

B3galt2 Cas9-CKO Strategy

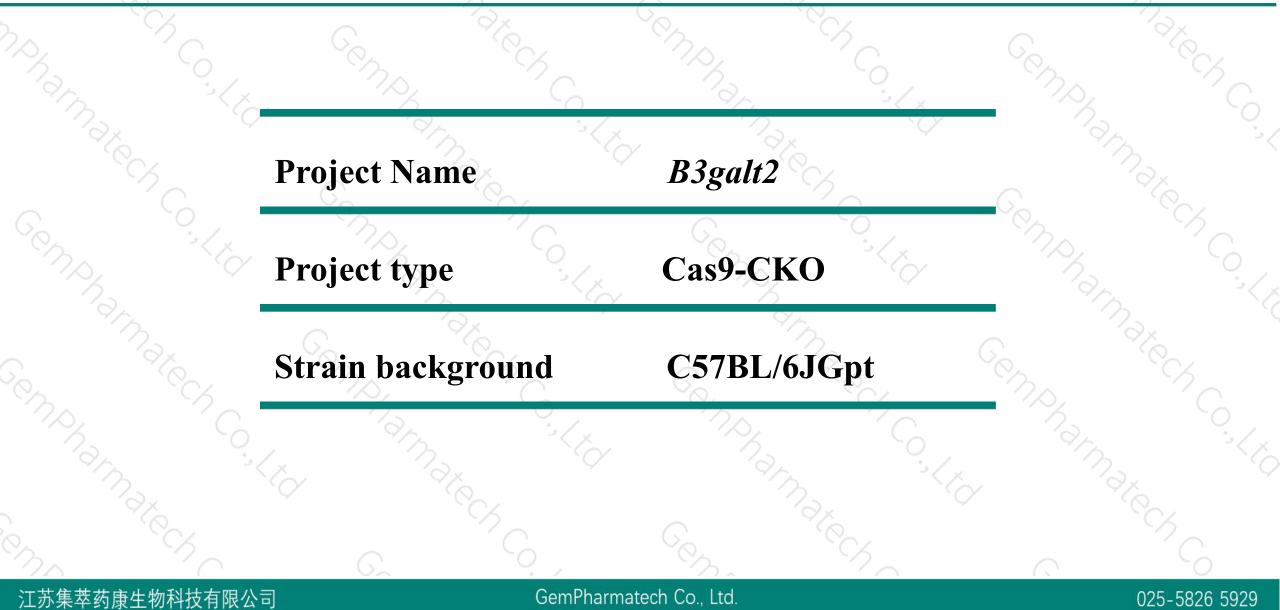
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Reviewer: JiaYu

Design Date: 2020-10-14

Project Overview

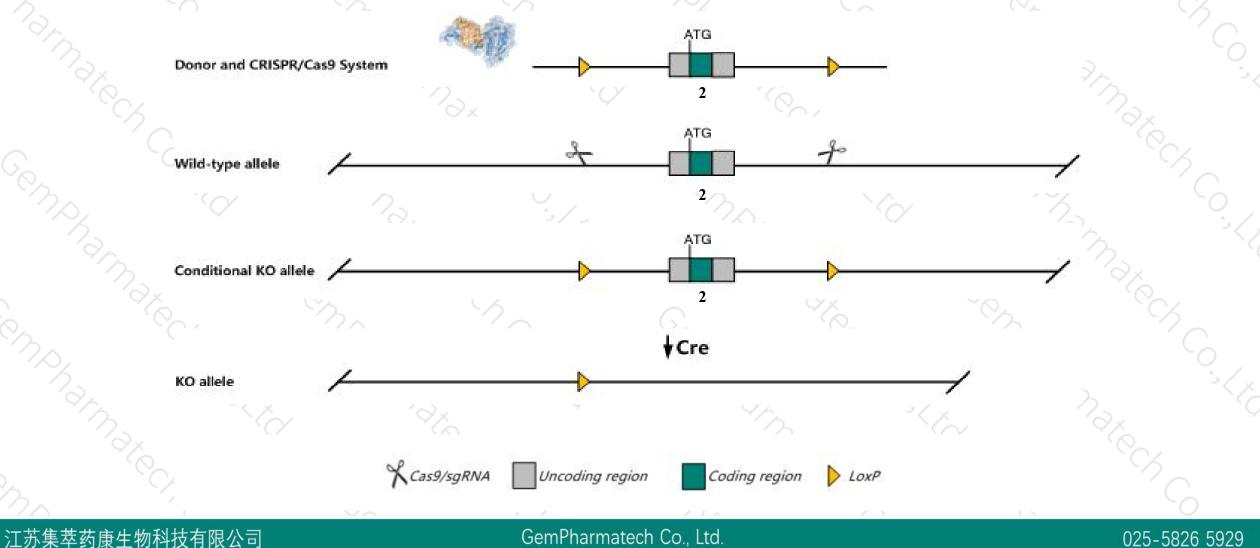




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *B3galt2* gene. The schematic diagram is as follows:





