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Inpp5d Cas9-CKO Strategy

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Reviewer: Yanhua Shen

Design Date: 2022-12-05

Overview

Target Gene Name

- Inpp5d

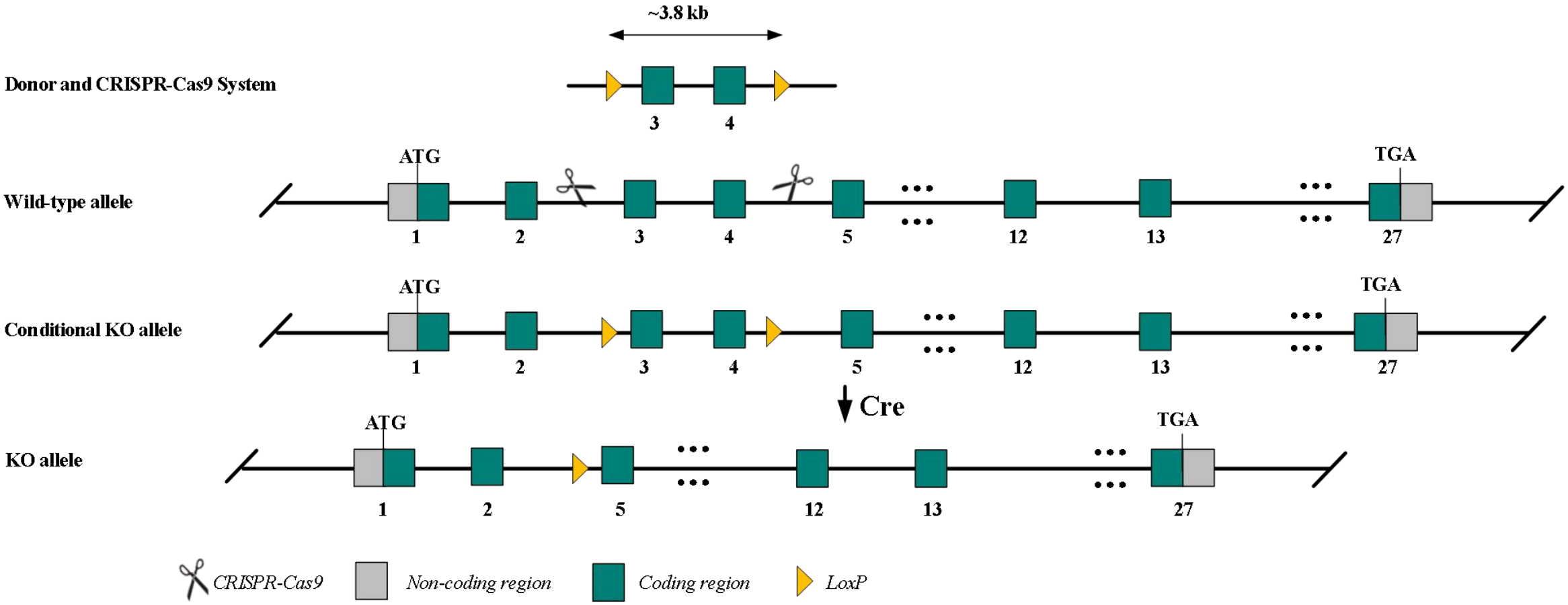
Project Type

- Cas9-CKO

Genetic Background

- C57BL/6JGpt

Strain Strategy

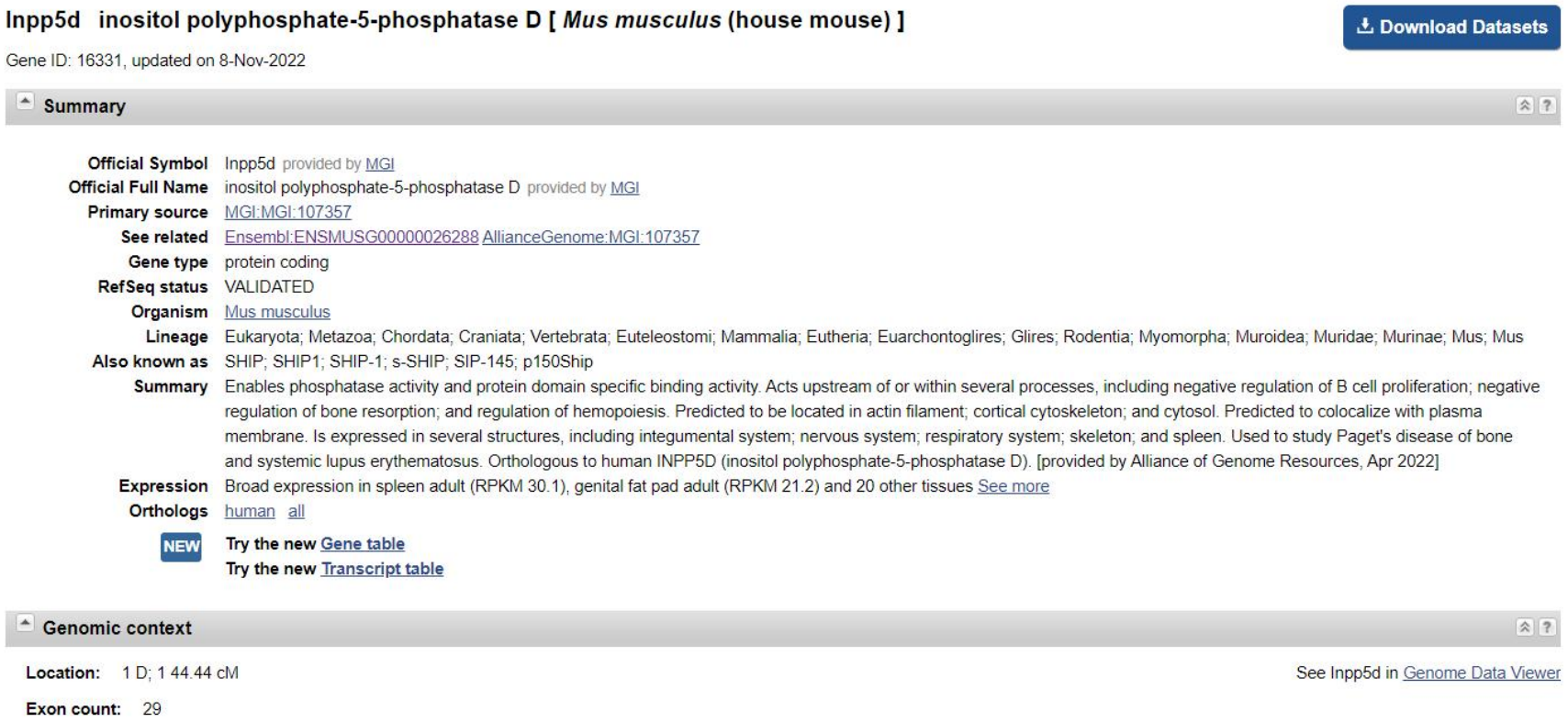


Schematic representation of CRISPR-Cas9 engineering used to edit the *Inpp5d* gene.

Technical Information

- The *Inpp5d* gene has 8 transcripts. According to the structure of *Inpp5d* gene, exon 3 and 4 of *Inpp5d*-210 (ENSMUST00000169754.8) transcript is recommended as the knockout region. The region contains 326 bp of coding sequences. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Inpp5d* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.

Gene Information



Inpp5d inositol polyphosphate-5-phosphatase D [*Mus musculus* (house mouse)]

Download Datasets

Gene ID: 16331, updated on 8-Nov-2022

Summary

Official Symbol	Inpp5d provided by MGI
Official Full Name	inositol polyphosphate-5-phosphatase D provided by MGI
Primary source	MGI:MGI:107357
See related	Ensembl:ENSMUSG00000026288 AllianceGenome:MGI:107357
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	SHIP; SHIP1; SHIP-1; s-SHIP; SIP-145; p150Ship
Summary	Enables phosphatase activity and protein domain specific binding activity. Acts upstream of or within several processes, including negative regulation of B cell proliferation; negative regulation of bone resorption; and regulation of hemopoiesis. Predicted to be located in actin filament; cortical cytoskeleton; and cytosol. Predicted to colocalize with plasma membrane. Is expressed in several structures, including integumental system; nervous system; respiratory system; skeleton; and spleen. Used to study Paget's disease of bone and systemic lupus erythematosus. Orthologous to human INPP5D (inositol polyphosphate-5-phosphatase D). [provided by Alliance of Genome Resources, Apr 2022]
Expression	Broad expression in spleen adult (RPKM 30.1), genital fat pad adult (RPKM 21.2) and 20 other tissues See more
Orthologs	human all

