

# *Mxi1* Cas9-CKO Strategy

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**Reviewer:**

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# Project Overview



**Project Name**

***Mxi1***

**Project type**

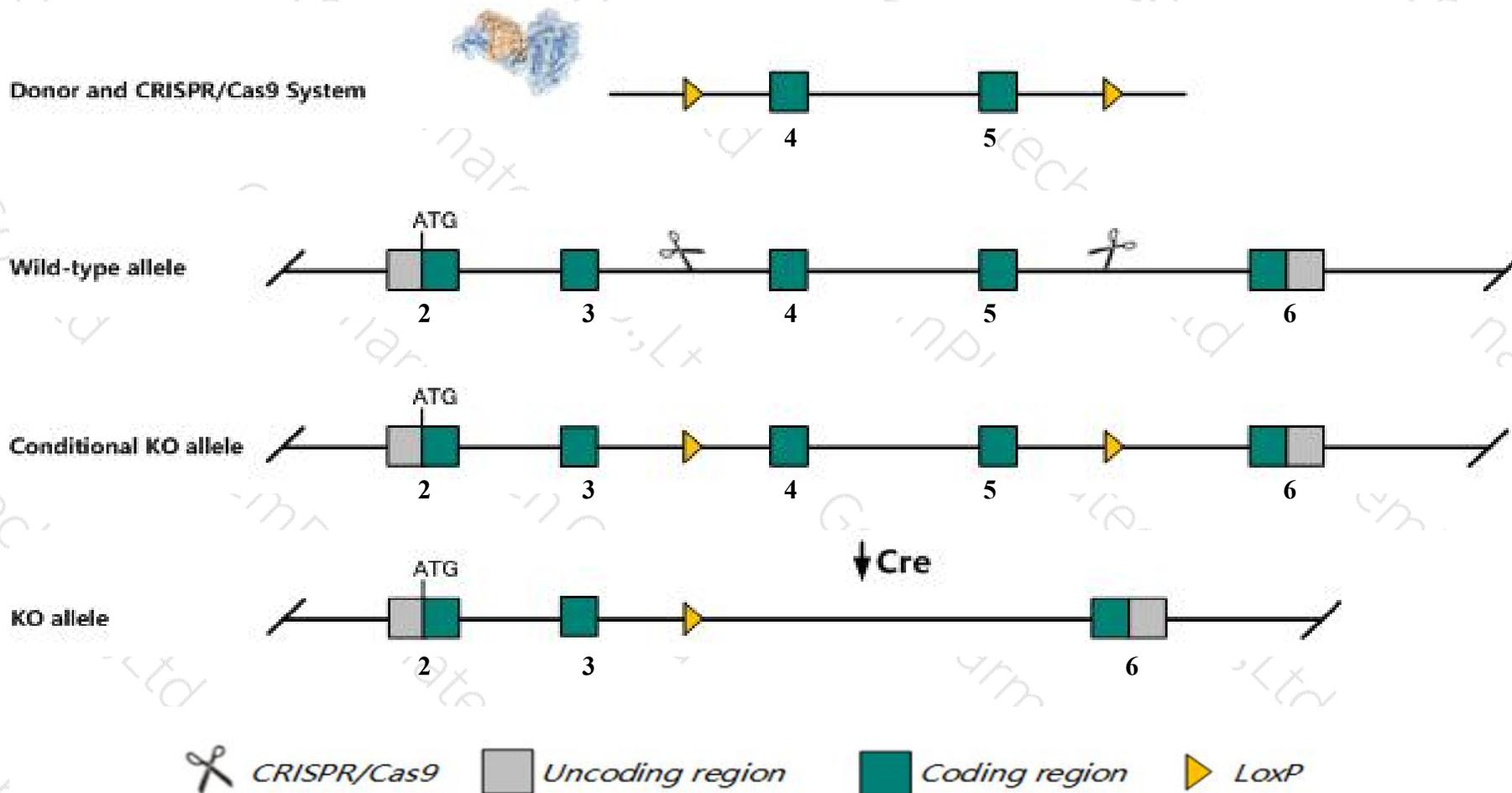
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Mx1* gene. The schematic diagram is as follows:



- The *Mxi1* gene has 9 transcripts. According to the structure of *Mxi1* gene, exon4-exon5 of *Mxi1-202* (ENSMUST00000025998.14) transcript is recommended as the knockout region. The region contains 287bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Mxi1* 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.

