Rpgrip1 Cas9-CKO Strategy

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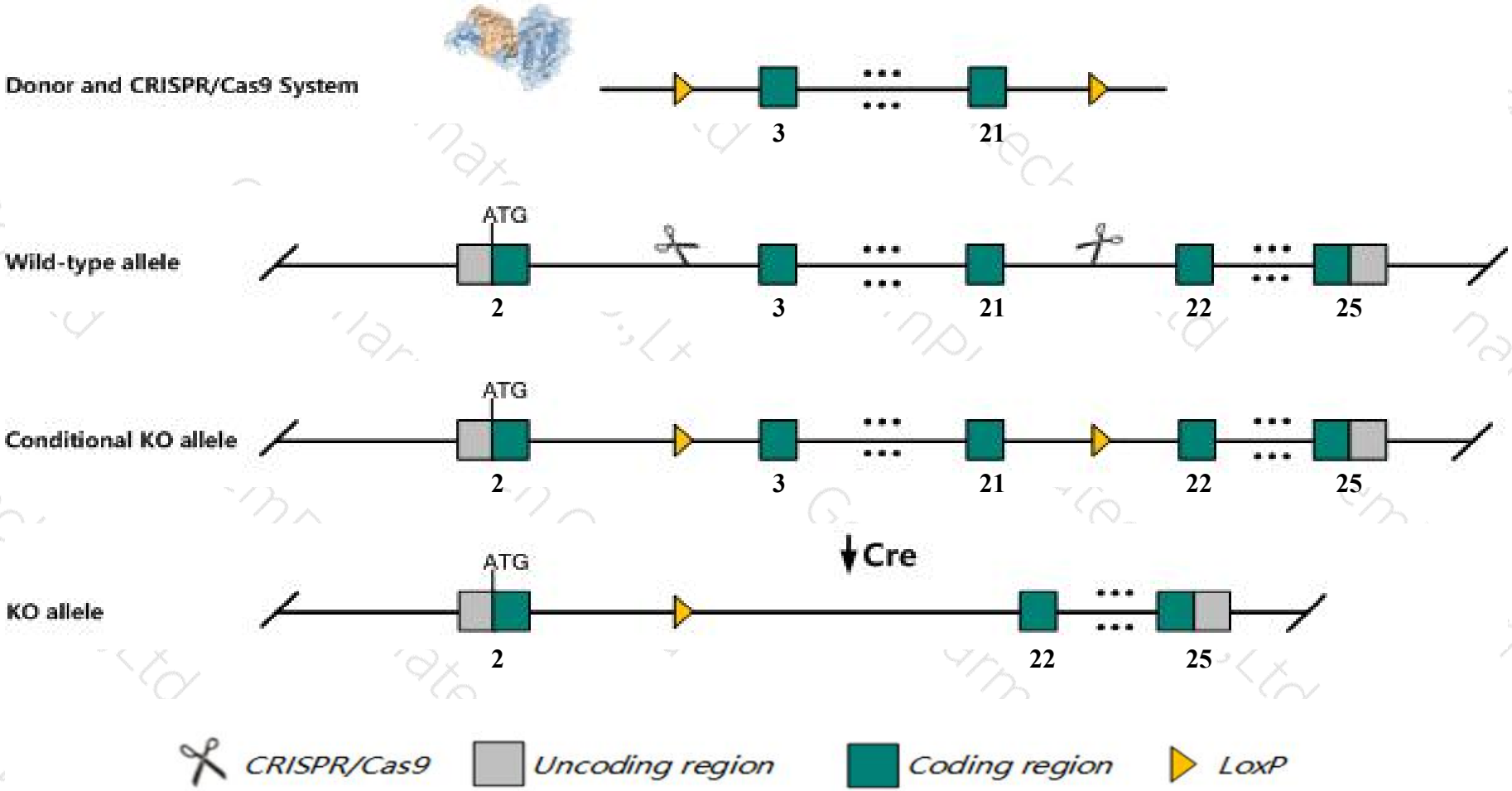
Design Date: 2020-1-20

Project Overview

Project Name	***Rpgrip1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rpgrip1* gene. The schematic diagram is as follows:



Technical routes

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- The *Rpgrip1* gene has 19 transcripts. According to the structure of *Rpgrip1* gene, exon3-exon21 of *Rpgrip1-202* (ENSMUST00000111603.9) transcript is recommended as the knockout region.The region contains 3401bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Rpgrip1* 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,Homozygous mutation of this gene results in photoreceptor cell dysmorphology. By 3 months of age mutant animals show near complete loss of photoreceptor cells.

