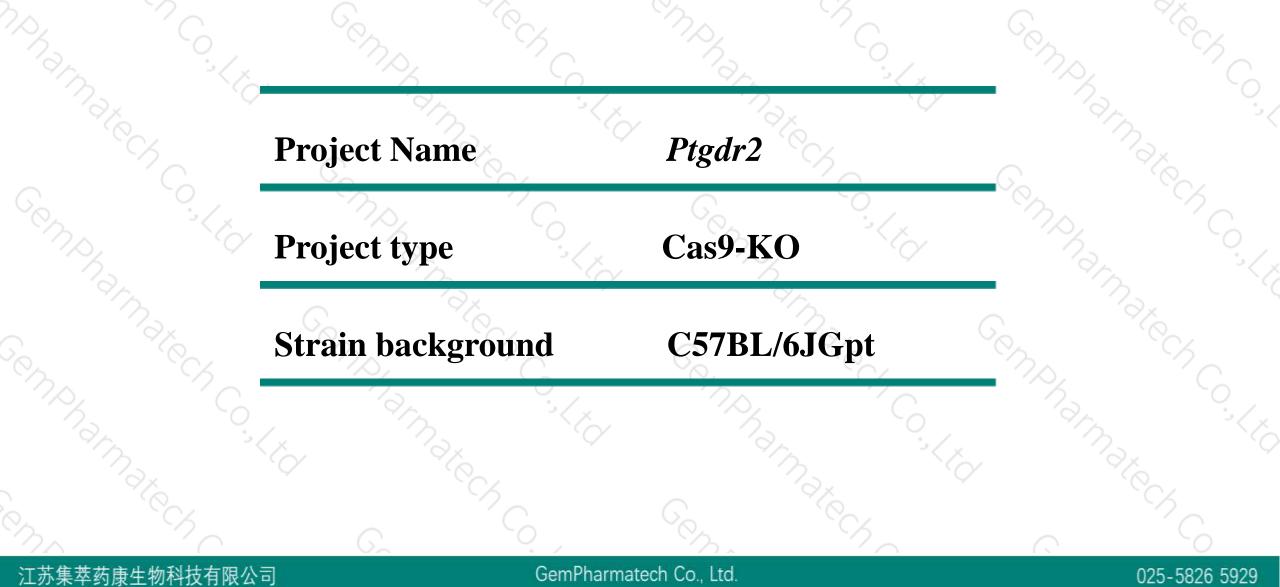


Ptgdr2 Cas9-KO Strategy

Designer: Yanhua Shen Reviewer: Xueting Zhang Design Date: 2020-3-3

Project Overview

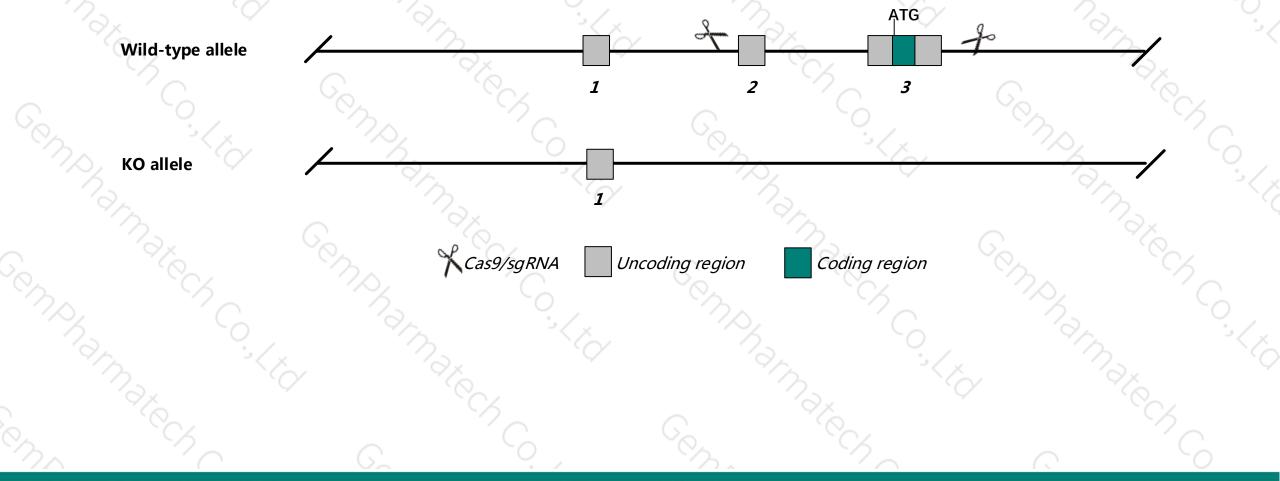






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This model will use CRISPR/Cas9 technology to edit the *Ptgdr2* gene. The schematic diagram is as follows:





- The *Ptgdr2* gene has 1 transcript. According to the structure of *Ptgdr2* gene, exon2-exon3 of *Ptgdr2-201* (ENSMUST00000037261.3) transcript is recommended as the knockout region. The region contains all 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 *Ptgdr2* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.





