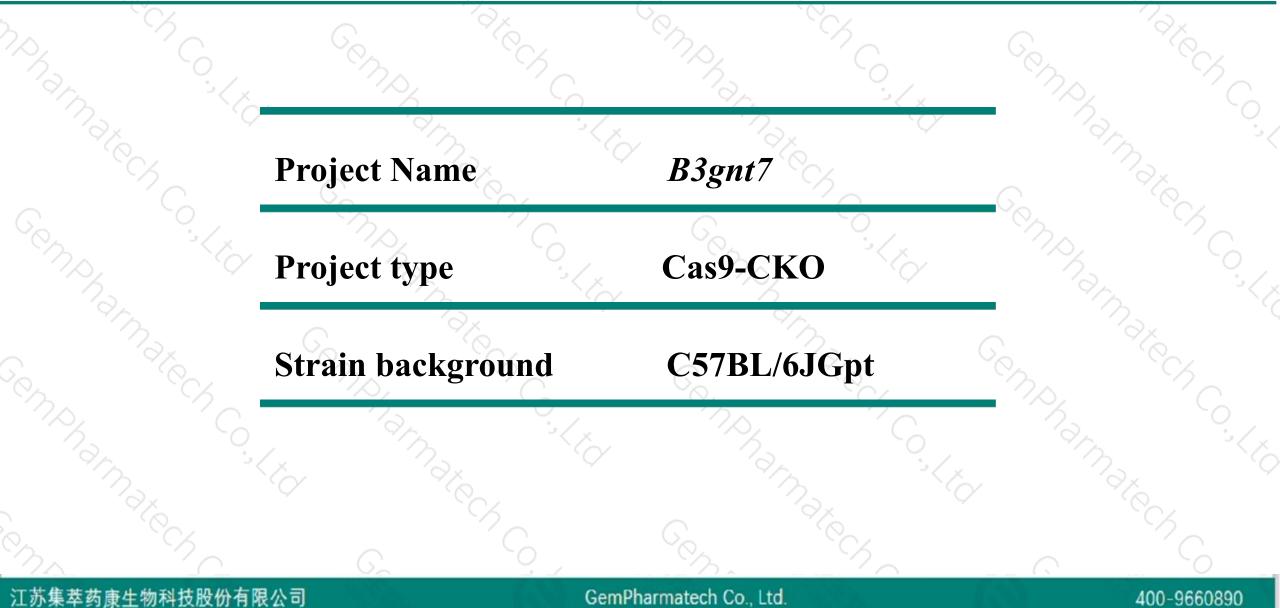


B3gnt7 Cas9-CKO Strategy

Designer: Yanhua Shen Reviewer: Xueting Zhang Design Date: 2020-4-8

Project Overview



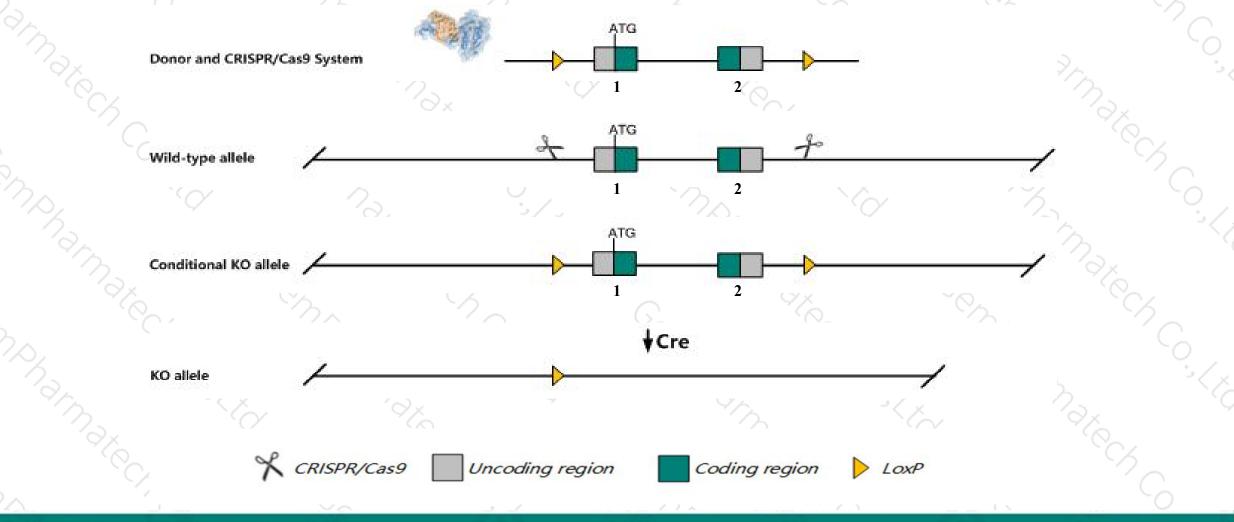


Conditional Knockout strategy



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This model will use CRISPR/Cas9 technology to edit the *B3gnt7* gene. The schematic diagram is as follows:



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The B3gnt7 gene has 3 transcripts. According to the structure of B3gnt7 gene, exon1-exon2 of B3gnt7-201 (ENSMUST00000113306.3) 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 *B3gnt7* gene. The brief process is as follows:CRISPR/Cas9 system and Donor 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.

