

Ica1 Cas9-CKO Strategy

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Project Overview

Project Name

Ica1

Project type

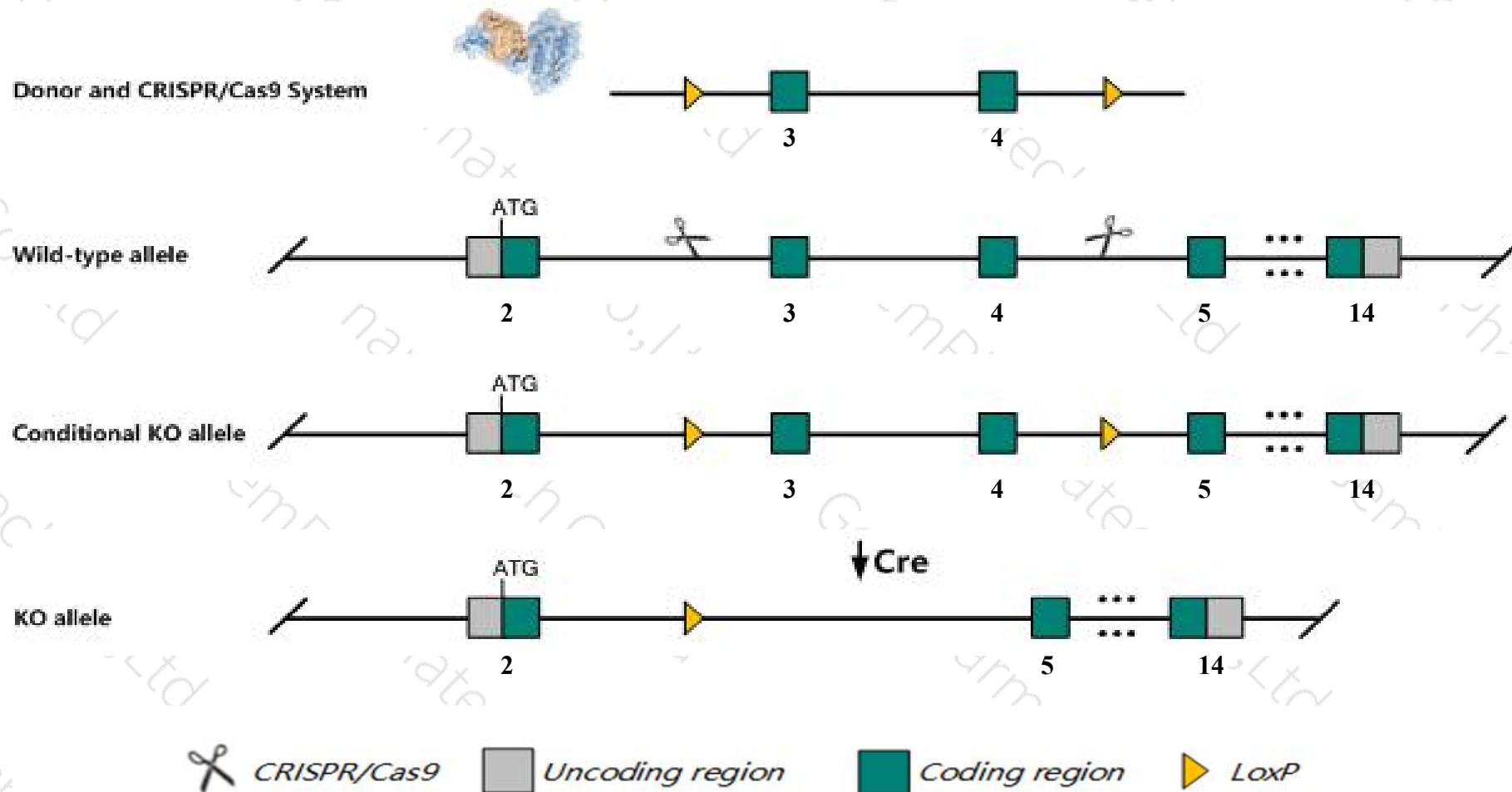
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Ical* gene has 12 transcripts. According to the structure of *Ical* gene, exon3-exon4 of *Ical*-201(ENSMUST00000038403.11) transcript is recommended as the knockout region. The region contains 236bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ical* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed. Cas9, gRNA 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.