- The *Rpgrip1* gene is located on the Chr14. 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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Rpgrip1 retinitis pigmentosa GTPase regulator interacting protein 1 [Mus musculus (house mouse)]

Gene ID: 77945, updated on 19-Mar-2019

Summary

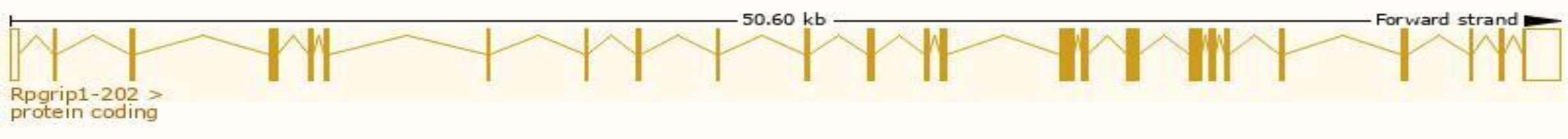
Official Symbol	Rpgrip1 provided by MGI
Official Full Name	retinitis pigmentosa GTPase regulator interacting protein 1 provided by MGI
Primary source	MGI:MGI:1932134
See related	Ensembl:ENSMUSG00000057132
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	0610005A07Rik, 4930401L23Rik, 4930505G06Rik, A930002K18Rik, AA415034, nmf247
Expression	Broad expression in testis adult (RPKM 34.8), CNS E11.5 (RPKM 6.0) and 17 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

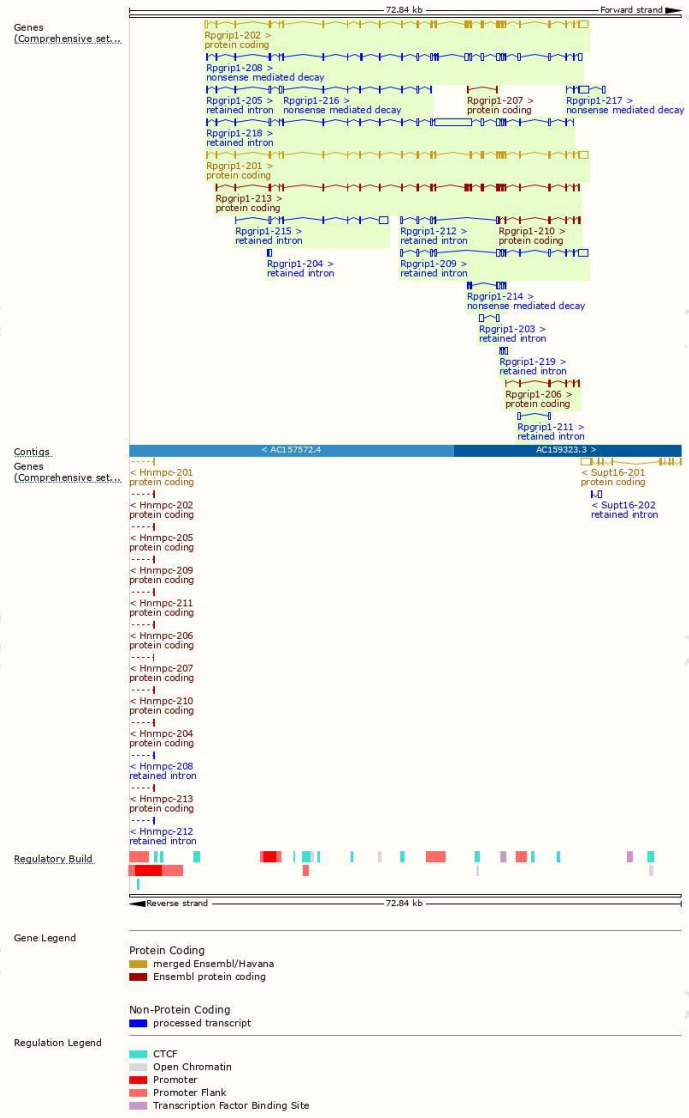
The gene has 19 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rpgrip1-202	ENSMUST00000111603.9	5380	1331aa	Protein coding	CCDS36918	Q9EPQ2	TSL:1 GENCODE basic APPRIS P2
Rpgrip1-201	ENSMUST00000111600.10	4565	1127aa	Protein coding	CCDS49490	E9QQ97	TSL:1 GENCODE basic
Rpgrip1-213	ENSMUST00000181401.7	3963	1320aa	Protein coding	-	E7CHD7	TSL:1 GENCODE basic APPRIS ALT2
Rpgrip1-210	ENSMUST00000181017.7	966	295aa	Protein coding	-	M0QWM1	CDS 5' incomplete TSL:2
Rpgrip1-206	ENSMUST00000180500.7	602	201aa	Protein coding	-	M0QW58	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:3
Rpgrip1-207	ENSMUST00000180513.1	213	71aa	Protein coding	-	M0QWT5	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Rpgrip1-208	ENSMUST00000180646.7	4557	289aa	Nonsense mediated decay	-	M0QWC0	TSL:1
Rpgrip1-217	ENSMUST00000181736.1	1733	82aa	Nonsense mediated decay	-	M0QWL3	CDS 5' incomplete TSL:5
Rpgrip1-214	ENSMUST00000181627.7	970	91aa	Nonsense mediated decay	-	M0QWI1	CDS 5' incomplete TSL:5
Rpgrip1-216	ENSMUST00000181709.1	757	191aa	Nonsense mediated decay	-	M0QW56	CDS 5' incomplete TSL:5
Rpgrip1-218	ENSMUST00000181823.8	7878	No protein	Retained intron	-	-	TSL:5
Rpgrip1-209	ENSMUST00000180901.7	3212	No protein	Retained intron	-	-	TSL:1
Rpgrip1-215	ENSMUST00000181673.2	1826	No protein	Retained intron	-	-	TSL:5
Rpgrip1-205	ENSMUST00000180484.7	945	No protein	Retained intron	-	-	TSL:1
Rpgrip1-212	ENSMUST00000181297.7	902	No protein	Retained intron	-	-	TSL:1
Rpgrip1-203	ENSMUST00000180388.1	834	No protein	Retained intron	-	-	TSL:3
Rpgrip1-219	ENSMUST00000181862.1	585	No protein	Retained intron	-	-	TSL:3
Rpgrip1-204	ENSMUST00000180433.1	465	No protein	Retained intron	-	-	TSL:3
Rpgrip1-211	ENSMUST00000181208.1	415	No protein	Retained intron	-	-	TSL:3