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- According to the existing MGI data, Mice homozygous for a knock-out allele show enhanced IL-5 production and eosinophil recruitment into the lung in an allergic airway inflammatory model. In contrast, mice homozygous for a second knock-out allele exhibit lower serum IgE levels and reduced IgE-mediated inflammatory responses in skin.
 Ccdc86 gene may be destroyed.
- ➤The *Ptgdr2* gene is located on the Chr19. 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)





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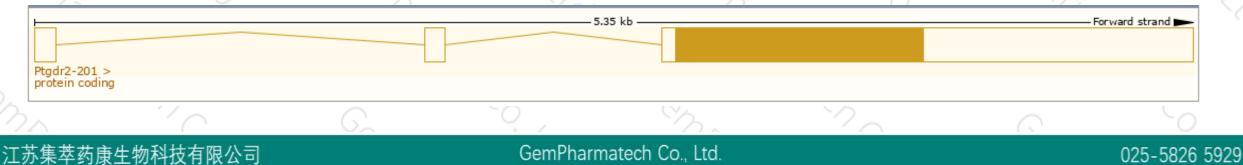
Transcript information (Ensembl)



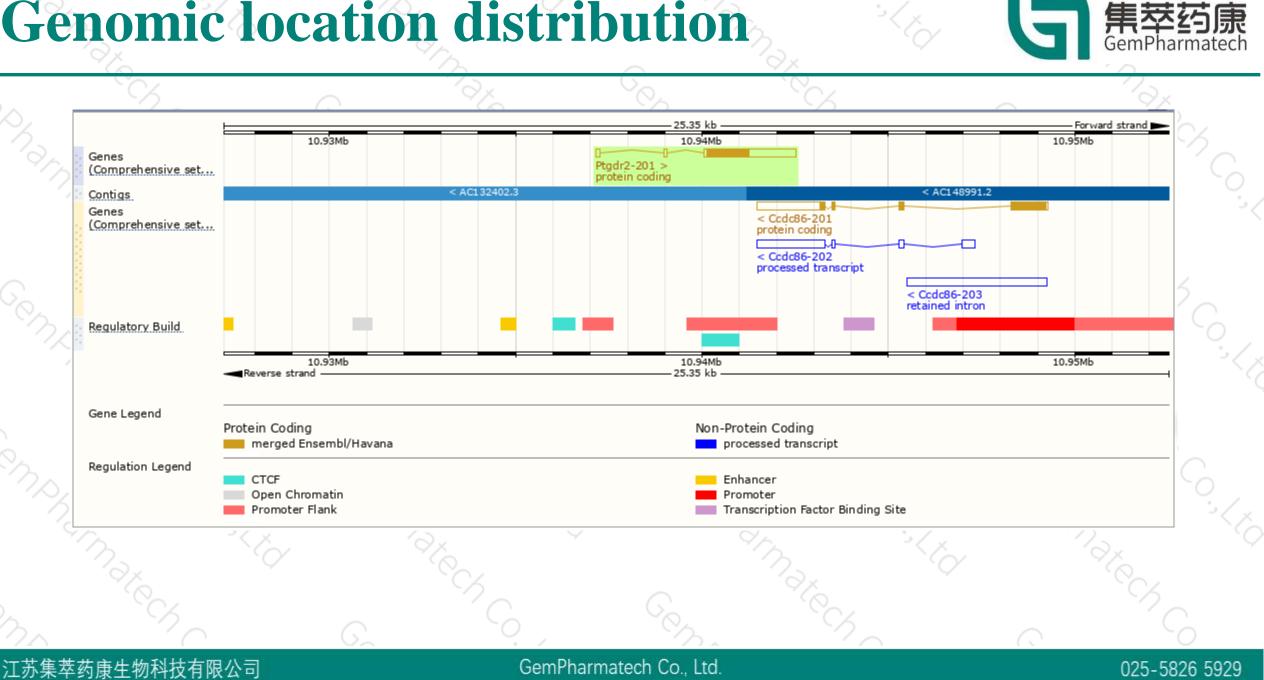
The gene has 1 transcript, all transcripts are shown below:

Na	me 🖕	Transcript ID	bp 🍦	Protein 🖕	Biotype 💧	CCDS	UniProt 🖕		Flags	4
Ptgo	dr2-201	ENSMUST0000037261.3	2644	<u>382aa</u>	Protein coding	<u>CCDS29592</u> &	<u>Q9Z2J6</u> ₽	TSL:1	GENCODE basic	APPRIS P1

The strategy is based on the design of *Ptgdr2-201* transcript, The transcription is shown below



Genomic location distribution



Protein domain

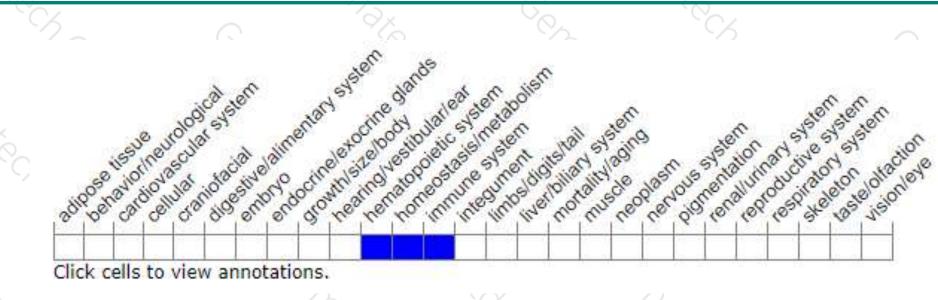


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	Low complexity (Seq) Superfamily	SSF81321							0.
	Prints	55761321	PR00526						
			-coupled receptor, rhodopsin					•	
	Pfam.	_	G protein-coupled receptor, rl	nod opsin-like					
	PROSITE profiles	5	PCR, rhodopsin-like, 7TM						5
	PROSITE patterns PANTHER			G protein-coupled r	eceptor, rhodopsin-like				í C
	LOUTIES.	PTHR24229 :SF8 PTHR24229							
	Gene3D	1.20.1070.10							~
	CDD	cd15118						-	
	All sequence SNPs/i		SNP and all other sources)		т т.		1	1.1	1
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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, Mice homozygous for a knock-out allele show enhanced IL-5 production and eosinophil recruitment into the lung in an allergic airway inflammatory model. In contrast, mice homozygous for a second knock-out allele exhibit lower serum IgE levels and reduced IgE-mediated inflammatory responses in skin.

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If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