- According to the existing MGI data, Homozygous null mice show multisystem anomalies including progressive hyperplasia in the spleen and prostate, degenerative changes in the kidney, and increased sensitivity to carcinogens. In addition, mutant embryo fibroblasts are more prone to transformation by the Myc and Ras oncogenes.
- Transcript *Mxi1-209* may not be affected.
- The *Mxi1* gene is located on the Chr19. 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)

## Mxi1 MAX interactor 1, dimerization protein [Mus musculus (house mouse)]

Gene ID: 17859, updated on 31-Jan-2019

### Summary



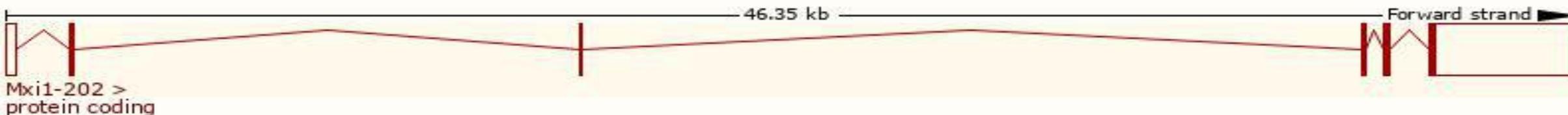
<b>Official Symbol</b>	Mxi1 provided by <a href="#">MGI</a>
<b>Official Full Name</b>	MAX interactor 1, dimerization protein provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:97245</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000025025</a>
<b>Gene type</b>	protein coding
<b>RefSeq status</b>	REVIEWED
<b>Organism</b>	<a href="#">Mus musculus</a>
<b>Lineage</b>	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
<b>Also known as</b>	Gm10197, Mad2, bHLHc11
<b>Summary</b>	This gene encodes a protein containing a helix-loop-helix domain characteristic of transcription factors, which allows heterodimerization and sequence-specific DNA binding. The encoded protein is related to a family of Myc/Max/Mad proteins that are involved in the regulation of several cellular processes. The protein encoded by this gene is a transcriptional repressor thought to negatively regulate Myc function. Three alternatively spliced transcripts encoding different isoforms have been described. [provided by RefSeq, Jul 2008]
<b>Expression</b>	Ubiquitous expression in small intestine adult (RPKM 8.9), testis adult (RPKM 8.5) and 28 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