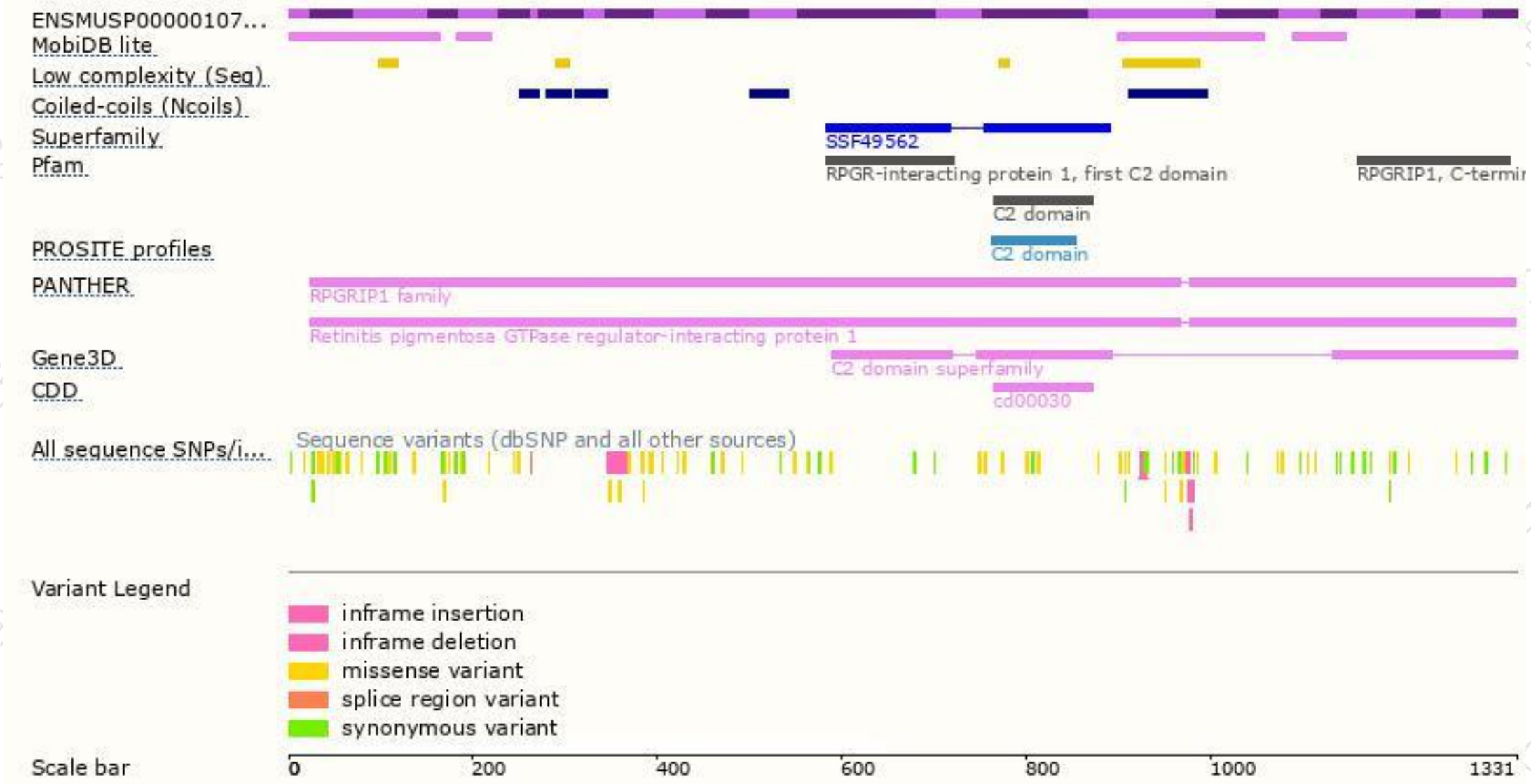
The strategy is based on the design of *Rpgrip1-202* transcript,The transcription is shown below



