

Hint1 Cas9-KO Strategy

Designer:

Design Date:

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Reviewer:

2019-11-5

Project Overview



Project Name Hint1

Project type Cas9-KO

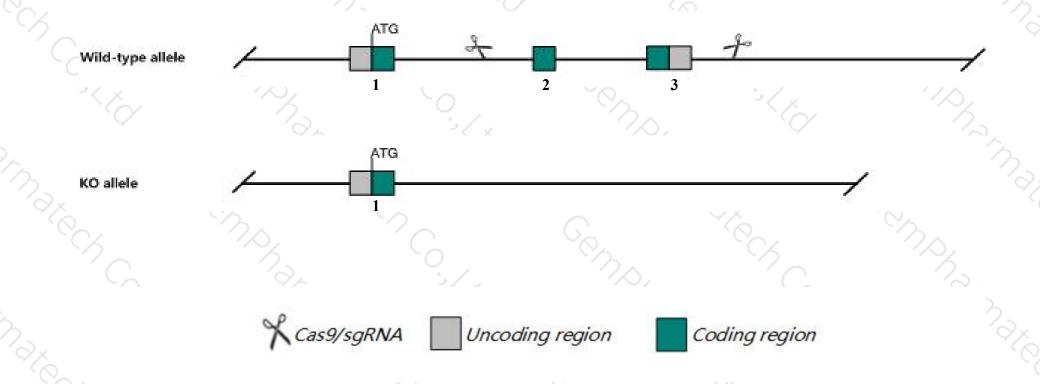
Strain background C57BL/6J

GemPharmatech Co., Ltd.

Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Hint1* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Hint1* gene has 2 transcripts. According to the structure of *Hint1* gene, exon2-exon3 of *Hint1-201* (ENSMUST00000020504.5) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Hint1* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

Notice



- ➤ According to the existing MGI data, homozygous mutant animals do not exhibit an overt phenotype, though one line of mutant mice was shown to be more susceptible to carcinogen-induced tumors than wild-type.
- The *Hint1* gene is located on the Chr11. 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.

Gene information (NCBI)



Hint1 histidine triad nucleotide binding protein 1 [Mus musculus (house mouse)]

Gene ID: 15254, updated on 3-Nov-2019



↑ ?

Official Symbol Hint1 provided by MGI

Official Full Name histidine triad nucleotide binding protein 1 provided by MGI

Primary source MGI:MGI:1321133

See related Ensembl: ENSMUSG00000020267

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 Hint; lpk1; PKCI-1; PRKCNH1; AA673479

Expression Ubiquitous expression in kidney adult (RPKM 346.0), duodenum adult (RPKM 283.7) and 28 other tissues See more

Orthologs <u>human</u> all

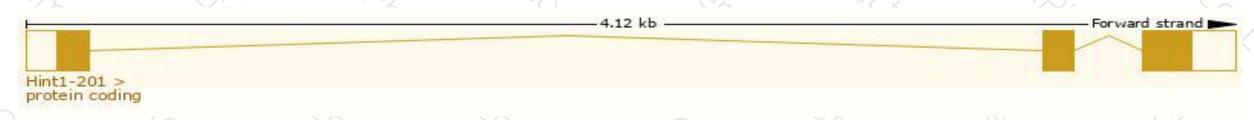
Transcript information (Ensembl)



