

# C57BL/6NGpt-*Slc15a4* Cas9-KO Strategy

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**Reviewer: Daohua Xu**

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# Project Overview

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**Project Name**

***Slc15a4***

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**Project type**

**Cas9-KO**

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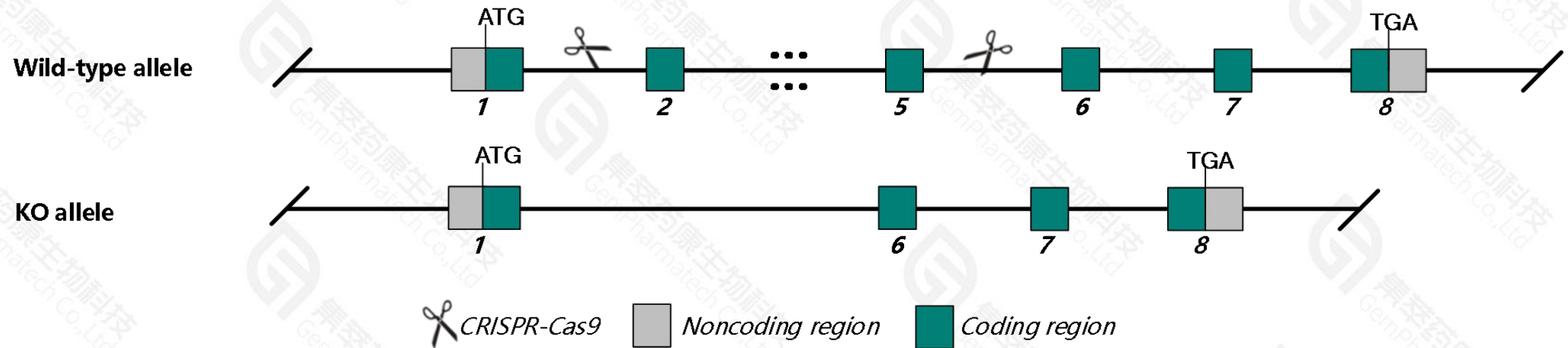
**Strain background**

**C57BL/6NGpt**

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# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Slc15a4* gene. The schematic diagram is as follows:



# Technical routes

- The *Slc15a4* gene has 11 transcripts. According to the structure of *Slc15a4* gene, exon 2-exon 5 of MGP\_C57BL6NJ\_T0075038.1 transcript is recommended as the knockout region. The region contains 706bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Slc15a4* gene. The brief process is as follows: CRISPR-Cas9 system were microinjected into the fertilized eggs of C57BL/6NGpt 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/6NGpt mice.



- According to the existing MGI data, mice homozygous for an ENU-induced mutation display abrogation of both Toll-like receptor (TLR)-induced type I IFN and proinflammatory cytokine production by plasmacytoid dendritic cells. Mice homozygous for a knock-out allele show impaired TLR9-and NOD1-mediated cytokine production and decreased susceptibility to DSS-induced colitis.
- The *Slc15a4* gene is located on the Chr 5. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

# Gene information (NCBI)

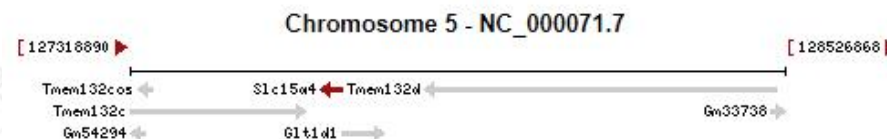
## Slc15a4 solute carrier family 15, member 4 [ *Mus musculus* (house mouse) ]

Gene ID: 100561, updated on 24-Apr-2022

[Download Datasets](#)

### Summary

Official Symbol	Slc15a4 provided by MGI
Official Full Name	solute carrier family 15, member 4 provided by MGI
Primary source	MGI:MGI:2140796
See related	Ensembl:ENSMUSG00000029416 AllianceGenome:MGI:2140796
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	<i>Mus musculus</i>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	PHT1; PTR4; AA987064; AW742963; C130069N12Rik
Summary	Enables L-histidine transmembrane transporter activity and peptide:proton symporter activity. Involved in several processes, including L-histidine transmembrane export from vacuole; positive regulation of pattern recognition receptor signaling pathway; and regulation of isotype switching to IgG isotypes. Located in early endosome membrane and lysosomal membrane. Is integral component of endosome membrane and integral component of lysosomal membrane. Is expressed in embryo. Orthologous to human SLC15A4 (solute carrier family 15 member 4). [provided by Alliance of Genome Resources, Apr 2022]
Expression	Ubiquitous expression in spleen adult (RPKM 27.6), mammary gland adult (RPKM 16.0) and 28 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>
<b>NEW</b>	Try the new <a href="#">Gene table</a> Try the new <a href="#">Transcript table</a>

