Sox10 Cas9-CKO Strategy

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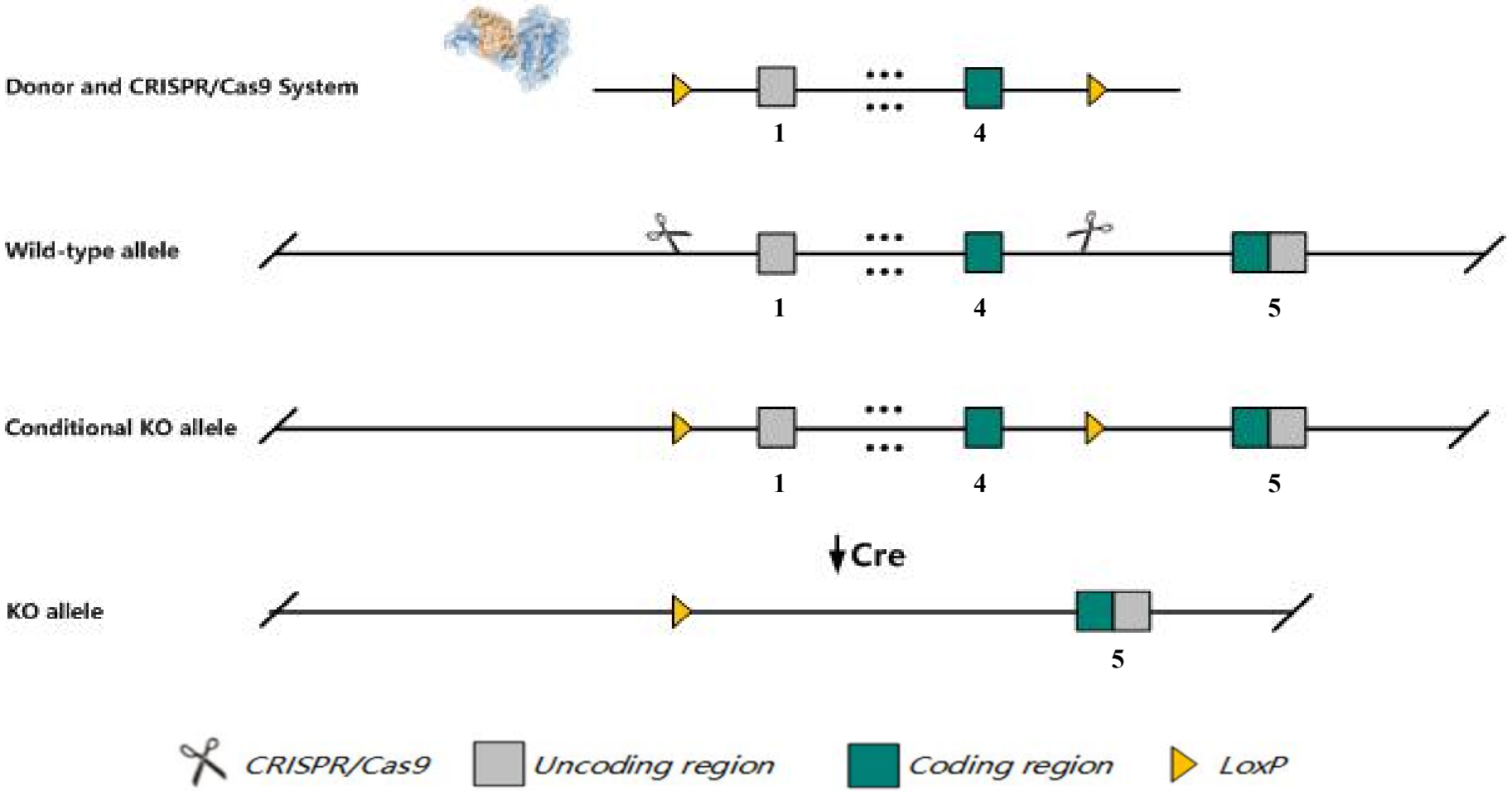
Project Overview

Project Name	***Sox10***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Sox10* gene. The schematic diagram is as follows:



Technical routes

The *Sox10* gene has 4 transcripts. According to the structure of *Sox10* gene, exon1-exon4 of *Sox10-203*(ENSMUST00000230532.1) transcript is recommended as the knockout region.The region contains start condon ATG. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Sox10* 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.

