

Rasgrp4 Cas9-CKO Strategy

Designer:

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Project Overview

Project Name

Rasgrp4

Project type

Cas9-CKO

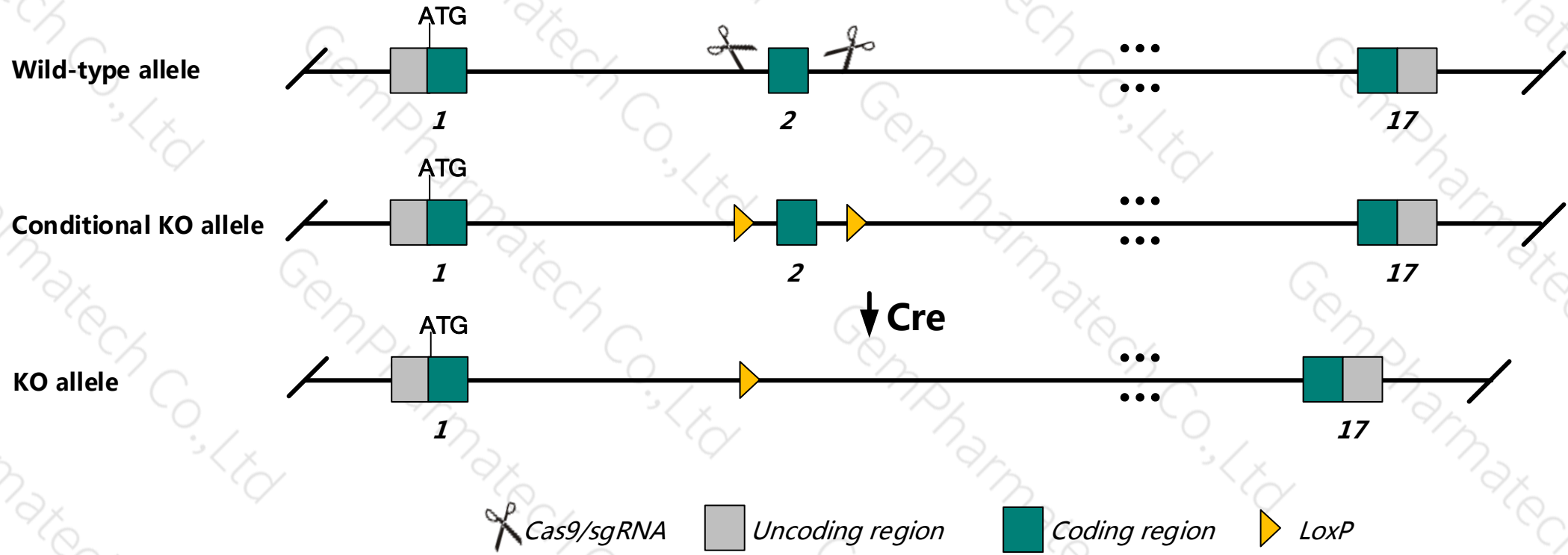
Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rasgrp4* gene. The schematic diagram is as follows:

Donor and CRISPR/Cas9 System



- The *Rasgrp4* gene has 14 transcripts. According to the structure of *Rasgrp4* gene, exon2 of *Rasgrp4*-205 transcript is recommended as the knockout region. The region contains 185bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rasgrp4* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 was knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues or cell types.

- The *Rasgrp4* gene is located on the Chr7. 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 gene transcription and translation processes, all risks cannot be predicted under existing information.

Gene information (NCBI)

Rasgrp4 RAS guanyl releasing protein 4 [*Mus musculus* (house mouse)]

Gene ID: 233046, updated on 31-Jan-2019

Summary

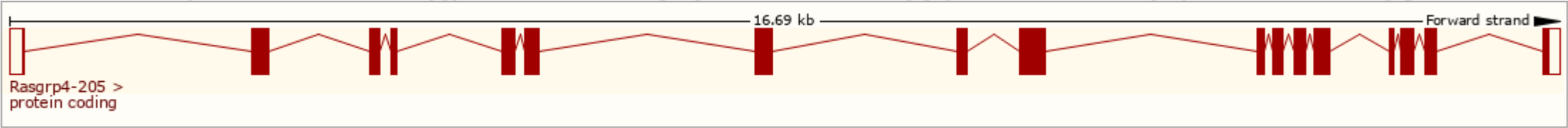
Official Symbol	Rasgrp4 provided by MGI
Official Full Name	RAS guanyl releasing protein 4 provided by MGI
Primary source	MGI:MGI:2386851
See related	Ensembl:ENSMUSG00000030589
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Expression	Ubiquitous expression in thymus adult (RPKM 8.9), duodenum adult (RPKM 8.4) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