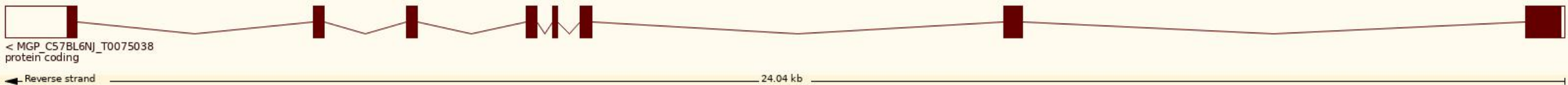


# Transcript information (Ensembl)

The gene has 11 transcripts, and all transcripts are shown below:

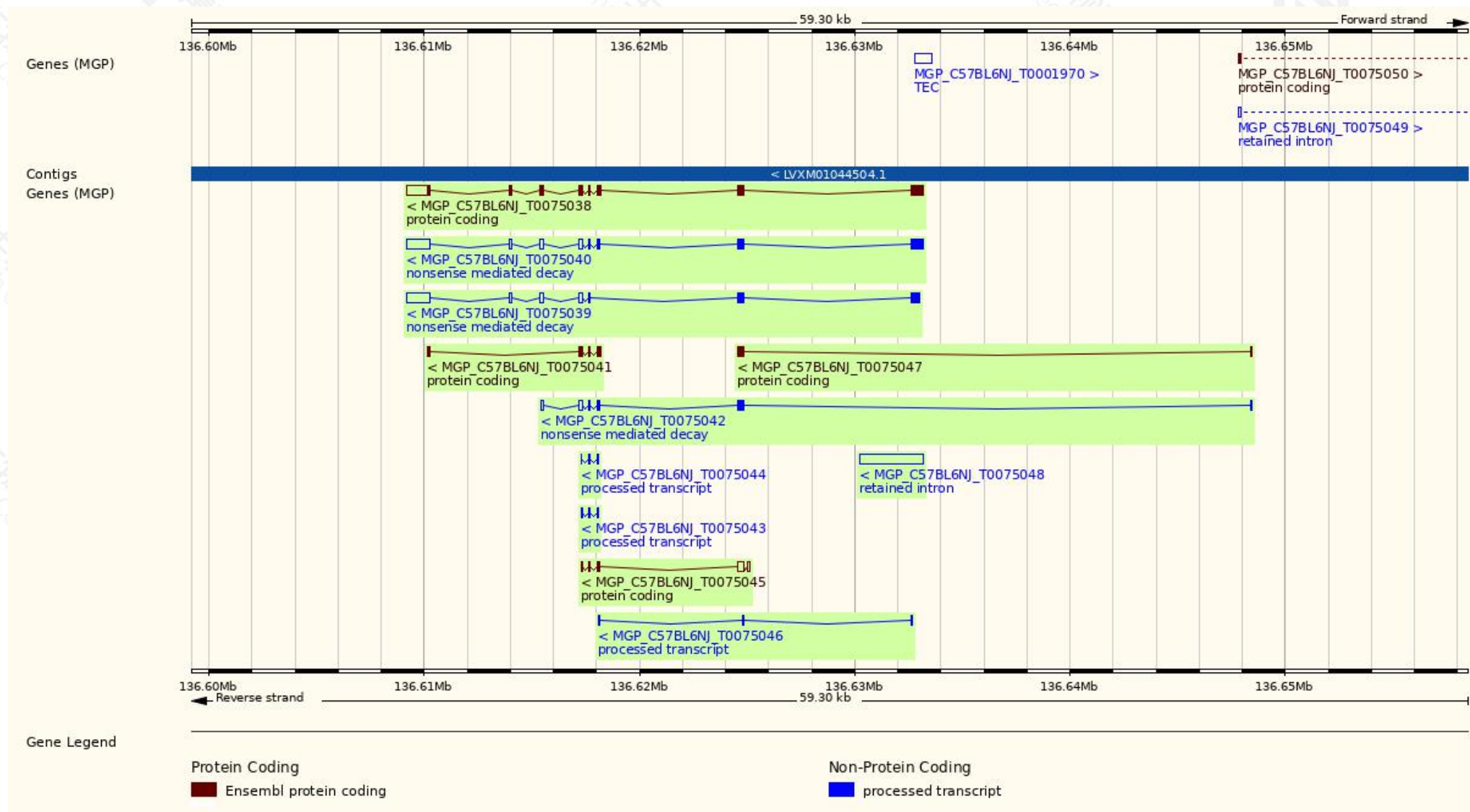
Transcript ID	Name	bp	Protein	Biotype	CCDS	UniProt Match	Flags
<a href="#">MGP_C57BL6NJ_T0075038.1</a>	-	2732	<a href="#">574aa</a>	Protein coding	<a href="#">CCDS19688</a>	<a href="#">A0A0G2JDS1</a> <a href="#">D3Z4X0</a> <a href="#">D3Z5E9</a> <a href="#">D6RDC2</a> <a href="#">F6QFB4</a> <a href="#">F7AZ26</a> <a href="#">Q91W98</a>	Ensembl Canonical
<a href="#">MGP_C57BL6NJ_T0075039.1</a>	-	2370	<a href="#">245aa</a>	Nonsense mediated decay	-	-	-
<a href="#">MGP_C57BL6NJ_T0075040.1</a>	-	2673	<a href="#">296aa</a>	Nonsense mediated decay	-	-	-
<a href="#">MGP_C57BL6NJ_T0075041.1</a>	-	507	<a href="#">168aa</a>	Protein coding	-	-	-
<a href="#">MGP_C57BL6NJ_T0075042.1</a>	-	779	<a href="#">126aa</a>	Nonsense mediated decay	-	-	-
<a href="#">MGP_C57BL6NJ_T0075043.1</a>	-	219	No protein	Processed transcript	-	-	-
<a href="#">MGP_C57BL6NJ_T0075044.1</a>	-	166	No protein	Processed transcript	-	-	-
<a href="#">MGP_C57BL6NJ_T0075045.1</a>	-	679	<a href="#">81aa</a>	Protein coding	-	-	-
<a href="#">MGP_C57BL6NJ_T0075046.1</a>	-	200	No protein	Processed transcript	-	-	-
<a href="#">MGP_C57BL6NJ_T0075047.1</a>	-	333	<a href="#">105aa</a>	Protein coding	-	-	-
<a href="#">MGP_C57BL6NJ_T0075048.1</a>	-	2937	No protein	Retained intron	-	-	-

