Hspg2 Cas9-KO Strategy

Designer: Reviewer:

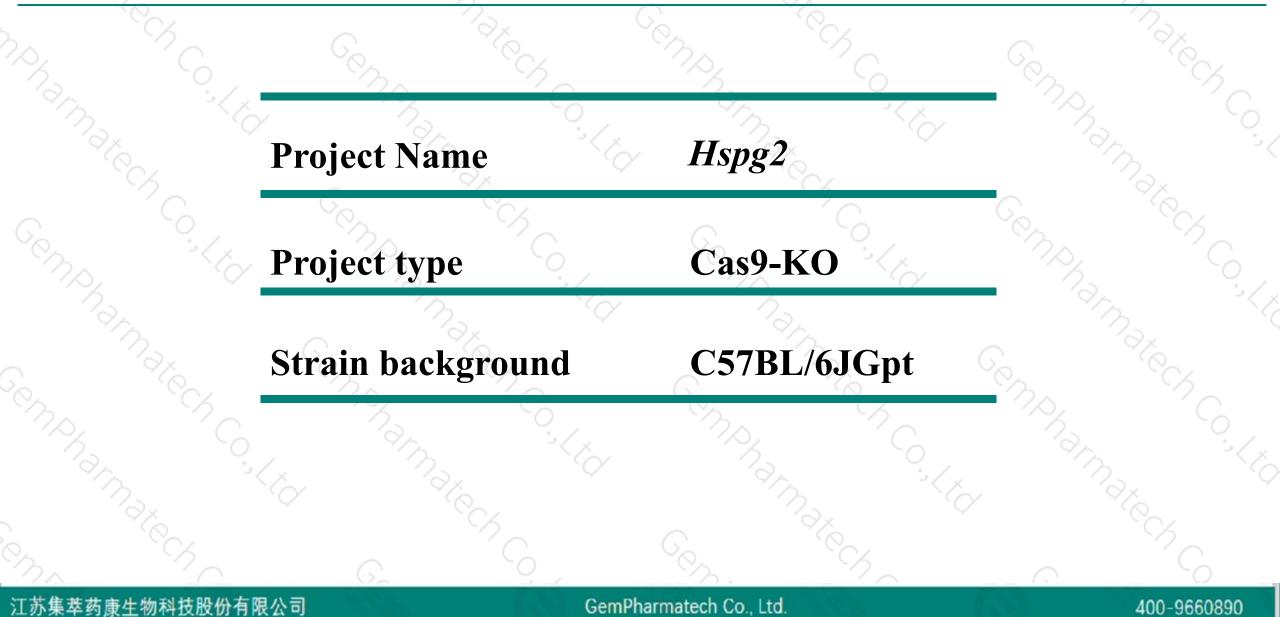
Design Date:

Longyun Hu Yun Li 2019-12-18

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Project Overview

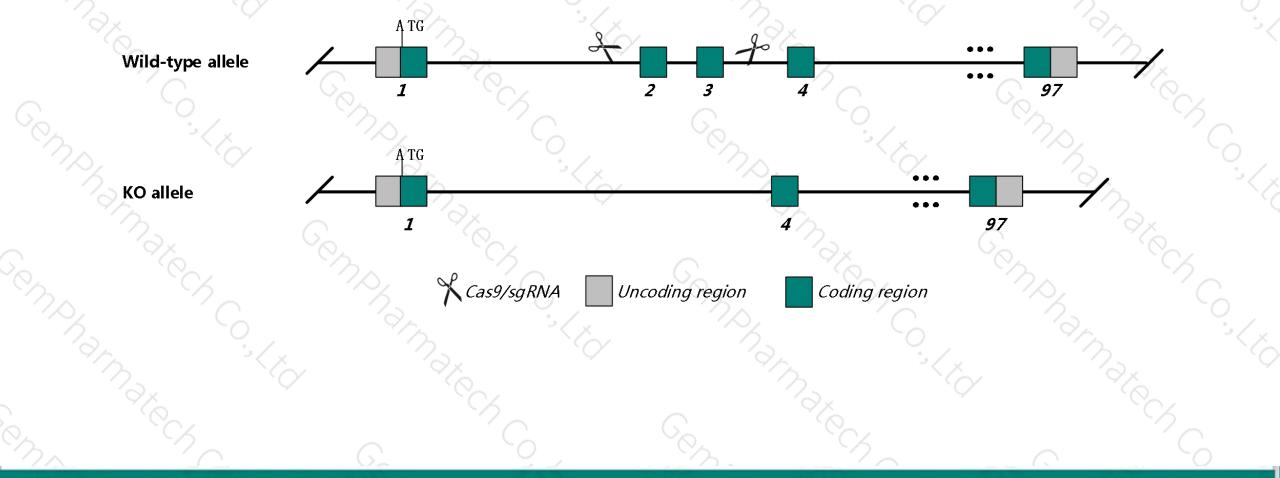




Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Hspg2* gene. The schematic diagram is as follows:



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Technical routes



The *Hspg2* gene has 3 transcripts. According to the structure of *Hspg2* gene, exon2-3 of *Hspg2*-203 transcript is recommended as the knockout region. The region contains 181bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Hspg2* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA 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.