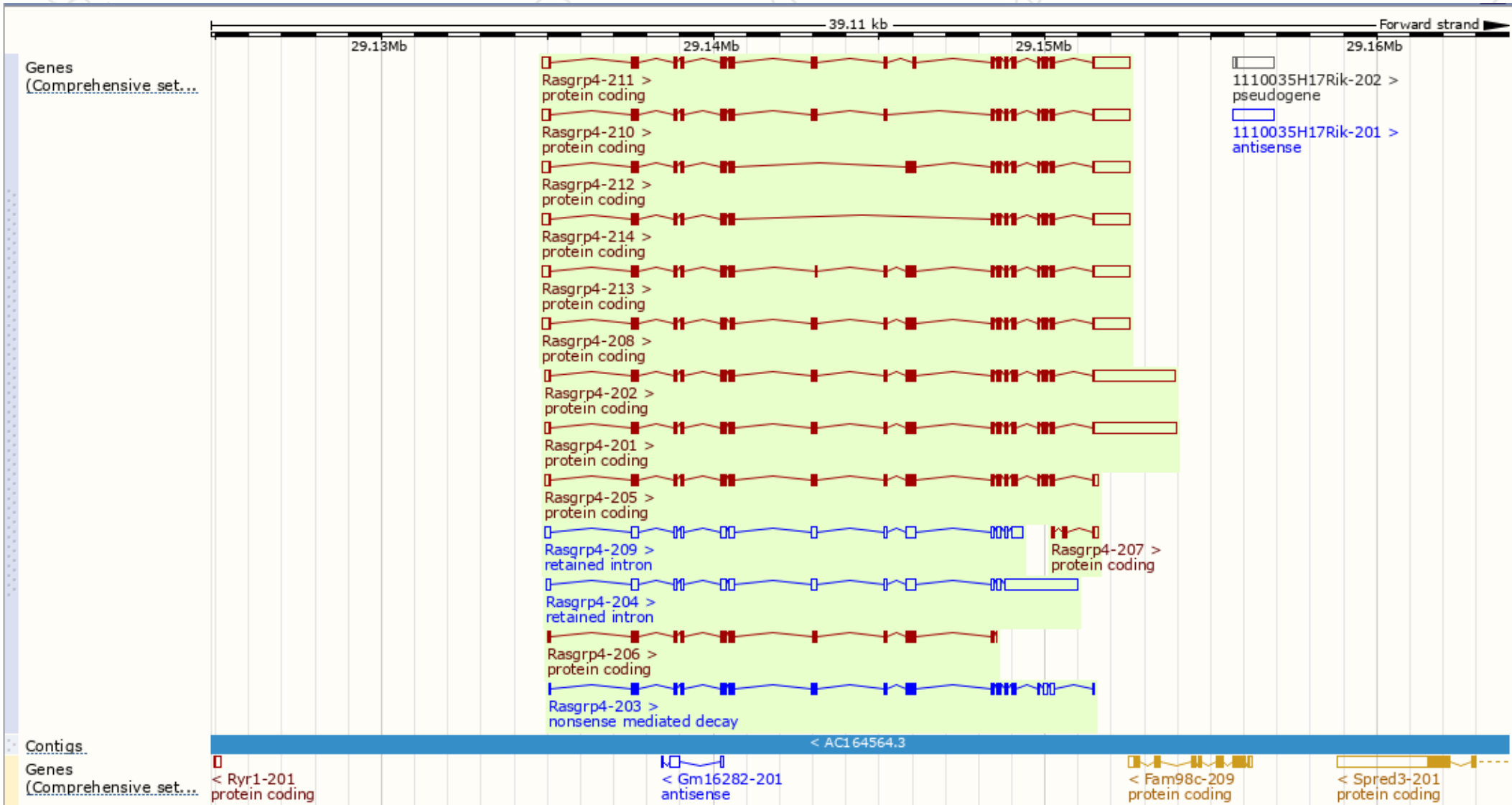
The gene has 14 transcripts, and all transcripts are shown below :