NEW Try the new Gene table
Try the new Transcript table

Genomic context

Location: 1 D; 1 44.44 cM — See Inpp5d in Genome Data Viewer

Exon count: 29

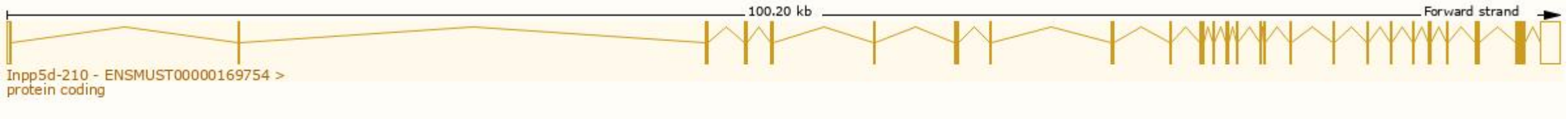
Source: https://www.ncbi.nlm.nih.gov/

Transcript Information

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

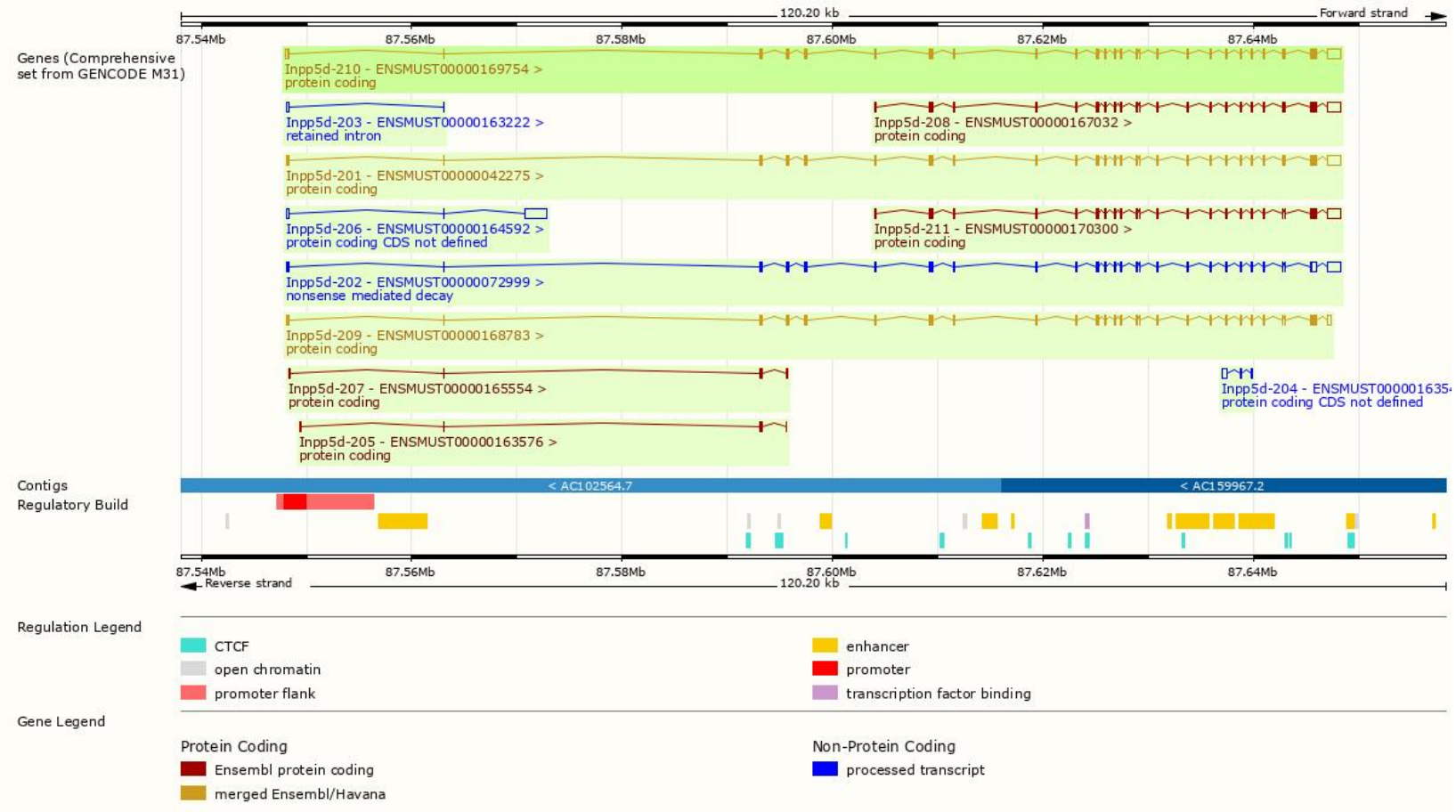
Transcript ID	Name	bp	Protein	Biotype	CCDS	UniProt Match	Flags
ENSMUST00000042275.15	Inpp5d-201	4880	1190aa	Protein coding	CCDS48311	Q9ES52-2	GENCODE basic APPRIS ALT2 TSL:1
ENSMUST00000072999.13	Inpp5d-202	4677	959aa	Nonsense mediated decay		Q9ES52-4	TSL:1
ENSMUST00000163222.2	Inpp5d-203	364	No protein	Retained intron		-	TSL:2
ENSMUST00000163548.2	Inpp5d-204	577	No protein	Protein coding CDS not defined		-	TSL:2
ENSMUST00000163576.2	Inpp5d-205	344	35aa	Protein coding		E9Q752	TSL:3 CDS 3' incomplete
ENSMUST00000164592.8	Inpp5d-206	2480	No protein	Protein coding CDS not defined		-	TSL:1
ENSMUST00000165554.2	Inpp5d-207	370	124aa	Protein coding		F7BZA7	TSL:5 CDS 5' and 3' incomplete
ENSMUST00000167032.2	Inpp5d-208	4125	928aa	Protein coding		Q9ES52-5	GENCODE basic TSL:2
ENSMUST00000168783.8	Inpp5d-209	3854	1130aa	Protein coding	CCDS48310	Q9ES52-3	GENCODE basic APPRIS ALT2 TSL:1
ENSMUST00000169754.8	Inpp5d-210	4937	1191aa	Protein coding	CCDS35655	Q9ES52-1	Ensembl Canonical GENCODE basic APPRIS P5 TSL:1
ENSMUST00000170300.8	Inpp5d-211	3942	867aa	Protein coding		Q9ES52-6	GENCODE basic TSL:2