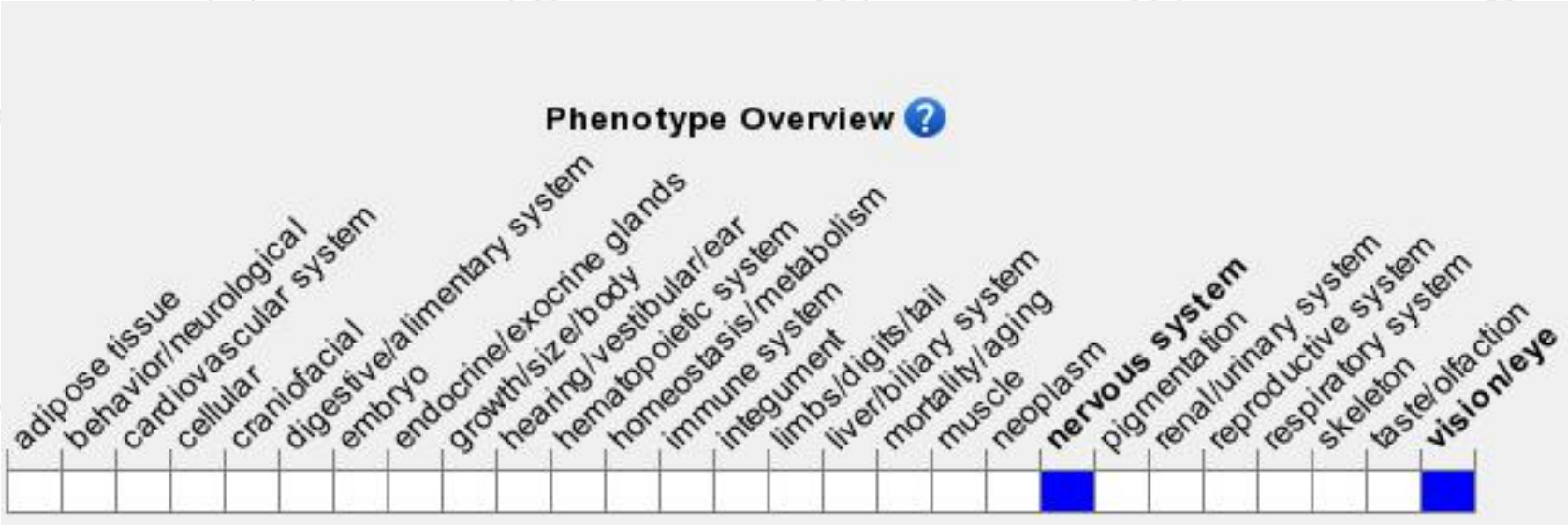
Genomic location distribution



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 photoreceptor cell dysmorphology. By 3 months of age mutant animals show near complete loss of photoreceptor cells.

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

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