- According to the existing MGI data, homozygous mutation of this gene results in diabetes and spontaneous lethality at 4-5 months of age on a NOD background, however mice on a 129/Sv background are normal. Onset of diabetes starts 4 weeks later than wild-type NOD mice and mutants are resistant to cyclophosphamide-accelerated diabetes.
- Transcript *Ical*-206 and *Ical*-207 may not be affected.
- The *Ical* gene is located on the Chr6. 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)

Ica1 islet cell autoantigen 1 [Mus musculus (house mouse)]

Gene ID: 15893, updated on 13-Mar-2020

Summary

Official Symbol Ica1 provided by [MGI](#)

Official Full Name islet cell autoantigen 1 provided by [MGI](#)

Primary source [MGI:MGI:96391](#)

See related [Ensembl:ENSMUSG00000062995](#)

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 69kDa, ICA69

Expression Broad expression in testis adult (RPKM 6.7), cortex adult (RPKM 5.0) and 19 other tissues [See more](#)

Orthologs [human](#) [all](#)

Transcript information (Ensembl)

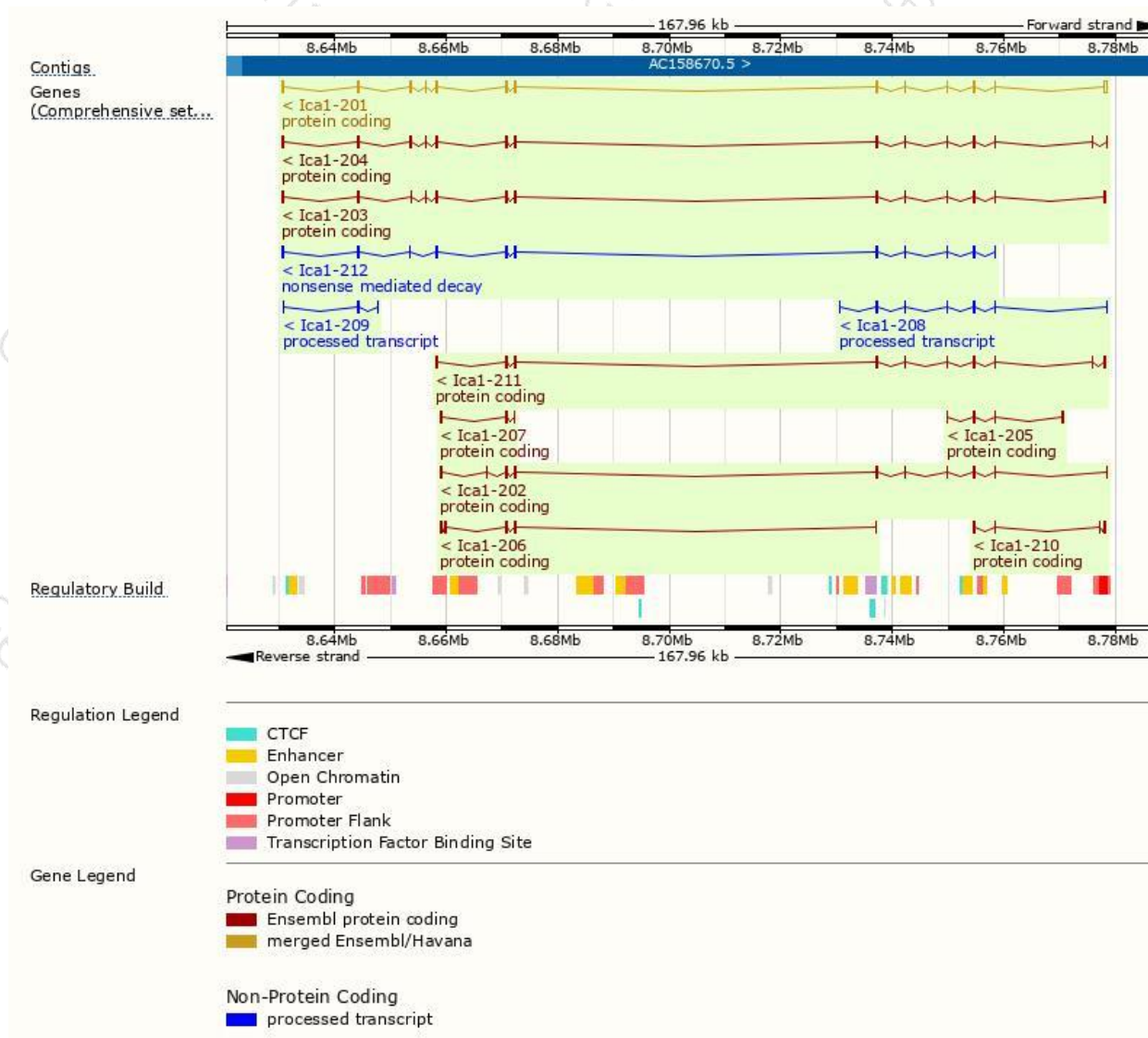
The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ica1-201	ENSMUST0000038403.11	2075	478aa	Protein coding	CCDS19911	P97411	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P2
Ica1-204	ENSMUST00000115520.7	1876	478aa	Protein coding	CCDS19911	P97411	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P2
Ica1-203	ENSMUST00000115519.7	1706	465aa	Protein coding	-	D3Z118	TSL:5 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT2
Ica1-202	ENSMUST00000115518.7	1256	310aa	Protein coding	-	D3Z119	TSL:1 GENCODE basic
Ica1-211	ENSMUST00000153390.7	1138	277aa	Protein coding	-	D3Z376	CDS 3' incomplete TSL:5
Ica1-206	ENSMUST00000126430.1	565	141aa	Protein coding	-	F7BG11	CDS 5' incomplete TSL:3
Ica1-210	ENSMUST00000151758.1	441	45aa	Protein coding	-	D3Z699	CDS 3' incomplete TSL:3
Ica1-205	ENSMUST00000126039.7	404	73aa	Protein coding	-	D3Z020	CDS 3' incomplete TSL:3
Ica1-207	ENSMUST00000127398.7	226	66aa	Protein coding	-	F6UY19	CDS 5' incomplete TSL:3
Ica1-212	ENSMUST00000156695.7	1493	302aa	Nonsense mediated decay	-	S4R217	TSL:5
Ica1-208	ENSMUST00000135113.1	960	No protein	Processed transcript	-	-	TSL:1
Ica1-209	ENSMUST00000145870.1	470	No protein	Processed transcript	-	-	TSL:2