Show/hide columns (1 hidden) Filter							
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rasgrp4-201	ENSMUST00000032811.11	4621	673aa	Protein coding	CCDS85257	Q8BTM9	TSL:1 GENCODE basic APPRIS P1
Rasgrp4-205	ENSMUST00000159975.7	2296	678aa	Protein coding	CCDS85258	Q8BTM9	TSL:1 GENCODE basic
Rasgrp4-202	ENSMUST00000094617.10	4617	679aa	Protein coding	-	E9QKB7	TSL:5 GENCODE basic
Rasgrp4-208	ENSMUST00000161522.7	3242	659aa	Protein coding	-	E0CZ06	TSL:5 GENCODE basic
Rasgrp4-213	ENSMUST00000204845.2	3182	639aa	Protein coding	-	A0A0N4SUQ3	TSL:5 GENCODE basic
Rasgrp4-211	ENSMUST00000203380.2	3077	604aa	Protein coding	-	A0A0N4SUN7	TSL:5 GENCODE basic
Rasgrp4-210	ENSMUST00000203070.2	3008	581aa	Protein coding	-	A0A0N4SW33	TSL:5 GENCODE basic
Rasgrp4-212	ENSMUST00000204194.2	2993	576aa	Protein coding	-	A0A0N4SVV6	TSL:5 GENCODE basic
Rasgrp4-214	ENSMUST00000205027.2	2717	484aa	Protein coding	-	A0A0N4SVP1	TSL:5 GENCODE basic
Rasgrp4-206	ENSMUST00000160194.7	1356	429aa	Protein coding	-	E0CX54	CDS 3' incomplete TSL:5
Rasgrp4-207	ENSMUST00000160396.1	375	85aa	Protein coding	-	F7BK25	CDS 5' incomplete TSL:5
Rasgrp4-203	ENSMUST00000159351.2	1997	578aa	Nonsense mediated decay	-	Q8BTM9	TSL:1
Rasgrp4-204	ENSMUST00000159898.2	3712	No protein	Retained intron	-	-	TSL:1
Rasgrp4-209	ENSMUST00000162946.7	2005	No protein	Retained intron	-	-	TSL:1

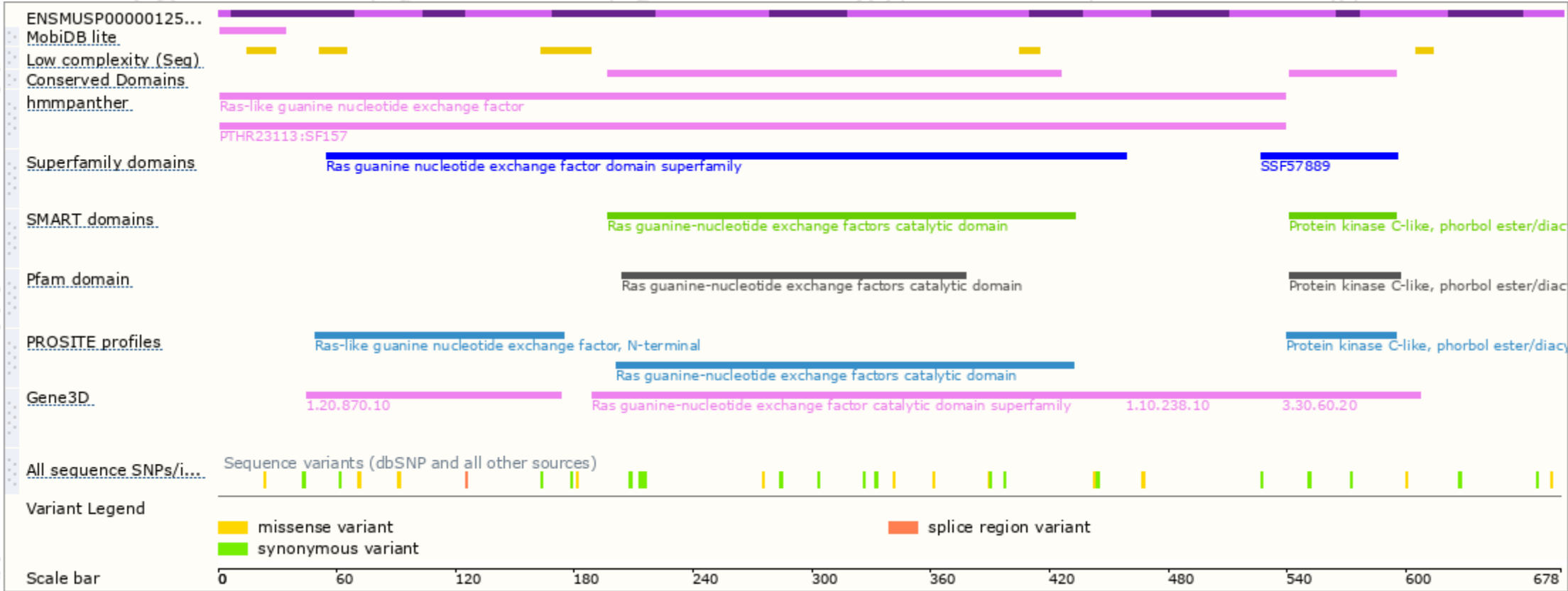
The strategy is based on the design of *Rasgrp4-205* transcript, The transcription is shown below