The strategy is based on the design of *Inpp5d*-210 transcript, the transcription is shown below:



Source: https://www.ensembl.org

Genomic Information



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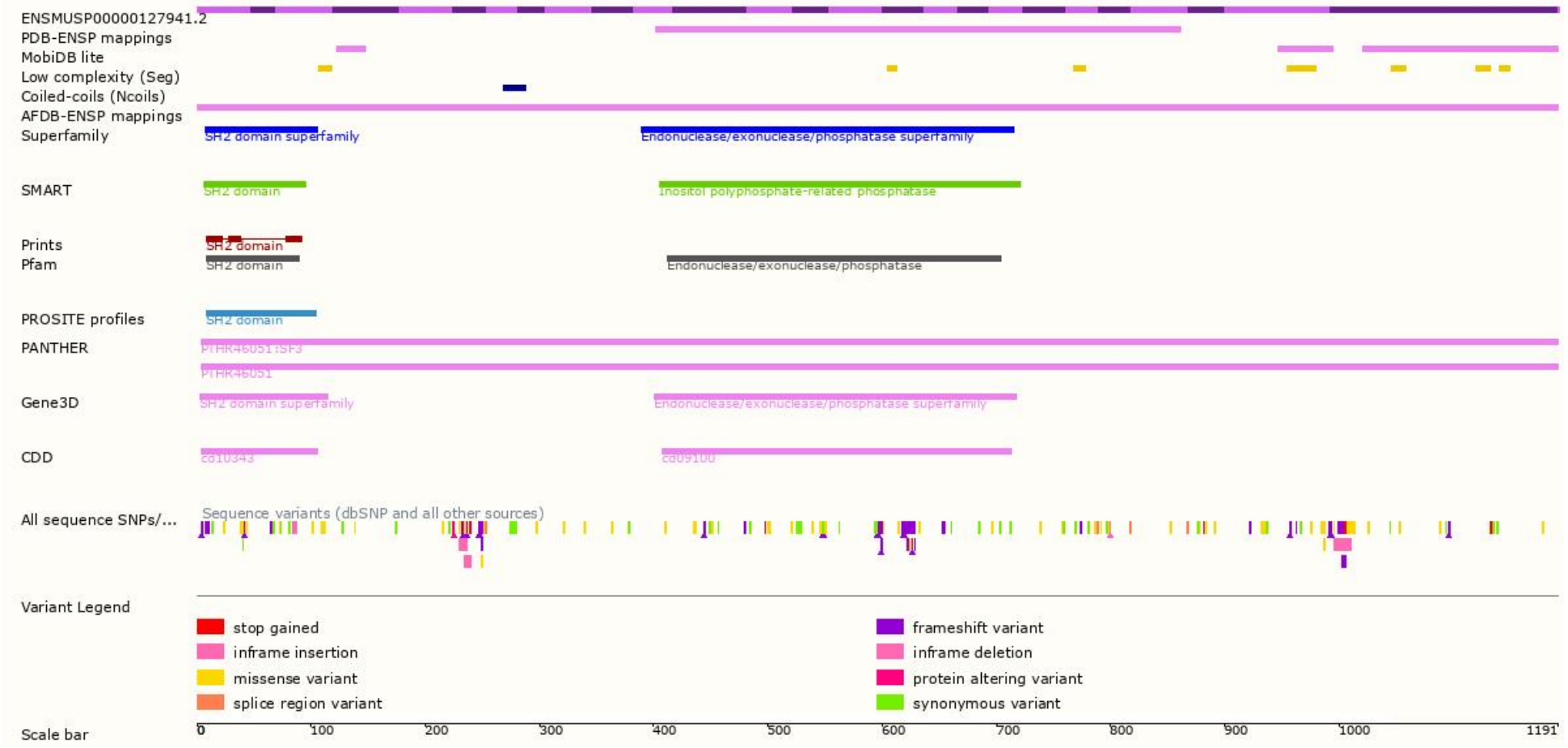
Source: : https://www.ensembl.org

