GemPharmatech™

Dpp6 Cas9-KO Strategy

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Reviewer: Yanhua Shen

Design Date: 2023-7-27

Overview

Target Gene Name

- Dpp6

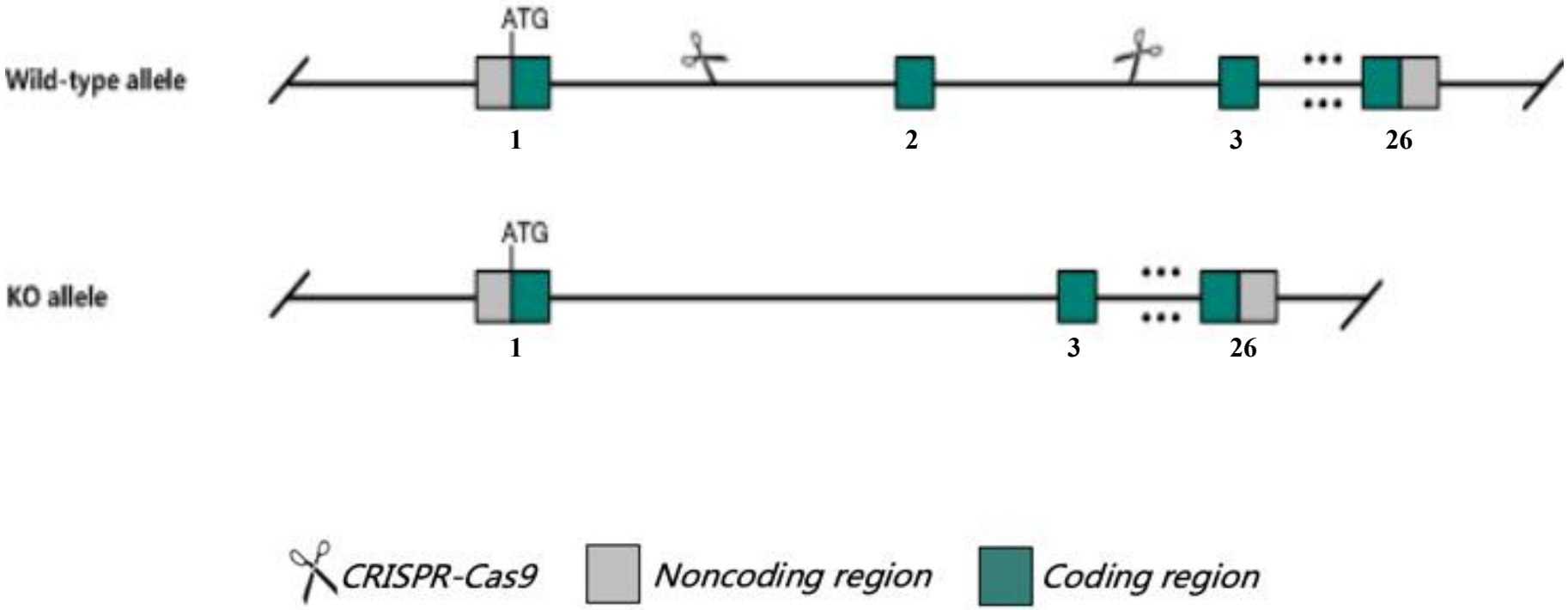
Project Type

- Cas9-KO

Genetic Background

- C57BL/6JGpt

Strain Strategy



Technical Information

- The *Dpp6* gene has 9 transcripts. According to the structure of *Dpp6* gene, exon2 of *Dpp6*-204 (ENSMUST00000122171.8) transcript is recommended as the knockout region. The region contains 115bp coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Dpp6* gene. The brief process is as follows: gRNAs were transcribed in vitro. Cas9 and gRNAs 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.