The strategy is based on the design of MGP\_C57BL6NJ\_T0075038.1 transcript,the transcription is shown below:



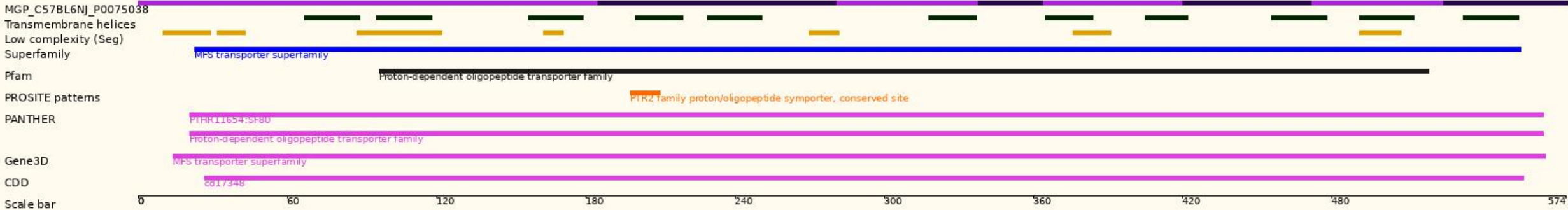


# Genomic location distribution

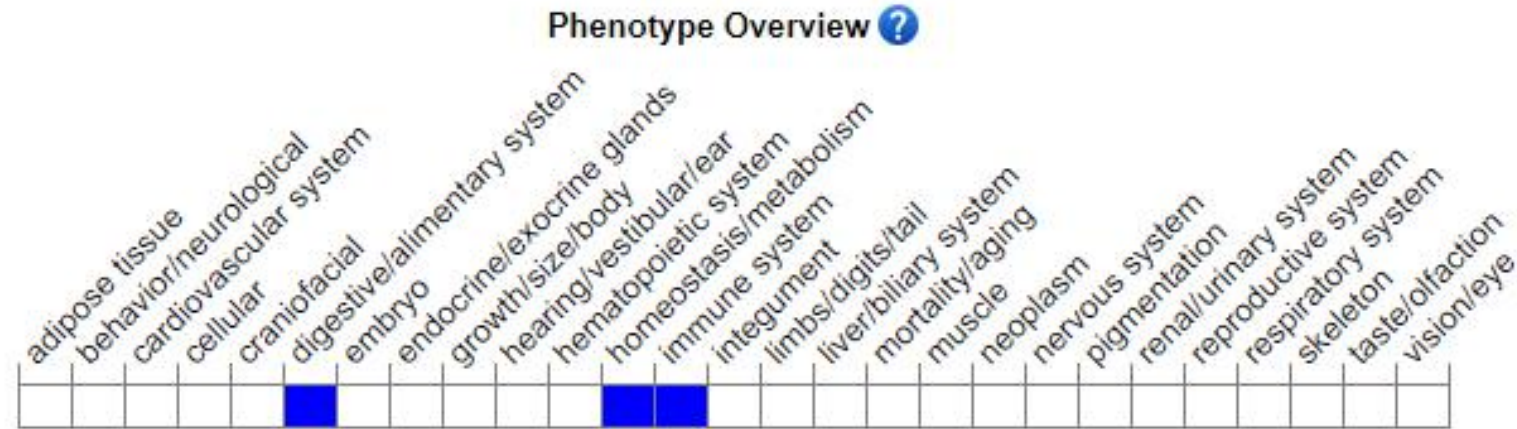




## Protein domains for MGP\_C57BL6NJ\_P0075038



# Mouse phenotype description(MGI)



*Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).*

Mice homozygous for an ENU-induced mutation display abrogation of both Toll-like receptor (TLR)-induced type I IFN and proinflammatory cytokine production by plasmacytoid dendritic cells. Mice homozygous for a knock-out allele show impaired TLR9-and NOD1-mediated cytokine production and decreased susceptibility to DSS-induced colitis.

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