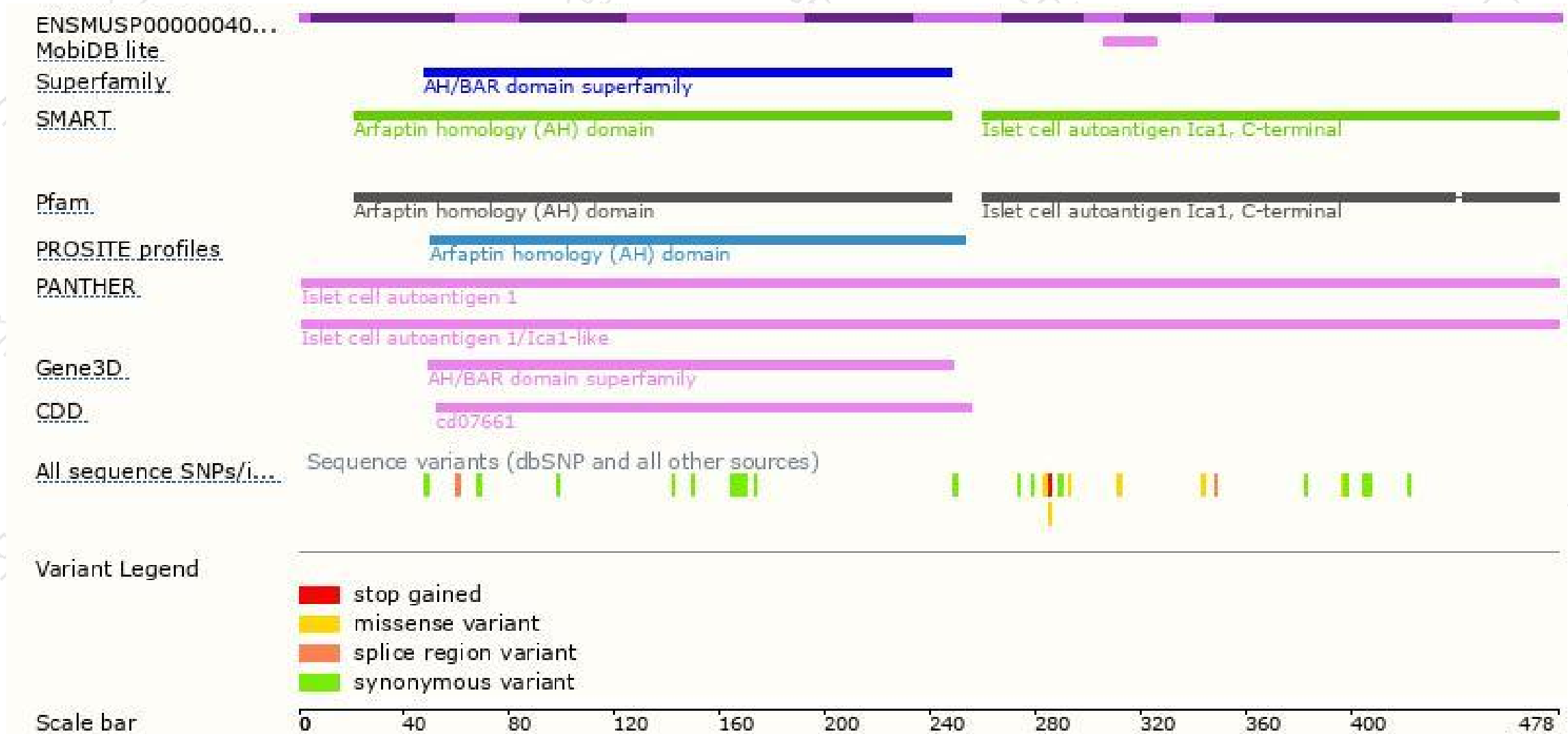
The strategy is based on the design of *Ica1-201* transcript,the transcription is shown below:



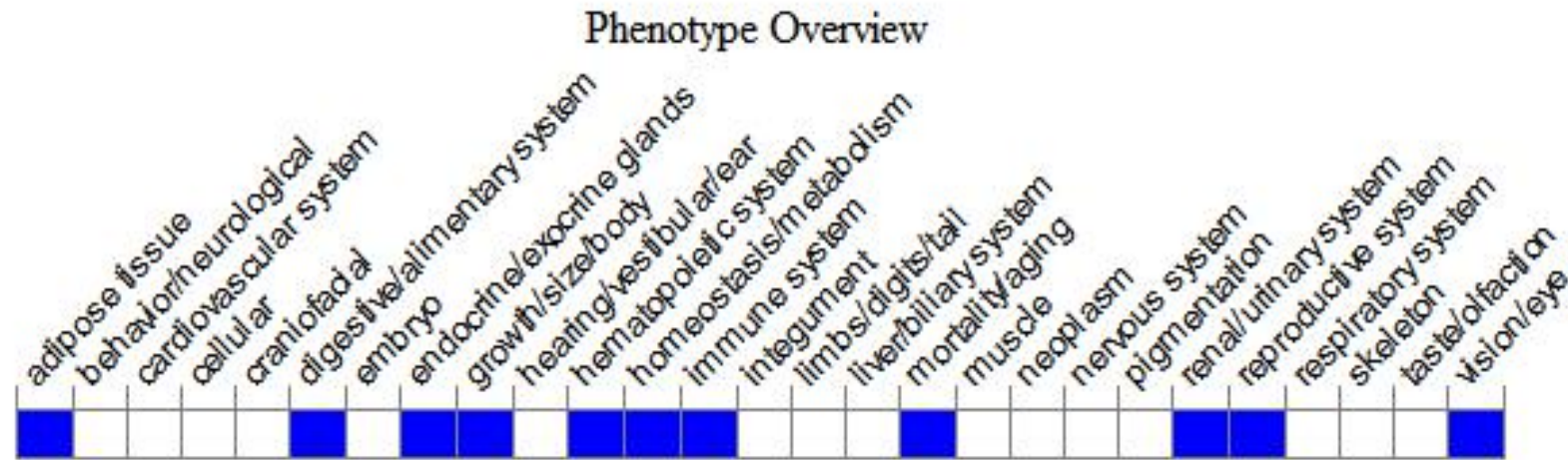
Genomic location distribution



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 mutation of this gene results in diabetes and spontaneous lethality at 4-5 months of age on a NOD background, however mice on a 129/Sv background are normal. Onset of diabetes starts 4 weeks later than wild-type NOD mice and mutants are resistant to cyclophosphamide-accelerated diabetes.

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

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