Gene Information

Dpp6 dipeptidylpeptidase 6 [*Mus musculus* (house mouse)]

Download Datasets

Gene ID: 13483, updated on 21-Jun-2023

Summary

Official Symbol	Dpp6 provided by MGI
Official Full Name	dipeptidylpeptidase 6 provided by MGI
Primary source	MGI:MGI:94921
See related	Ensembl:ENSMUSG00000061576 AllianceGenome:MGI:94921
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	Rw; Dpp-6; Peplb; D5Buc3; D5Buc4; D5Buc5; DPP VI; Gm1377; In(5)6H-p; B930011P16Rik
Summary	Enables potassium channel regulator activity. Involved in protein localization to plasma membrane and regulation of potassium ion transmembrane transport. Acts upstream of or within neuronal action potential and positive regulation of potassium ion transmembrane transport. Located in plasma membrane. Part of voltage-gated potassium channel complex. Is expressed in several structures, including 1st branchial arch; alimentary system; central nervous system; intervertebral disc; and nose. Human ortholog(s) of this gene implicated in amyotrophic lateral sclerosis; autosomal dominant non-syndromic intellectual disability 33; and spinal muscular atrophy. Orthologous to human DPP6 (dipeptidyl peptidase like 6). [provided by Alliance of Genome Resources, Apr 2022]
Expression	Biased expression in cerebellum adult (RPKM 32.7), frontal lobe adult (RPKM 23.3) and 6 other tissues See more
Orthologs	human all

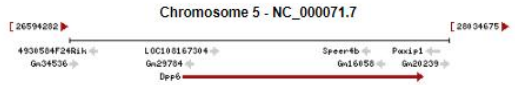
NEW Try the new Gene table
Try the new Transcript table

Genomic context

Location: 5 B1; 5 12.92 cM — See Dpp6 in Genome Data Viewer

Exon count: 29

Annotation release	Status	Assembly	Chr	Location
RS_2023_04	current	GRCm39 (GCF_000001635.27)	5	NC_000071.7 (27022355..27932498)
108.20200622	previous assembly	GRCm38.p6 (GCF_000001635.26)	5	NC_000071.6 (26817357..27727500)



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

Transcript Information

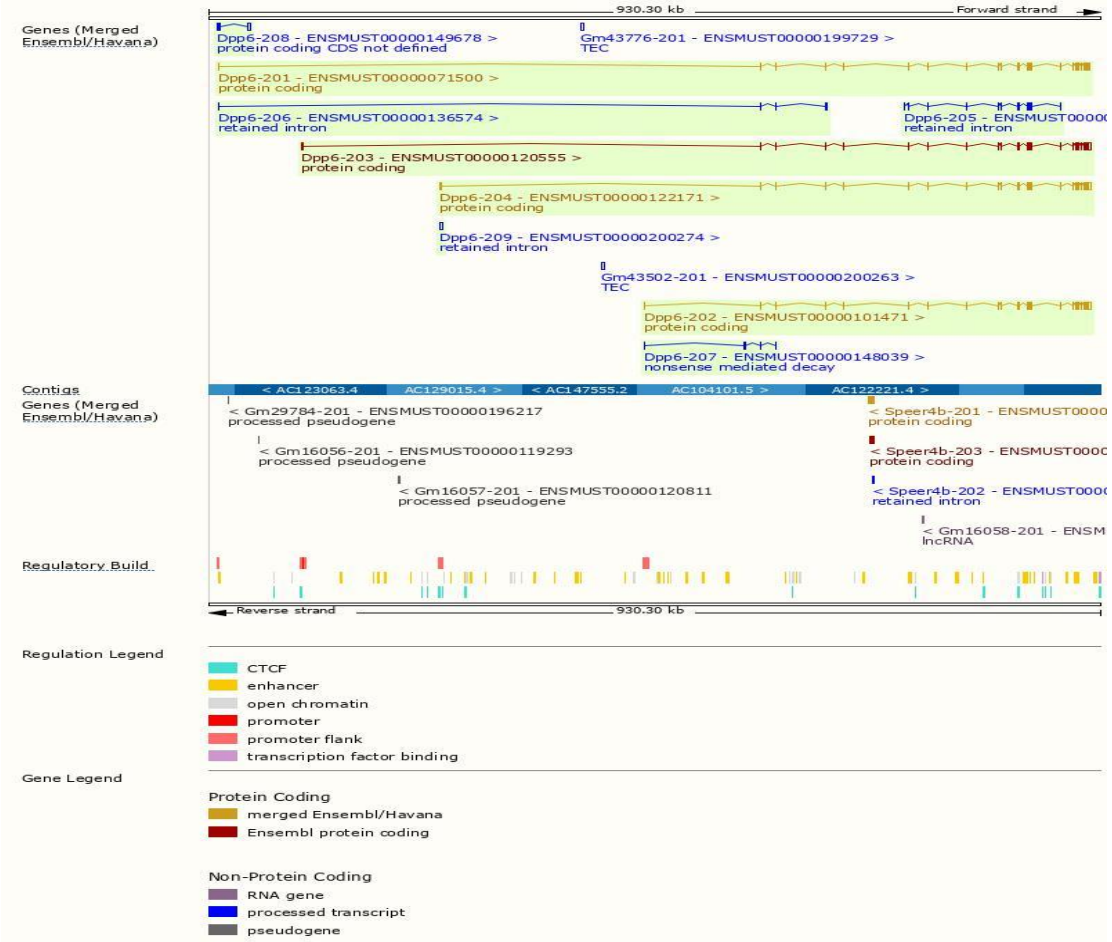
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Dpp6-204	ENSMUST00000122171.8	4720	859aa	Protein coding	CCDS51446		A single transcript chosen for a gene which is the most conserved, most highly expressed, has the longest coding sequence and is represented in other key resources, such as NCBI and UniProt. This is defined in detail on http://www.ensembl.org/info/genome/genebuild/canonical.html Ensembl Canonical , The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:1 ,
Dpp6-203	ENSMUST00000120555.8	4703	801aa	Protein coding	CCDS57332		The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:1 ,
Dpp6-202	ENSMUST00000101471.4	4491	803aa	Protein coding	CCDS51447		The GENCODE set is the gene set for human and mouse. GENCODE basic , APPRIS P4 , TSL:1 ,
Dpp6-201	ENSMUST00000071500.13	3826	804aa	Protein coding	CCDS19139		The GENCODE set is the gene set for human and mouse. GENCODE basic , APPRIS ALT1 , TSL:1 ,
Dpp6-207	ENSMUST00000148039.8	664	40aa	Nonsense mediated decay			TSL:5 ,
Dpp6-208	ENSMUST00000149678.2	2520	No protein	Protein coding CDS not defined			TSL:1 ,
Dpp6-209	ENSMUST00000200274.2	2738	No protein	Retained intron			TSL:NA ,
Dpp6-206	ENSMUST00000136574.2	2506	No protein	Retained intron			TSL:1 ,
Dpp6-205	ENSMUST00000134175.2	1316	No protein	Retained intron			TSL:1 ,