- According to the existing MGI data, Homozygous targeted null mutants die either at embryonic day 10.5 with cardiac outflow defects and/or brain exencephaly or at birth with skeletal dysplasia including micromelia and craniofacial defects. An exon 3 deletion mutant shows only a lens defect.
- The Hspg2 gene is located on the Chr 4. 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 gene transcription and translation processes, all risks cannot be predicted under existing information.

Gene information (NCBI)



\$ 7

Hspg2 perlecan (heparan sulfate proteoglycan 2) [Mus musculus (house mouse)]

Gene ID: 15530, updated on 14-May-2019

Summary

Official Symbol	Hspg2 provided by MGI							
Official Full Name	perlecan (heparan sulfate proteoglycan 2) provided by MGI							
Primary source	MGI:MGI:96257							
See related	Ensembl:ENSMUSG0000028763							
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	Pcn; Plc; per; HSPG; Al852380							
Expression	Broad expression in adrenal adult (RPKM 35.2), subcutaneous fat pad adult (RPKM 30.9) and 20 other tissues See more							
Orthologs	human all							

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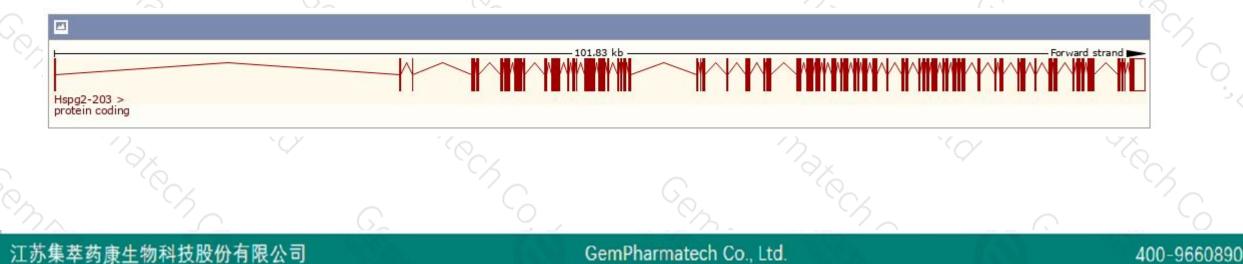
Transcript information (Ensembl)



The gene has 3 transcripts, and all transcripts are shown below :

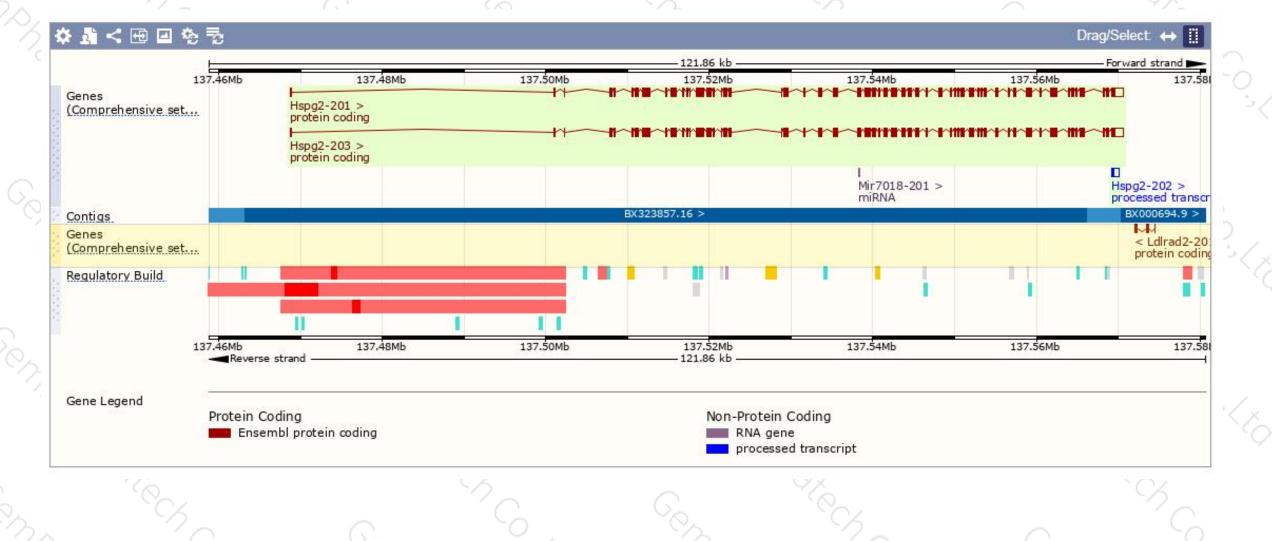
Show/hide columns (1 hidden)									
Name 💧	Transcript ID 💧	bp 🖕	Protein 💧	Biotype 💧	CCDS 💧	UniProt	Flags		
Hspg2-203	ENSMUST00000171332.1	14176	<u>4383aa</u>	Protein coding	<u>CCDS51333</u> &	E9PZ16@	TSL:5	GENCODE basic	APPRIS P2
Hspg2-201	ENSMUST0000030547.14	14187	<u>4375aa</u>	Protein coding	1.55	<u>B1B0C7</u> @	TSL:5	GENCODE basic	APPRIS ALT
Hspg2-202	ENSMUST00000155648.1	820	No protein	Processed transcript	-	-		TSL:2	

