

# Inhbe Cas9-KO Strategy

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#### Overview

#### Target Gene Name

• Inhbe

Project Type

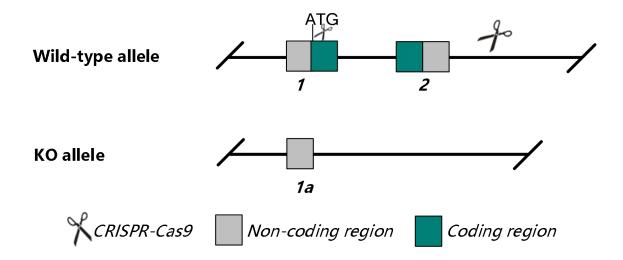
• Cas9-KO

Genetic Background

• C57BL/6JGpt



### Strain Strategy

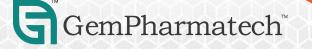


Schematic representation of CRISPR-Cas9 engineering used to edit the Inhbe gene.



#### Technical Information

- The *Inhbe* gene has 1 transcripts. According to the structure of *Inhbe* gene, exon1-2 of *Inhbe*-201 (ENSMUST0000059718.6) transcript is recommended as the knockout region. The region contains all the coding sequences. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Inhbe* 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

Inhbe inhibin beta-E [ Mus musculus (house mouse) ]

**L** Download Datasets

Gene ID: 16326, updated on 5-Aug-2022

Summary \$ ? Official Symbol Inhbe provided by MGI Official Full Name inhibin beta-E provided by MGI Primary source MGI:MGI:109269 See related Ensembl:ENSMUSG00000047492 AllianceGenome:MGI:109269 Gene type protein coding RefSeg status REVIEWED Organism Mus musculus Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae: Mus: Mus Summary This gene encodes a member of the TGF-beta (transforming growth factor-beta) superfamily of proteins. The encoded preproprotein is proteolytically processed to generate an inhibin beta subunit. Inhibins have been implicated in regulating numerous cellular processes including cell proliferation, apoptosis, immune response and hormone secretion. This gene may be upregulated under conditions of endoplasmic reticulum stress, and this protein may inhibit cellular proliferation and growth in pancreas and liver. [provided by RefSeq, Sep 2016] Expression Biased expression in liver E18 (RPKM 23.1) and liver adult (RPKM 22.2) See more Orthologs human all Try the new Gene table NEW Try the new Transcript table Genomic context \$ ? Location: 10 D3: 10 74.5 cM See Inhbe in Genome Data Viewer Exon count: 2 Annotation release Status Assembly Chr Location

GRCm39 (GCF 000001635.27) NC\_000076.7 (127185271..127187717, complement) current 10 108.20200622 GRCm38.p6 (GCF\_000001635.26) NC 000076.6 (127349402..127351848, complement) previous assembly 10 MGSCv37 (GCF 000001635.18) Build 37.2 previous assembly 10 NC\_000076.5 (126786458..126788828, complement)

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

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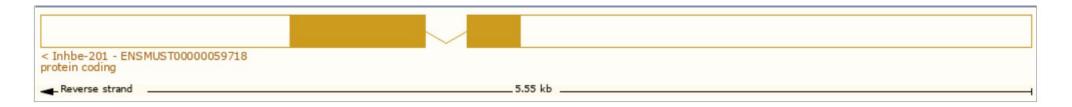
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#### Transcript Information

The gene has 1 transcript, the transcript is shown below:

Transcript ID 👙	Name 🍦	bp 🖕	Protein 🖕	Biotype 🍦	CCDS 🍦	UniProt Match	Flags			
ENSMUST0000059718.6	Inhbe-201	<mark>531</mark> 9	<u>350aa</u>	Protein coding	<u>CCDS24239</u> &	<u>008717</u> &	Ensembl Canonical	GENCODE basic	APPRIS P1	TSL:1

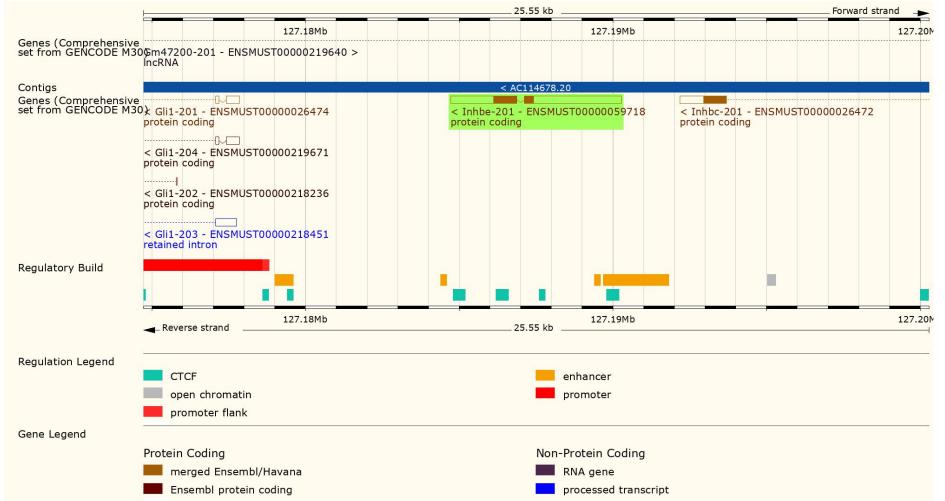
#### The strategy is based on the design of *Inhbe*-201 transcript, the transcription is shown below:



Source: https://www.ensembl.org



#### Genomic Information



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

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#### Protein Information

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ENSMUSP0000005397 Low complexity (Seg) Cleavage site (Sig AFDB-ENSP mappings Superfamily					Cystine-knot cytokin	e	
SMART					Iransforming gro	wth factor-beta, C-ter	minal
Prints Pfam	Inhibin, beta C subunit				Transforming gro	wth factor-beta, C-ter	minal
PROSITE profiles					Iransforming growth fac	tor-beta, C-terminal	
PROSITE patterns					iransfor	ming growth factor be	ta, conserved
PANTHER	PTHR11848:SF6						
19 (A)	ransforming growth factor-beta-related						
Gene3D					Cystine-knot cytokine		
CDD					cd19406		
All sequence SNPs/	Sequence variants (dbSNP and all o	ther sources)		1			1
Variant Legend	missense variant		s	ynonymous varian	t		
Scale bar	<b>b</b> 40 8	0 120	160	200	240	280	350

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

## Mouse Phenotype Information (MGI)

• Mice homozygous for a null mutation are fertile with normal liver and reproductive morphology and physiology.



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

#### Important Information

- The knockout region is close to the 5' side of the transcript Gli1 gene. The expression of Gli1 gene may be affected.
- Theknockout region is overlapped with the predictd gene Gm47200. The expression of Gm47200 gene may be affected.
- *Inhbe* is located on Chr10. 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.

