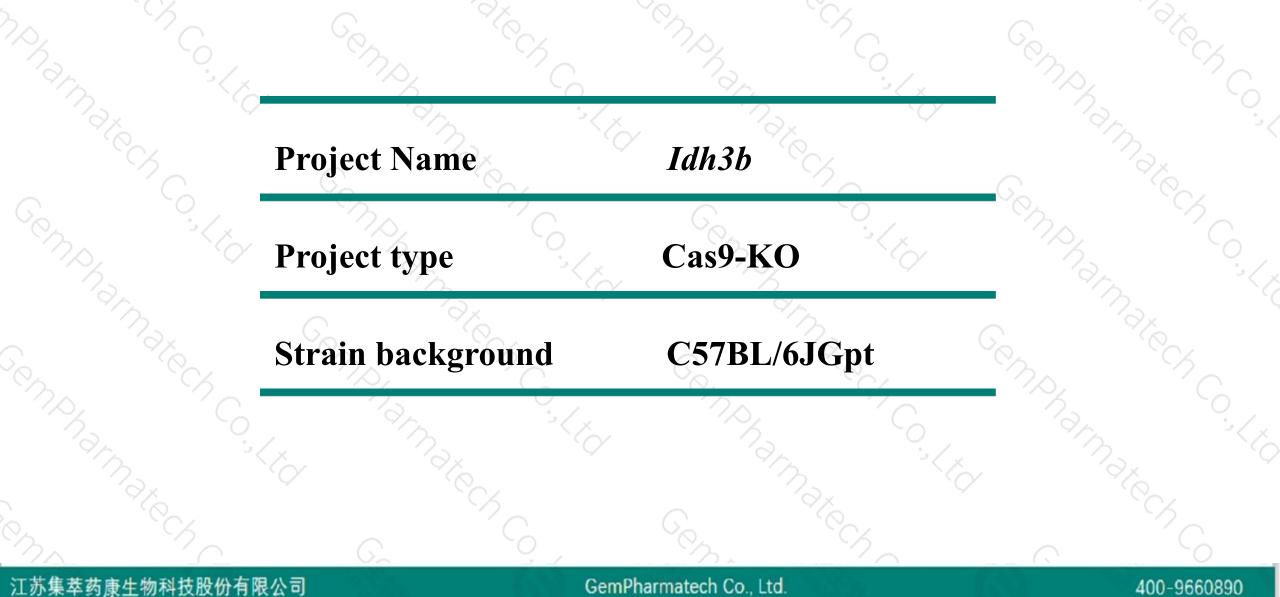


Idh3b Cas9-KO Strategy

Designer: Xueting Zhang Reviewer:Yanhua Shen Date:2020-04-07

Project Overview

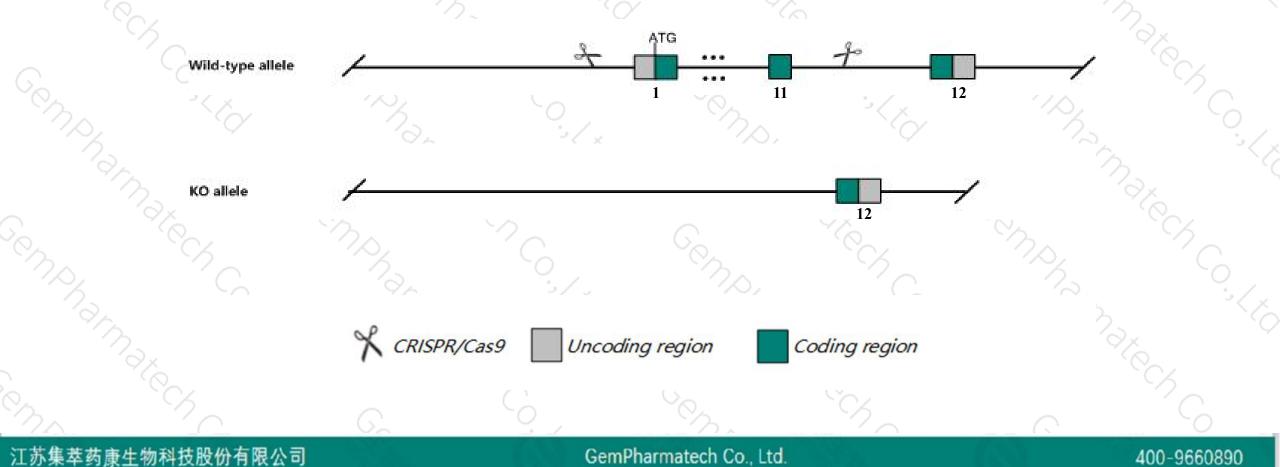




Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Idh3b* gene. The schematic diagram is as follows:





- The *Idh3b* gene has 5 transcripts. According to the structure of *Idh3b* gene, exon1-exon11 of *Idh3b-201* (ENSMUST0000028892.10) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- > In this project we use CRISPR/Cas9 technology to modify *Idh3b* gene. The brief process is as follows: CRISPR/Cas9 system

- > According to the existing MGI data, Homozygous knockout does not cause eye phenotypes.
- The knockout region is near to the C-terminal of Nop56 gene, this strategy may influence the regulatory function of the C-terminal of Nop56 gene.
- The *Idh3b* gene is located on the Chr2. 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.

Notice

Gene information (NCBI)



Summary					*
Official Symbol	Idh3b provided by MGI				
Official Full Name	isocitrate dehydrogena	ase 3 (NAD+) beta provided by MGI			
Primary source	MGI:MGI:2158650				
See related	Ensembl:ENSMUSG0	0000027406			
Gene type	protein coding				
RefSeq status	VALIDATED				
Organism	Mus musculus				
Lineage			; Mammali	a; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;	
Also known as	Muroidea; Muridae; Mu C78231	unnae; Mus; Mus			