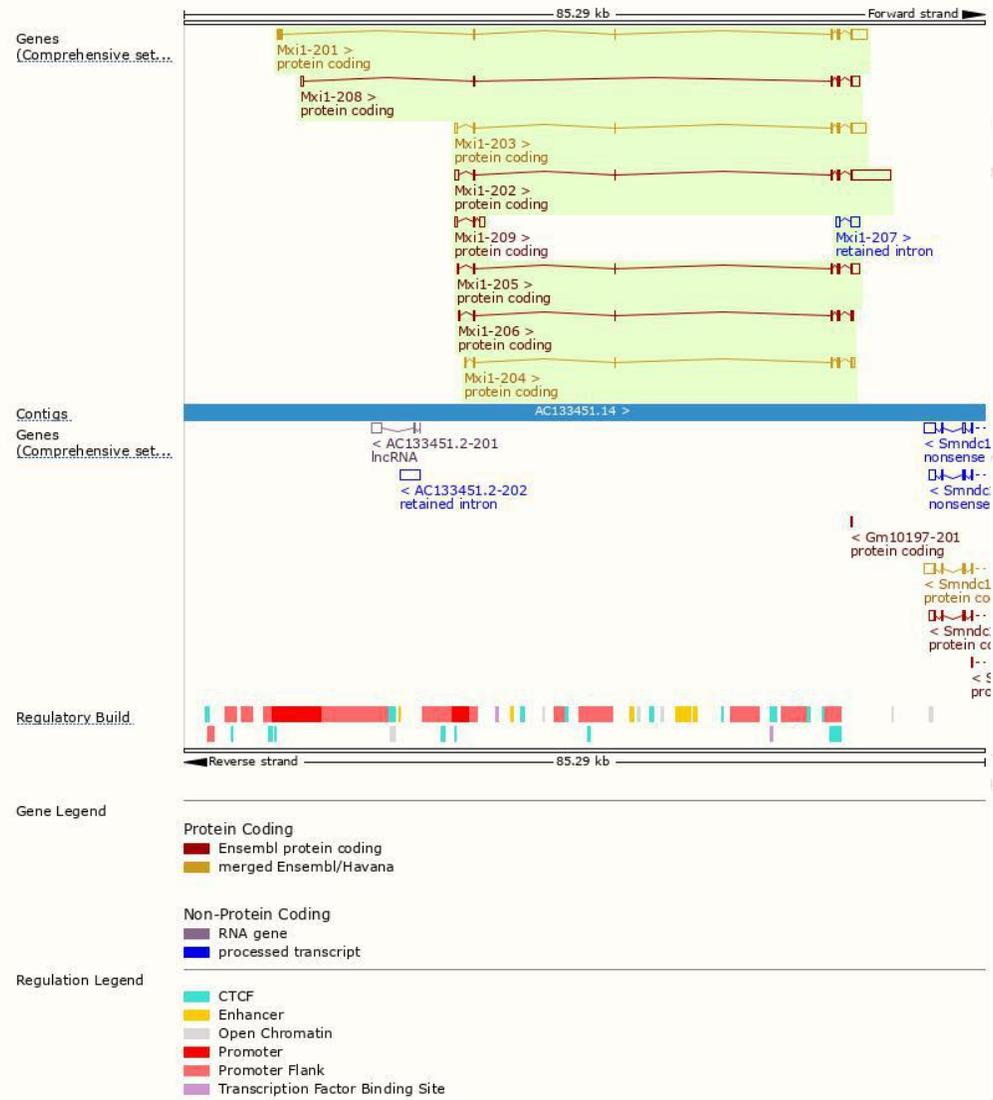
The gene has 9 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Mxi1-202	<a href="#">ENSMUST00000025998.14</a>	5036	<a href="#">192aa</a>	Protein coding	<a href="#">CCDS38023</a>	<a href="#">P50540</a>	TSL:1 GENCODE basic
Mxi1-201	<a href="#">ENSMUST00000003870.14</a>	2627	<a href="#">295aa</a>	Protein coding	<a href="#">CCDS38022</a>	<a href="#">Q3U3X2</a>	TSL:1 GENCODE basic
Mxi1-203	<a href="#">ENSMUST00000111737.2</a>	2340	<a href="#">228aa</a>	Protein coding	<a href="#">CCDS29901</a>	<a href="#">P50540 Q3USD3</a>	TSL:1 GENCODE basic APPRIS P1
Mxi1-205	<a href="#">ENSMUST00000235880.1</a>	1438	<a href="#">192aa</a>	Protein coding	<a href="#">CCDS38023</a>	-	GENCODE basic
Mxi1-204	<a href="#">ENSMUST00000235201.1</a>	1018	<a href="#">192aa</a>	Protein coding	<a href="#">CCDS38023</a>	-	GENCODE basic
Mxi1-206	<a href="#">ENSMUST00000236973.1</a>	833	<a href="#">192aa</a>	Protein coding	<a href="#">CCDS38023</a>	-	GENCODE basic
Mxi1-208	<a href="#">ENSMUST00000237480.1</a>	1544	<a href="#">182aa</a>	Protein coding	-	-	GENCODE basic
Mxi1-209	<a href="#">ENSMUST00000237837.1</a>	939	<a href="#">84aa</a>	Protein coding	-	-	GENCODE basic
Mxi1-207	<a href="#">ENSMUST00000237295.1</a>	1249	No protein	Retained intron	-	-	

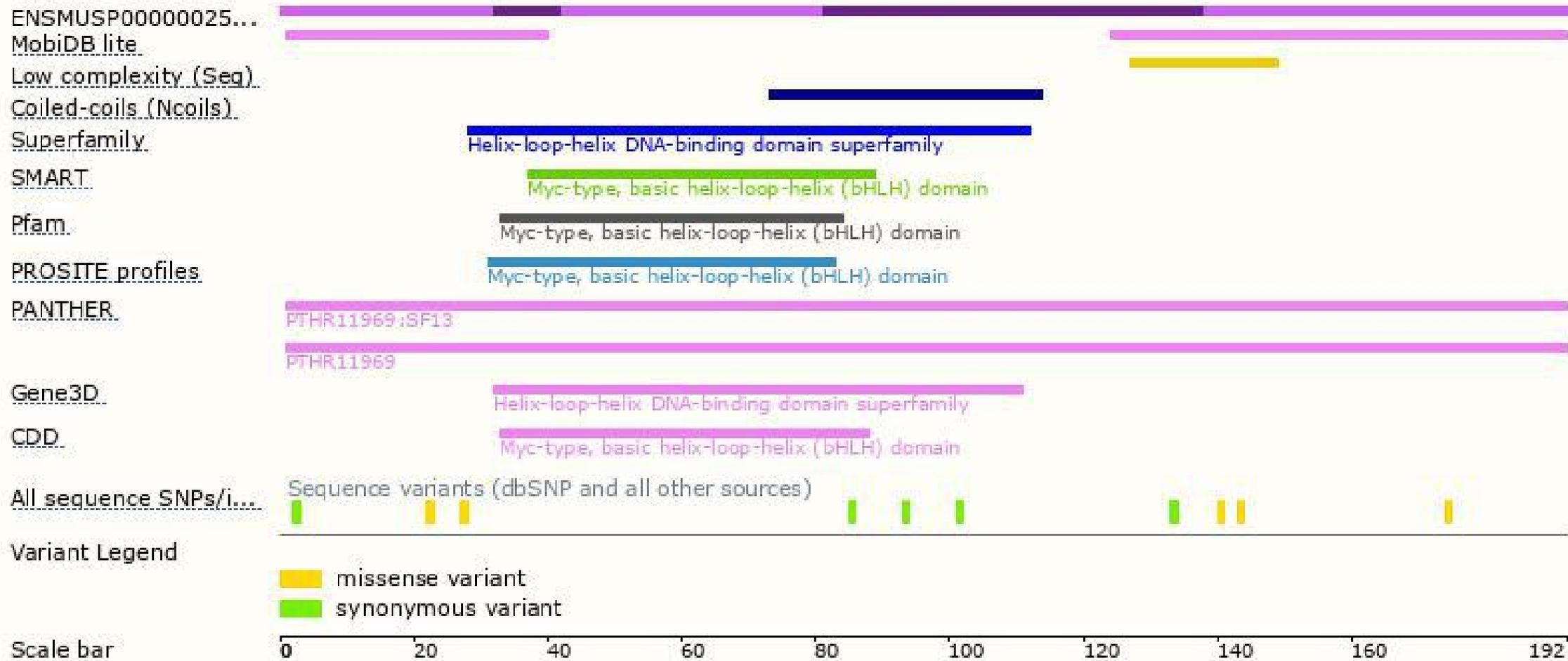
The strategy is based on the design of *Mxi1-202* transcript, The transcription is shown below



