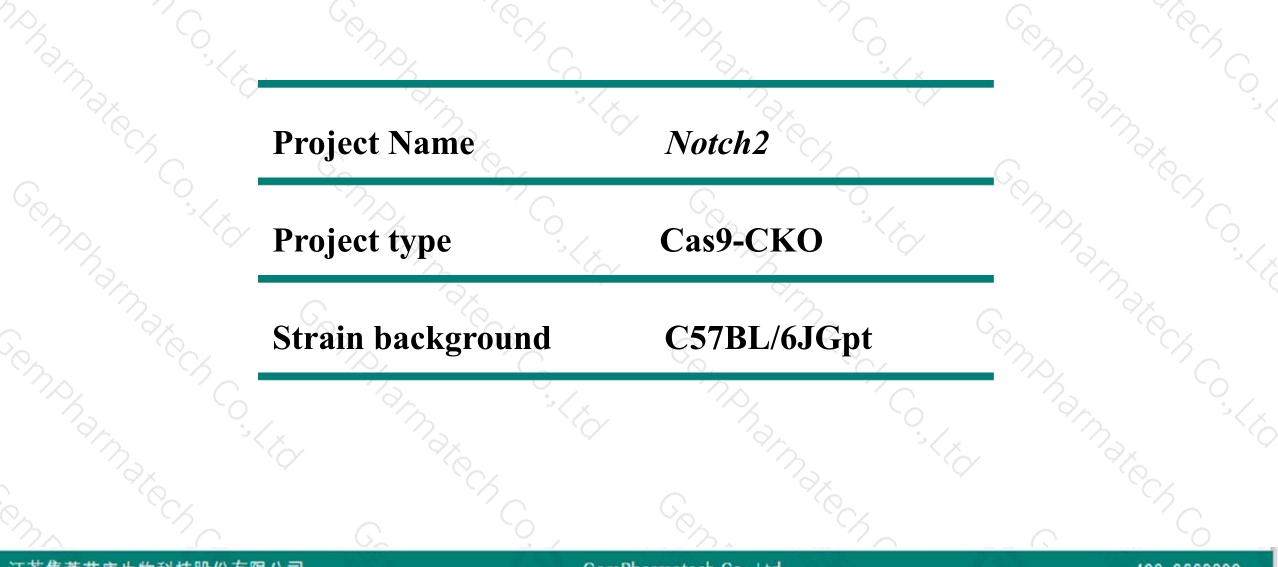


Notch2 Cas9-CKO Strategy



Project Overview





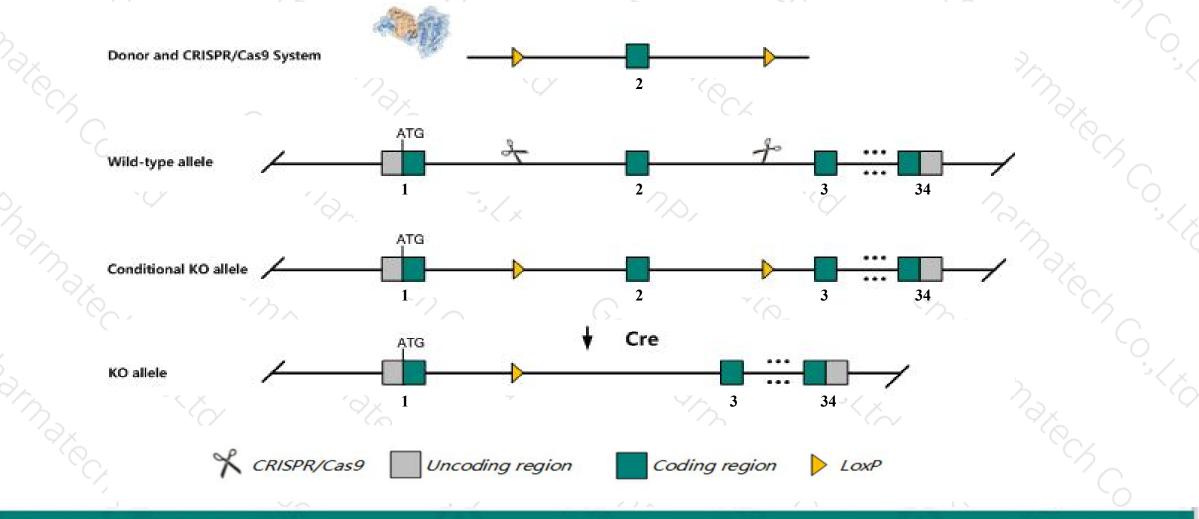
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Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Notch2* gene. The schematic diagram is as follows:



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 The Notch2 gene has 2 transcripts. According to the structure of Notch2 gene, exon2 of Notch2-201 (ENSMUST00000079812.7) transcript is recommended as the knockout region. The region contains 82bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Notch2* 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, Homozygotes for null alleles exhibit defects in embryonic development resulting in embryonic or neonatal lethality.
- The Notch2 gene is located on the Chr3. 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)



\$?