Expression		in heart adult (RPKM 305.9), kidney adult (F	2PKM 164	3) and 28 other tissues See more	
		in near addit (in this boold), hancy addit (i			
Orthologs	human all				
Orthologs Genomic context	<u>numan an</u>				*
	<u>numan an</u>			See Idh3b in <u>Genome Data 1</u>	
Genomic context	<u>numan</u> <u>an</u>			See Idh3b in <u>Genome Data 1</u>	
Genomic context	<u>numan</u> <u>an</u> Status	Assembly	Chr	See Idh3b in <u>Genome Data 1</u>	
Genomic context Location: 2; 2 F1 Exon count: 12		Assembly GRCm38.p6 (<u>GCF_000001635.26</u>)	Chr 2		

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Transcript information (Ensembl)



The gene has 5 transcripts, all transcripts are shown below:

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Name 🖕	Transcript ID 💧	bp 🖕	Protein 🖕	Biotype 🍦	CCDS 🍦	UniProt 🖕	Flags
Idh3b-201	ENSMUST0000028892.10	1616	<u>384aa</u>	Protein coding	<u>CCDS16738</u> &	<u>Q91VA7</u> &	TSL:1 GENCODE basic APPRIS P2
Idh3b-205	ENSMUST00000239288.1	1233	<u>380aa</u>	Protein coding	-	11 <u>2</u> 1	GENCODE basic APPRIS ALT2
Idh3b-204	ENSMUST00000184538.1	452	<u>80aa</u>	Nonsense mediated decay	-	<u>V9GXV0</u> &	CDS 5' incomplete TSL:3
Idh3b-203	ENSMUST00000149843.1	2049	No protein	Retained intron	-	-	TSL:1
Idh3b-202	ENSMUST00000124868.1	620	No protein	Retained intron	-	-	TSL:3

The strategy is based on the design of *Idh3b-201* transcript, The transcription is shown below