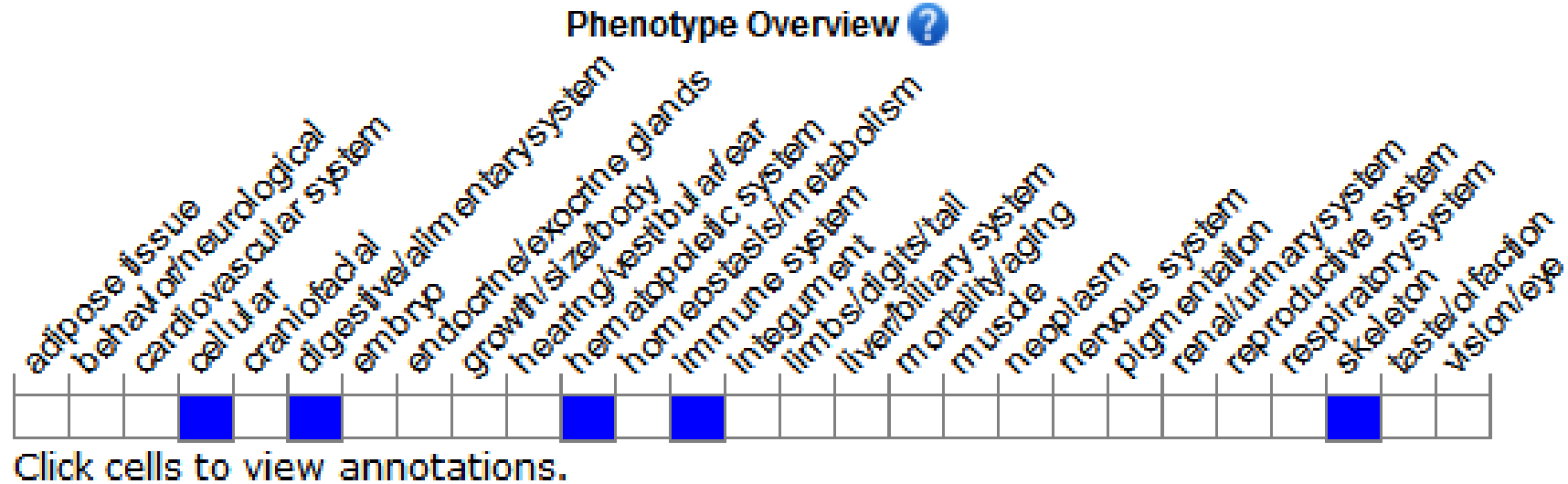
Genomic location distribution



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 a knock-out allele exhibit impaired neutrophil reactive oxygen species production and chemotaxis in vitro. Mice homozygous for another knock-out allele exhibit decreased susceptibility to induced colitis and arthritis.

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