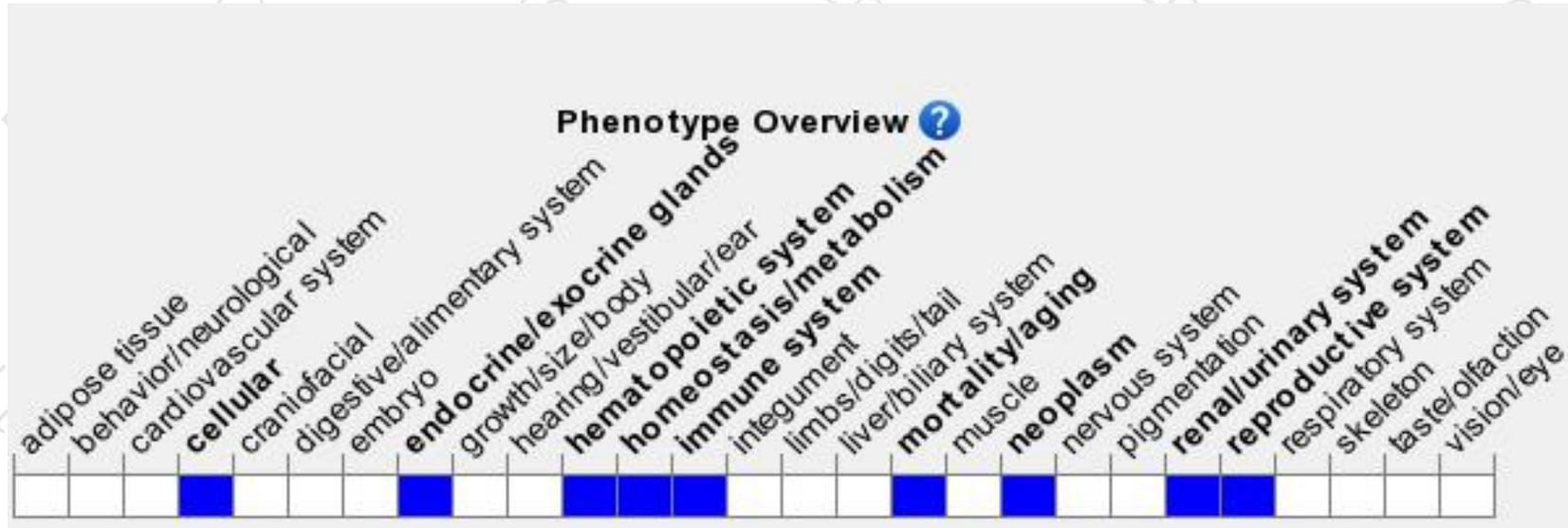
# Genomic location distribution



# 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 null mice show multisystem anomalies including progressive hyperplasia in the spleen and prostate, degenerative changes in the kidney, and increased sensitivity to carcinogens. In addition, mutant embryo fibroblasts are more prone to transformation by the Myc and Ras oncogenes.

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

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