Notice

According to the existing MGI data,homozygotes for null mutations lack peripheral glial cells, melanocytes, and autonomic and enteric neurons, and die neonatally or sooner. Heterozygotes exhibit white spotting and megacolon.

The KO region contains functional region of the *Gm10863* gene.Knockout the region may affect the function of *Gm10863* gene.

The *Sox10* gene is located on the Chr15. 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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Sox10 SRY (sex determining region Y)-box 10 [Mus musculus (house mouse)]

Gene ID: 20665, updated on 13-Mar-2020

Summary

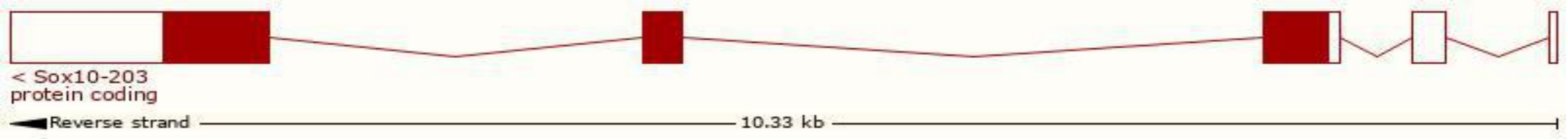
Official Symbol	Sox10 provided by MGI
Official Full Name	SRY (sex determining region Y)-box 10 provided by MGI
Primary source	MGI:MGI:98358
See related	Ensembl:ENSMUSG00000033006
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Dom, Sox21, gt
Expression	Biased expression in mammary gland adult (RPKM 22.0), cerebellum adult (RPKM 18.7) and 14 other tissues See more
Orthologs	human all

