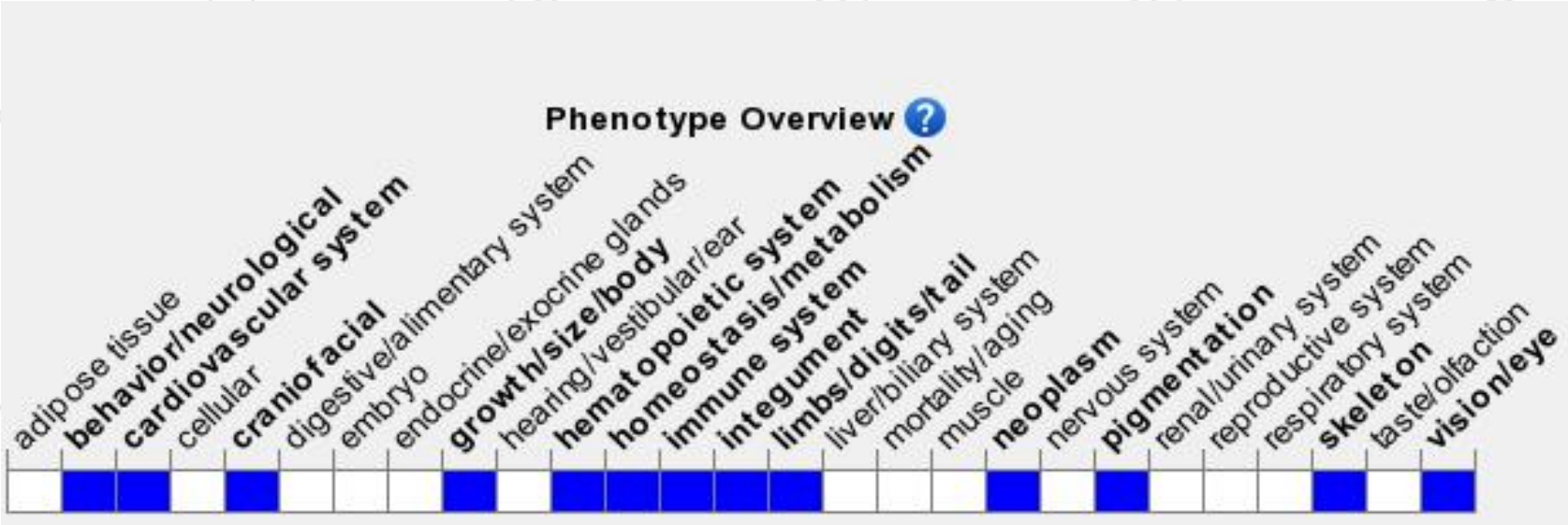
Genomic location distribution



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/).

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

Tel: 400-9660890

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GemPharmatech Co.,Ltd