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

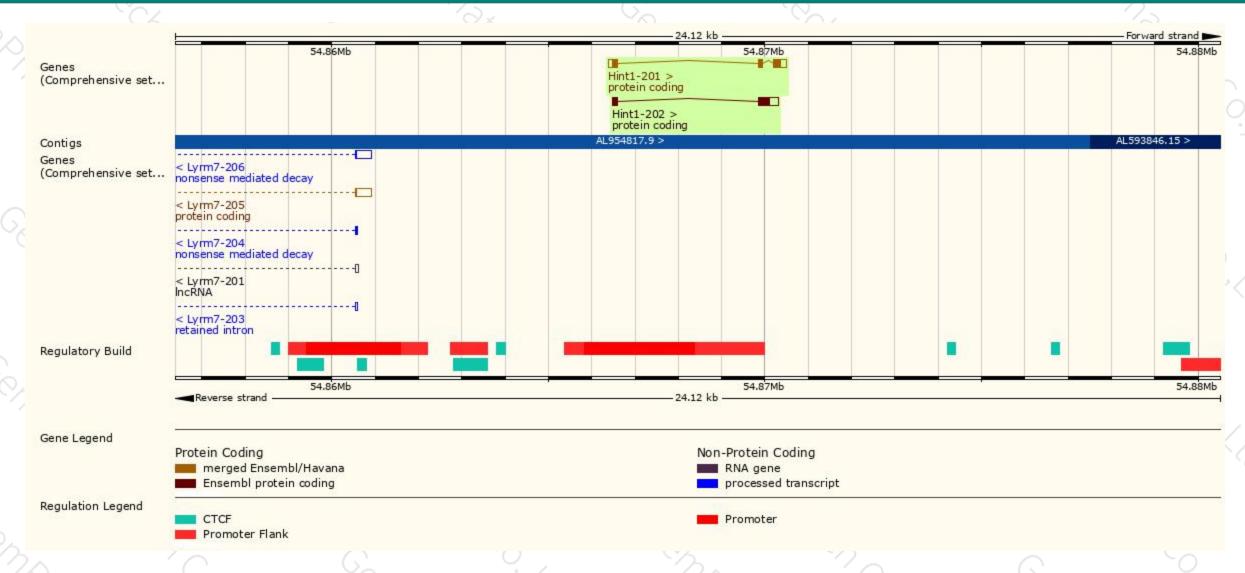
Name	Transcript ID	bp 🛊	Protein	Biotype 🍦	CCDS 🍦	UniProt	Flags		
Hint1-201	ENSMUST00000020504.5	636	<u>126aa</u>	Protein coding	CCDS24702 ₺	<u>P70349</u> ₽	TSL:1	GENCODE basic	APPRIS P1
Hint1-202	ENSMUST00000117710.1	580	<u>119aa</u>	Protein coding	-	<u>B0R1E3</u> ₽	(T	SL:2 GENCODE	basic

The strategy is based on the design of *Hint1-201* transcript, the transcription is shown below:



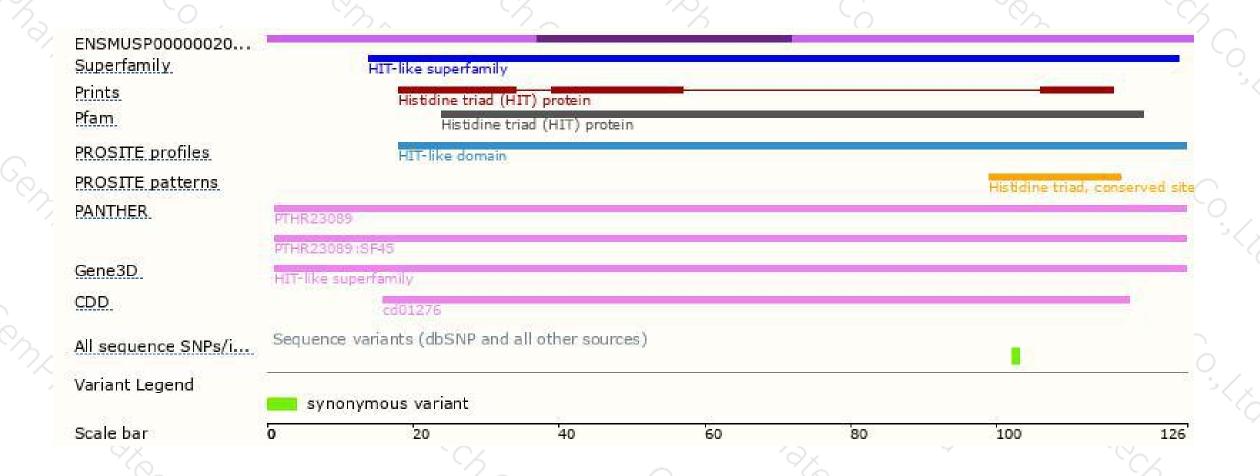
Genomic location distribution





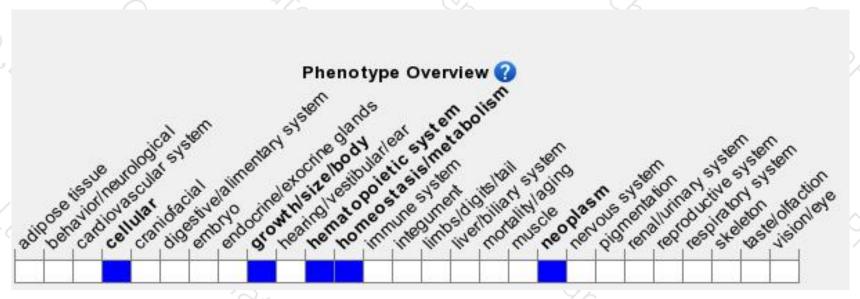
Protein domain





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 mutant animals do not exhibit an overt phenotype, though one line of mutant mice was shown to be more susceptible to carcinogen-induced tumors than wild-type.



If you have any questions, you are welcome to inquire.

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