Protein Information



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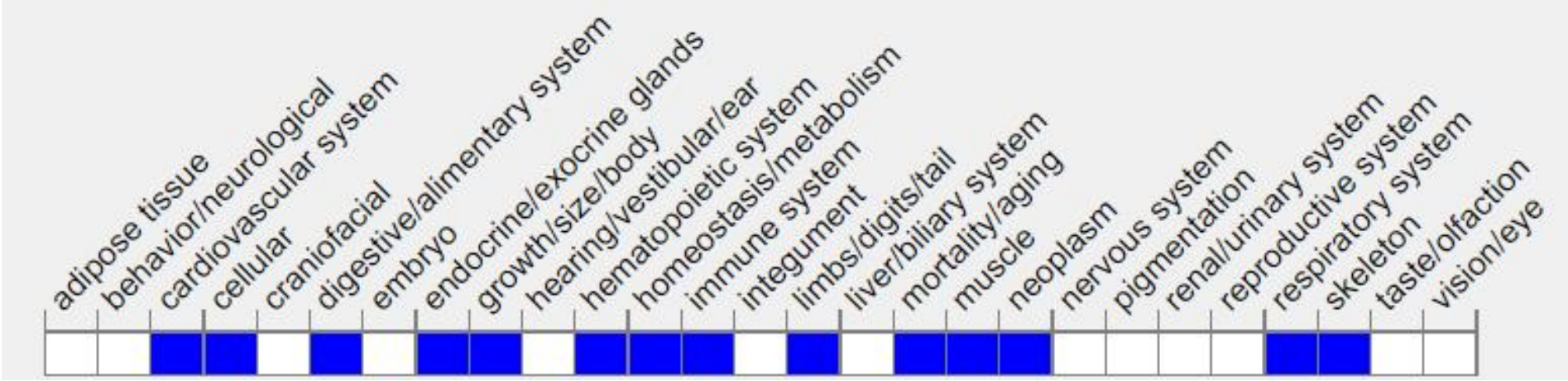
Source: : https://www.ensembl.org

Mouse Phenotype Information (MGI)



- Phenotypes affected by the mutations of *Inpp5d* gene are marked in blue. According to the existing MGI data, homozygous null mice fail to reject fully mismatched allogeneic marrow grafts, do not develop graft versus host disease, and show enhanced survival after such transplants. Homozygous splice site mutants exhibit wasting, granulocytic lung infiltration anddefective cytolysis by NK cells and CTLs.

Source: https://www.informatics.jax.org

Important Information

- The effect of this strategy on the expression of *Inpp5d*-208 and 211 transcripts in mice is currently unknown.
- *Inpp5d* is located on Chr 1. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.