The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.



- According to the existing MGI data, Mice homozygous for a knock-out allele exhibit decreased stroma and total cornea thickness.
- > Gm28626 and Gm16341 gene may be destroyed.
- The B3gnt7 gene is located on the Chr1. 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)



\$?

B3gnt7 UDP-GlcNAc:betaGal beta-1,3-N-acetylglucosaminyltransferase 7 [Mus musculus (house mouse)]

Gene ID: 227327, updated on 13-Mar-2020

Summary

Official Symbol	B3gnt7 provided by MGI
Official Full Name	UDP-GIcNAc:betaGal beta-1,3-N-acetylglucosaminyltransferase 7 provided by <u>MGI</u>
Primary source	MGI:MGI:2384394
See related	Ensembl:ENSMUSG0000079445
Gene type	protein coding
RefSeq status	VALIDATED
Organism	<u>Mus musculus</u>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
	Muroidea; Muridae; Murinae; Mus; Mus
Also known as	C330001H22Rik, beta-3GnT7
Expression	Biased expression in colon adult (RPKM 350.5), large intestine adult (RPKM 52.5) and 1 other tissueSee more
Orthologs	human all

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Transcript information (Ensembl)



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

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
B3gnt7-201	ENSMUST00000113306.3	2582	<u>397aa</u>	Protein coding	CCDS35645	<u>Q8K0J2</u>	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
B3gnt7-202	ENSMUST00000188695.1	1523	<u>155aa</u>	Protein coding	-	A0A087WQY3	CDS 3' incomplete TSL:1
B3gnt7-203	ENSMUST00000189618.1	1876	No protein	Retained intron	(2)	34	TSLINA

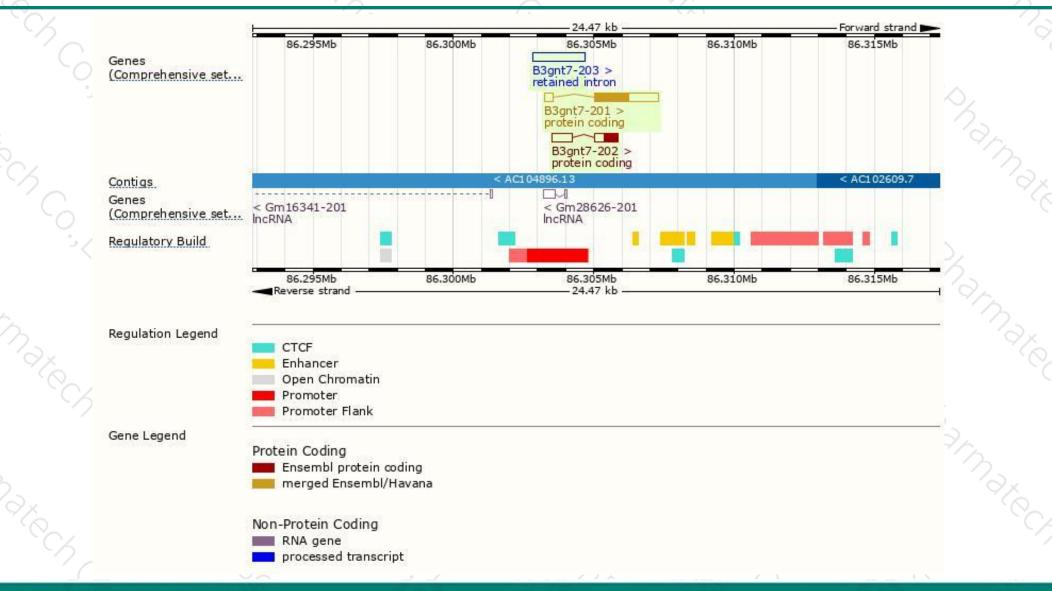
The strategy is based on the design of B3gnt7-201 transcript, The transcription is shown below

		- 4.0	07 kb	Forward strand 🗩		
B3gnt7-201 protein codin	>					
protein codin	g	100×				

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Genomic location distribution





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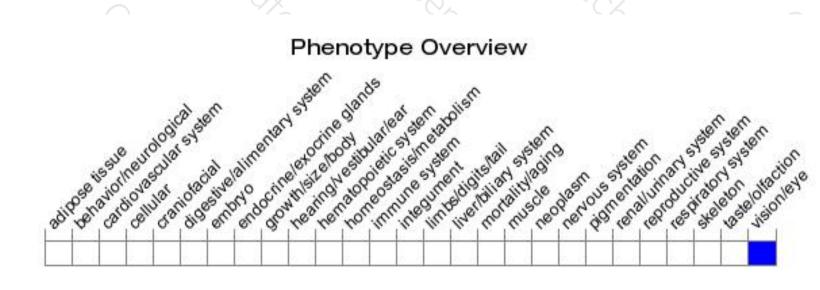
Protein domain



917			5	6		212				5.	
	ENSMUSP00000108 Transmembrane heli Low complexity (Seg) Pfam										
"Ch	PANTHER.	Glycosyl transferase, family 31									
1	Gene3D	PTHR11214:SF93									
	All sequence SNPs/i	Sequence variar	nts (dbSNP and	all other sou	arces)	1	1	0	T.	T.	
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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 exhibit decreased stroma and total cornea thickness.



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