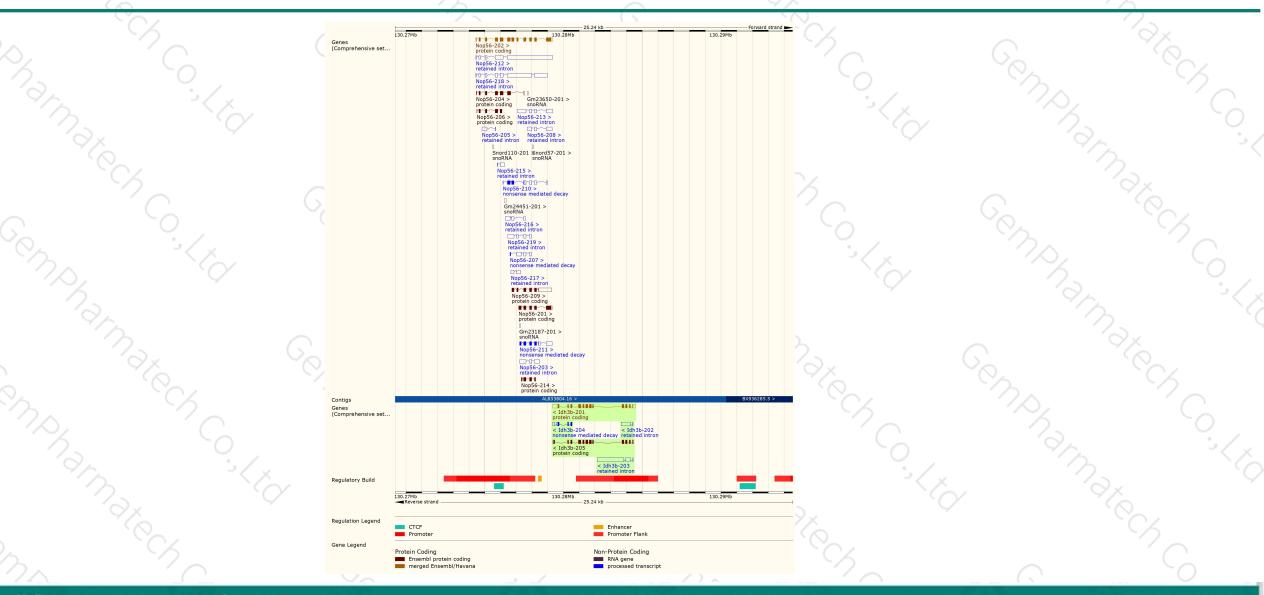


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Genomic location distribution



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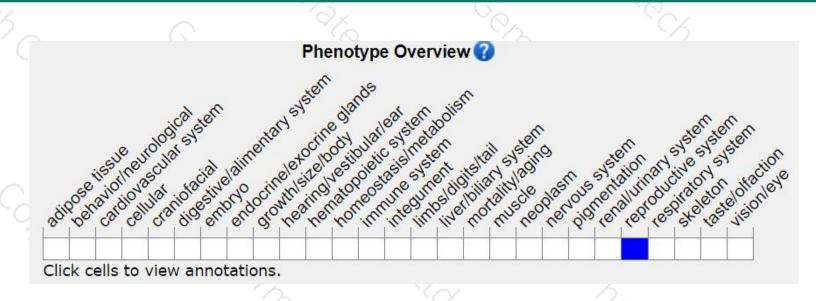
Protein domain



J. Max	ENSMUSP00000028		<u>5.</u>	3		<u></u>	Co ,	~~~~	2
d're	TIGRFAM Superfamily SMART	SSF5		ise NAD-dependent drogenase-like domain					
	Pfam PROSITE patterns PANTHER	PTHR11835	Isopropylmalate dehy	drogenase-like domain			Isocitrate/isop	propylmalate dehydro	genase, conserve
nspar,	Gene3D All sequence SNPs/i	PTHR11835:SF42 3.40.7 Sequence variants		r sources)	a cara			a.	
	All sequence SNPs/i Variant Legend	3.40.7 Sequence variants missense varia	18.10 (dbSNP and all other ant variant			splice region	variant	1	
Phan.	All sequence SNPs/i Variant Legend	3.40.7 Sequence variants	18.10 (dbSNP and all other ant	r sources)				320	384 73 73 73 73 73 74 74 74 74

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, Homozygous knockout does not cause eye phenotypes.



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