The strategy is based on the design of Hspg2-203 transcript, The transcription is shown below



Genomic location (Ensembl)



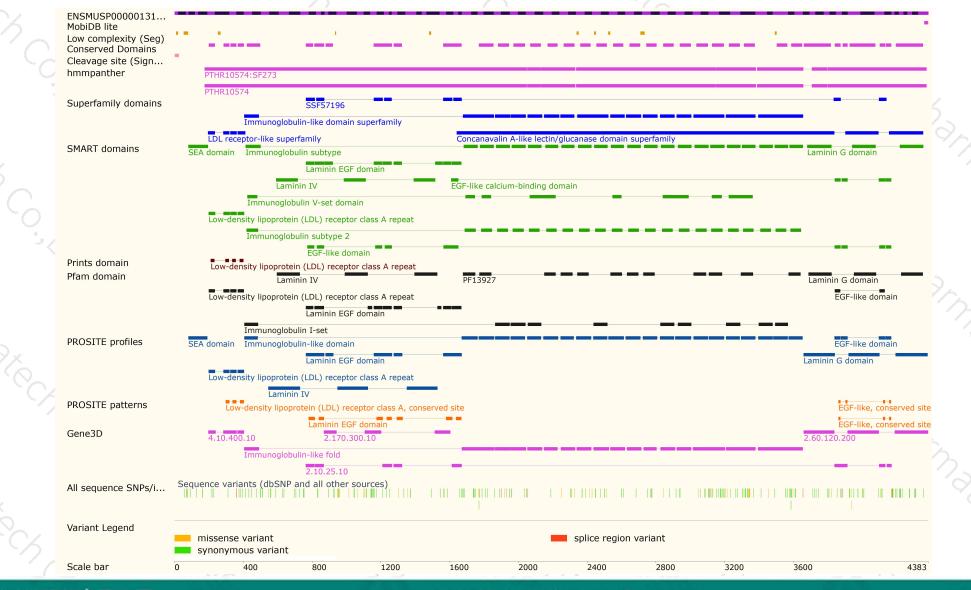


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Protein domain (Ensembl)



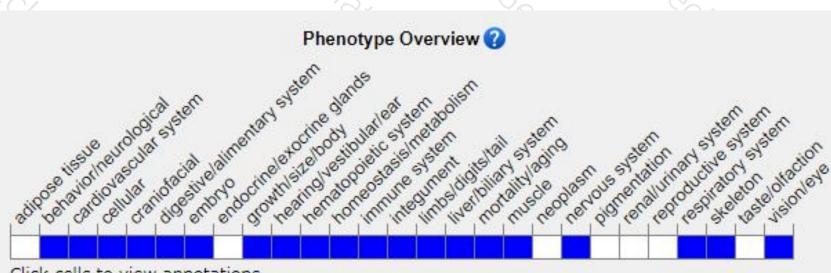


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Mouse phenotype description(MGI)





Click cells to view annotations.

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, Mutations in this locus affect cell-cycle regulation and apoptos is. Null homozygotes show high, early-onset tumor incidence; some have persistent hyaloid vasculature and cataracts. Truncated or temperature-sensitive alleles cause early aging phenotypes.

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