The strategy is based on the design of *Dpp6*-204 transcript, the transcription is shown below:

Source: https://www.ensembl.org

Genomic Information

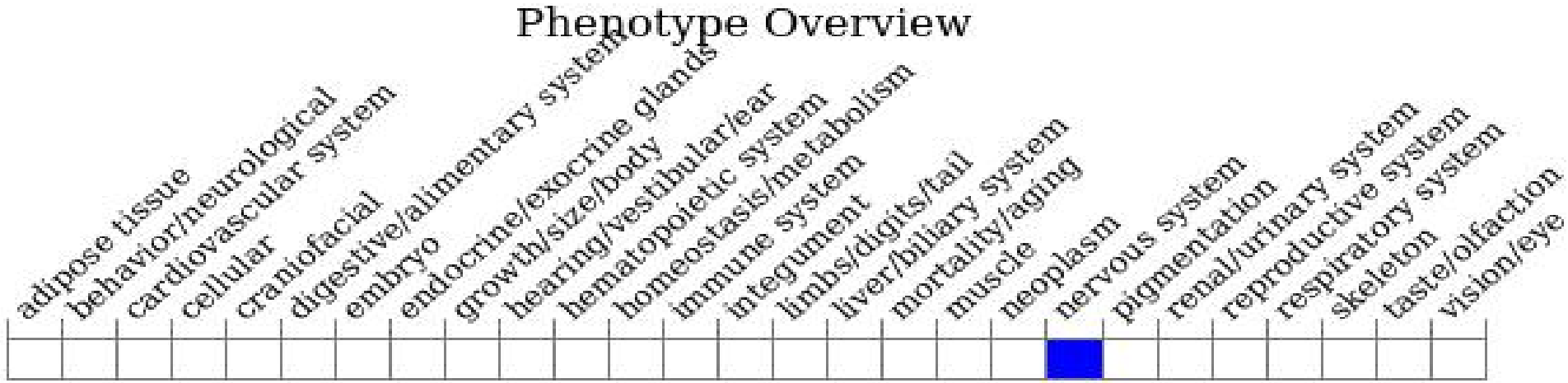


Source: : https://www.ensembl.org

Protein Information

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

Mouse Phenotype Information (MGI)