Transcript information Ensembl

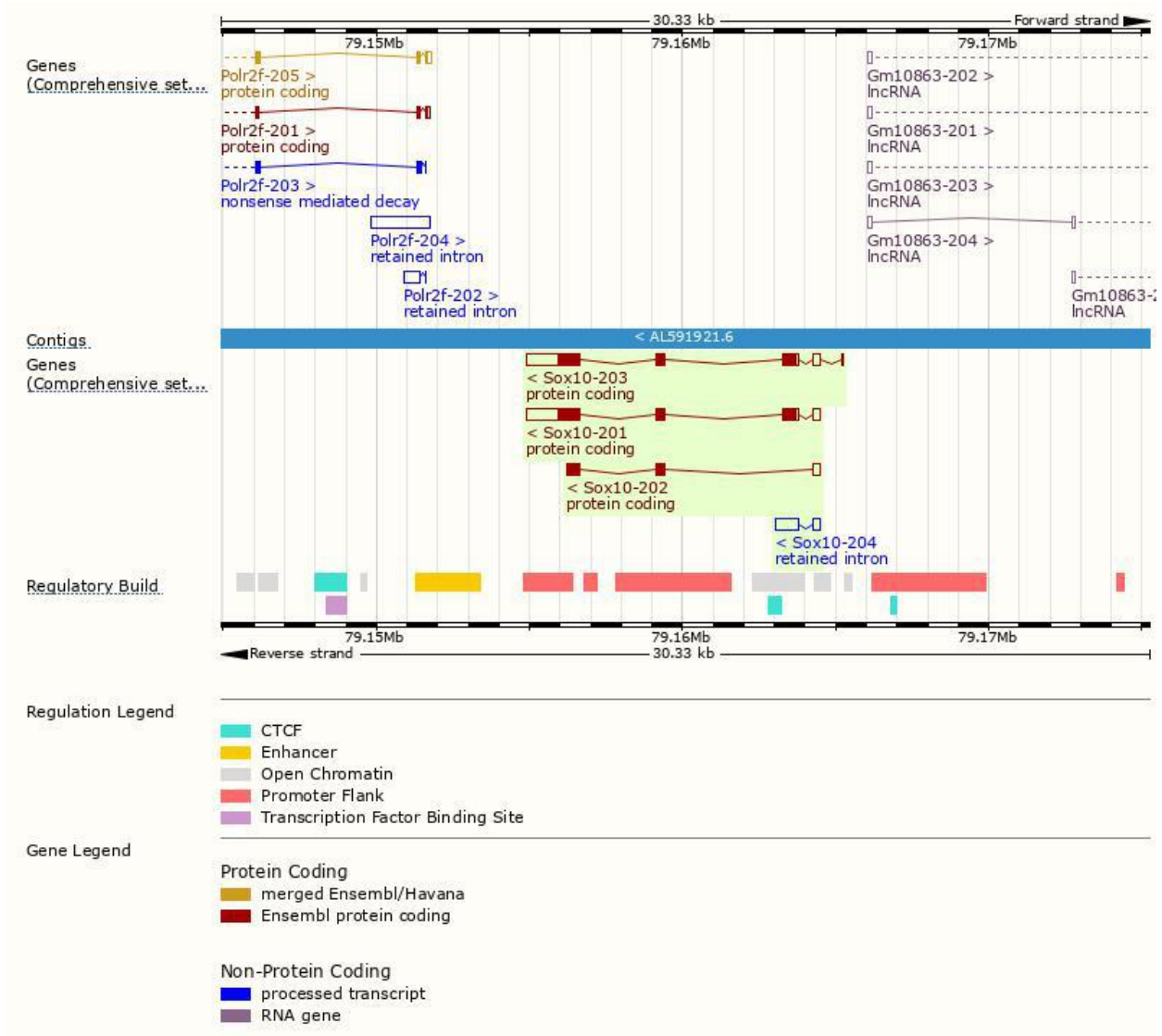
The gene has 4 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Sox10-203	ENSMUST00000230532.1	2780	466aa	Protein coding	CCDS49668	Q04888	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 P1
Sox10-201	ENSMUST00000040019.4	2713	466aa	Protein coding	CCDS49668	Q04888	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 P1
Sox10-202	ENSMUST00000230261.1	863	205aa	Protein coding	-	A0A2R8VI24	CDS 3' incomplete
Sox10-204	ENSMUST00000230891.1	982	No protein	Retained intron	-	-	

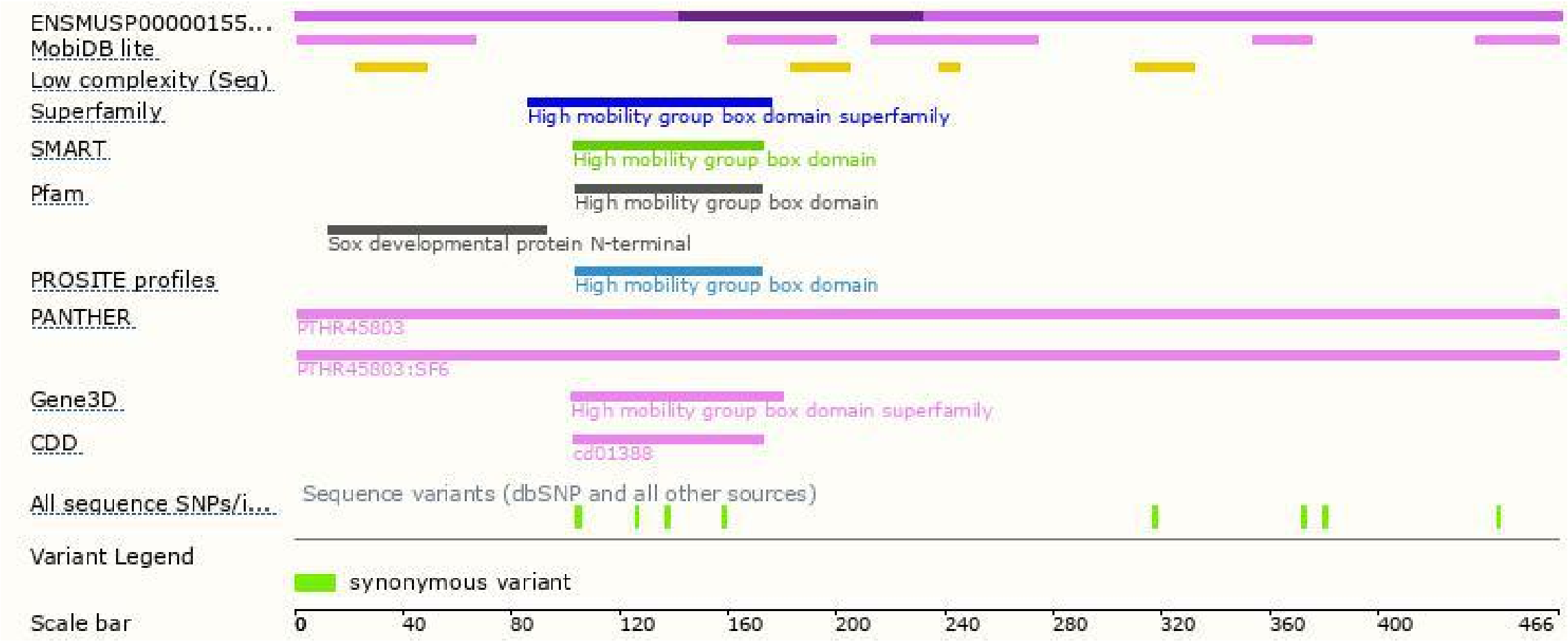
The strategy is based on the design of *Sox10-203* transcript,the transcription is shown below:



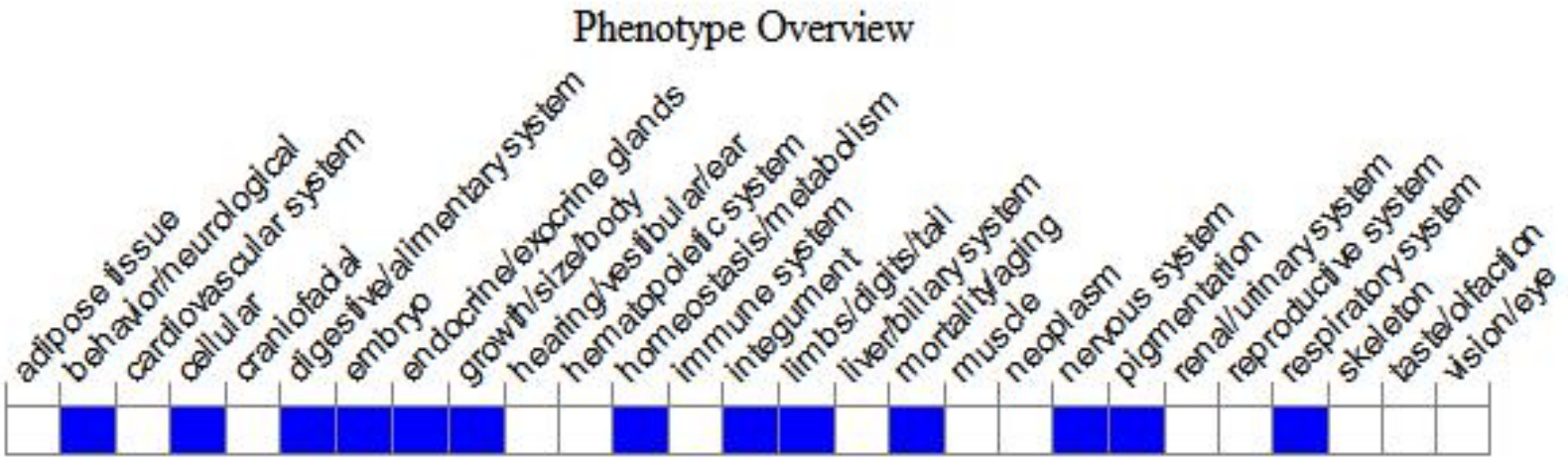
Genomic location distribution



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,homozygotes for null mutations lack peripheral glial cells, melanocytes, and autonomic and enteric neurons, and die neonatally or sooner. Heterozygotes exhibit white spotting and megacolon.

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