Notch2 notch 2 [Mus musculus (house mouse)]

Gene ID: 18129, updated on 2-Apr-2019

Summary

Official Symbol	Notch2 provided by MGI
Official Full Name	notch 2 provided byMGI
Primary source	MGI:MGI:97364
See related	Ensembl:ENSMUSG0000027878
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	AI853703, N2
Expression	Ubiquitous expression in spleen adult (RPKM 26.6), ovary adult (RPKM 22.8) and 28 other tissues See more
Orthologs	human all

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The gene has 2 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Notch2-201	ENSMUST00000079812.7	10500	<u>2473aa</u>	Protein coding	CCDS51013	<u>G5E8J0</u>	TSL:1 GENCODE basic APPRIS P1
Notch2-202	ENSMUST00000198324.1	1896	No protein	Retained intron	-		TSL:1

The strategy is based on the design of Notch2-201 transcript, The transcription is shown below

Notch2-201 > protein coding

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136.82 kb -

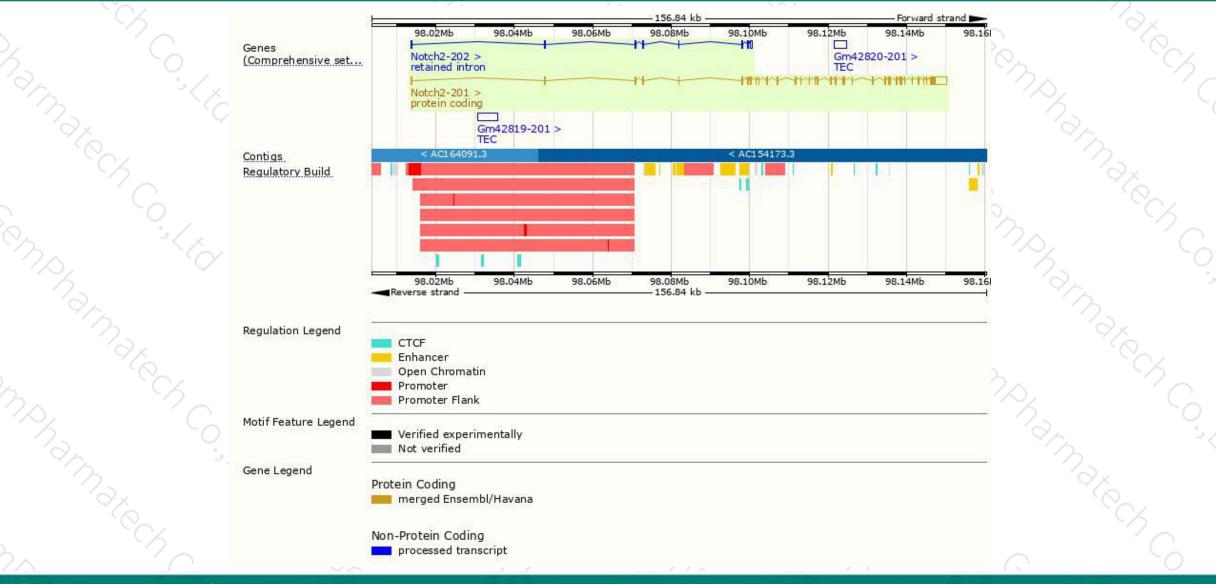
400-9660890

Forward strand

Genomic location distribution



400-9660890



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Protein domain



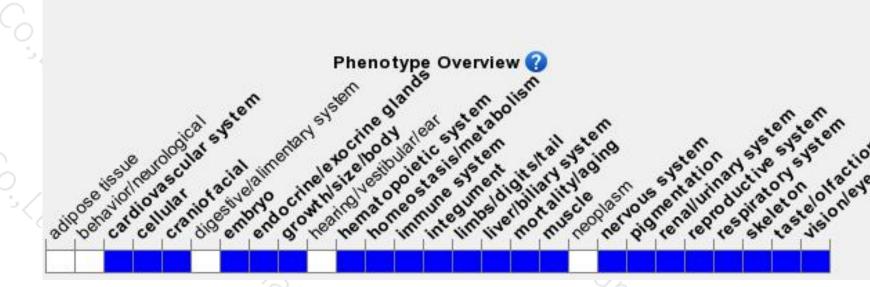
ENSMUSP0000078... Transmembrane heli... MobiDB lite . . Low complexity (Seg) Conserved Domains Cleavage site (Sign... PTHR43959 hmmpanther PTHR43959:SF3 Superfamily domains otch-like domain superfamily SSF57196 Growth factor receptor cysteine-rich domain superfamily Ankyrin repeat-containing domain su SMART domains EGF-like calcium-binding domain Doma Ankyrin repeat Notch domain Notch, NODP domain EGF-like domain Notch, NOD domain Prints domain PR01983 Neurogenic locus Notch 2 PR00010 Notch domain EGF-like domain Notch domain Pfam domain Doma Ankyrin repeat-containing domain EGF-like, conserved site EGF-like calcium-binding domain Notch, NODP domain Notch, NOD domain PROSITE profiles EGF-like domain Notch domain Ankyrin repeat-containing domain Ankyrin repeat -----PROSITE patterns -----...... GF-like calcium-binding, conserved site -----PIRSF domain Notch Gene3D 2.10.25.10 Ankyrin repeat-containing domain su 3.30.70.3310 Sequence variants (dbSNP and all other sources All sequence SNPs/i... Variant Legend missense variant splice region variant synonymous variant Scale bar 2473 400 800 1200 1600 2000

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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 alleles exhibit defects in embryonic development resulting in embryonic or neonatal lethality.



If you have any questions, you are welcome to inquire. Tel: 400-9660890