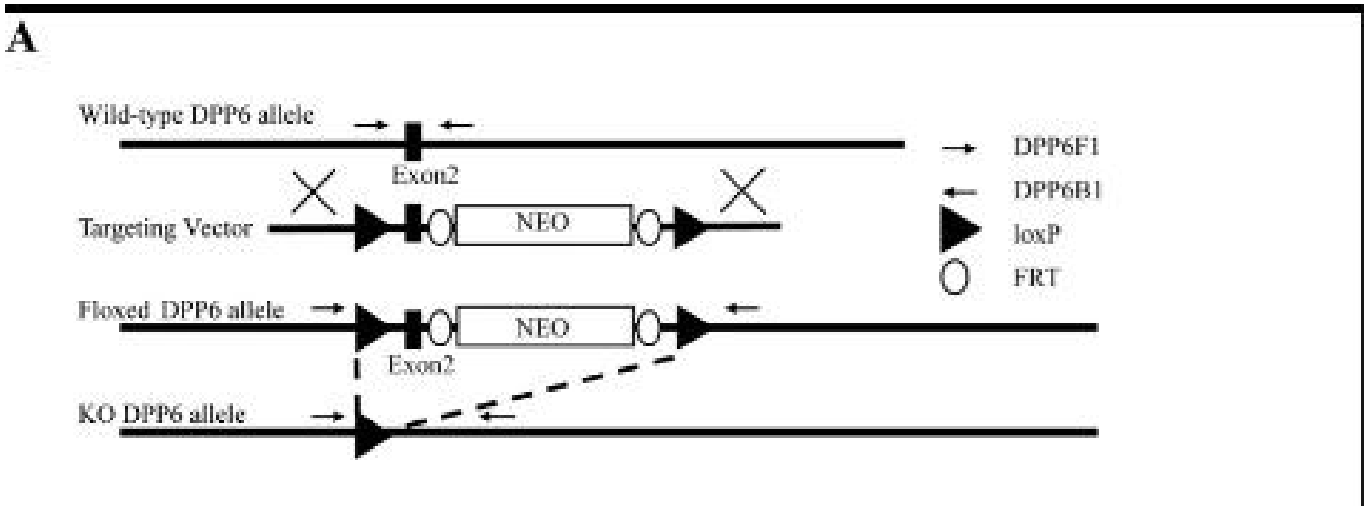
- Mice homozygous for a knock-out allele exhibit loss of A-type K+ current gradients in distal dendrites.

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

Important Information

- *Dpp6* is located on Chr5. 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 risks of the mutation on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

References



Construction and characterization of DPP6-KO mice

Targeted deletion of DPP6 exon 2 produces loss of DPP6 RNA and protein without producing compensatory changes in DPP10 expression. **(A)** A schematic describing the strategy used to delete a region of genomic DNA containing the second exon of DPP6. A targeting vector was 'knocked in' to the DPP6 locus via homologous recombination. Resulting mice were then crossed with mice expressing Cre-recombinase in germline cells causing recombination between the loxP sites flanking exon 2 and loss of the intervening genomic sequence. **(B)** PCR amplification product from a WT mouse, a mouse heterozygous for DPP6 deletion and a DPP6-KO mouse using primers DPP6F1 and DPP6B1 indicated in the schematic above. **(C)** Quantitative PCR amplification of DPP6 mRNA and mRNA from the homologous gene DPP10 showing loss of DPP6 amplification product in the DPP6-KO (WT, Ct=20.9 ± 0.3; KO, Ct=37.3 ± 1.7) and no change in the amount of DPP10 product in the DPP6-KO (WT, Ct=22.7 ± 0.4; KO, Ct=21.5 ± 0.7). All qPCR experiments were performed with whole brain mRNA. **(D)** Loss of DPP6 immunoreactivity in the DPP6-KO. **(E)** Retained pattern of DPP10 in situ hybridization in the DPP6-KO.

Sun W, Maffie JK, Lin L, Petralia RS, Rudy B, Hoffman DA. DPP6 establishes the A-type K(+) current gradient critical for the regulation of dendritic excitability in CA1 hippocampal neurons. Neuron. 2011 Sep 22;71(6):1102-15. doi: 10.1016/j.neuron.2011.08.008. Epub 2011 Sep 21. PMID: 21943606; PMCID: PMC3184237.