> The *B3galt2* gene has 1 transcript. According to the structure of *B3galt2* gene, exon2 of *B3galt2*-201(ENSMUST00000038252.3) 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 *B3galt2* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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, homozygous mice for a targeted mutation display hyperactivity, impaired motor coordination, decreased anxiety, increased startle reflexes, and decreased coping response.
- > The knockout region overlapped with the intron of Cdc73-201.Knockout the region may affect the function of Cdc73 gene.
- > The B3galt2 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)



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B3galt2 UDP-Gal:betaGlcNAc beta 1,3-galactosyltransferase, polypeptide 2 [Mus musculus (house mouse)]

Gene ID: 26878, updated on 13-Mar-2020

Summary

Official Symbol	B3galt2 provided by MGI
Official Full Name	UDP-Gal:betaGlcNAc beta 1,3-galactosyltransferase, polypeptide 2 provided byMGI
Primary source	MGI:MGI:1349461
See related	Ensembl:ENSMUSG0000033849
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
Expression	Biased expression in subcutaneous fat pad adult (RPKM 7.4), genital fat pad adult (RPKM 6.8) and 12 other tissues See more
Orthologs	human all

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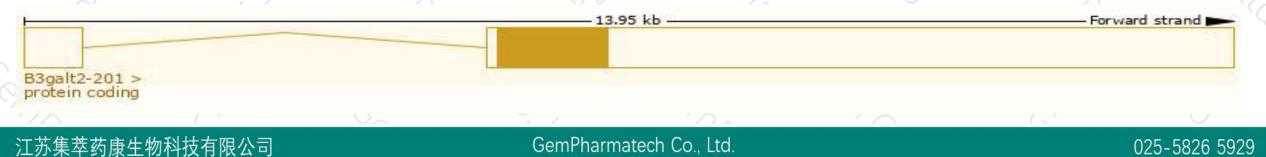
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The gene has 1 transcript, and the transcript is shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
B3galt2-201	ENSMUST0000038252.3	9288	<u>422aa</u>	Protein coding	CCDS15342	054905	TSL:1 GENCODE basic APPRIS P1
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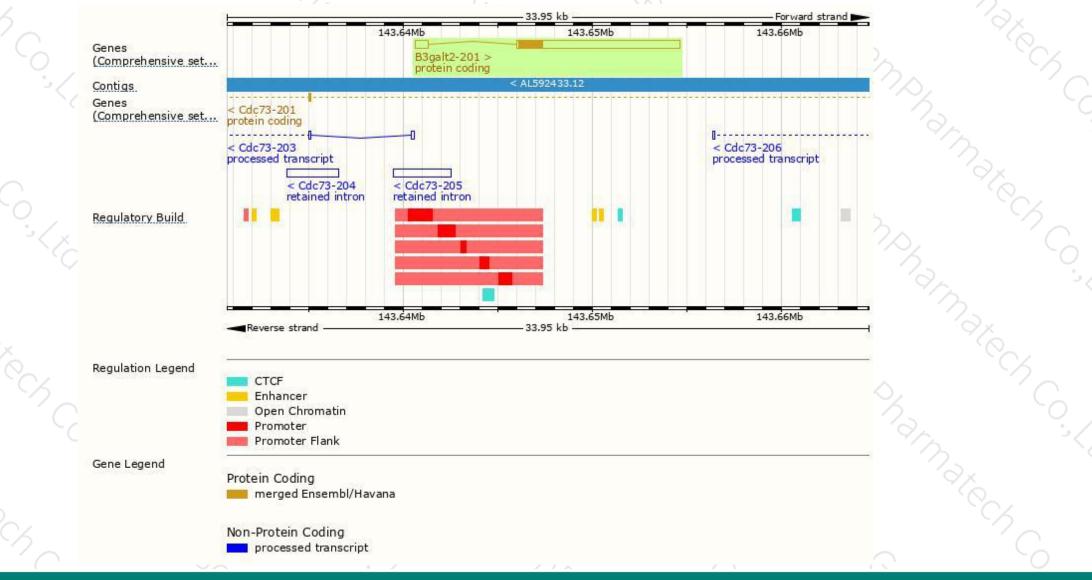
The strategy is based on the design of *B3galt2-201* transcript, the transcription is shown below:



Genomic location distribution



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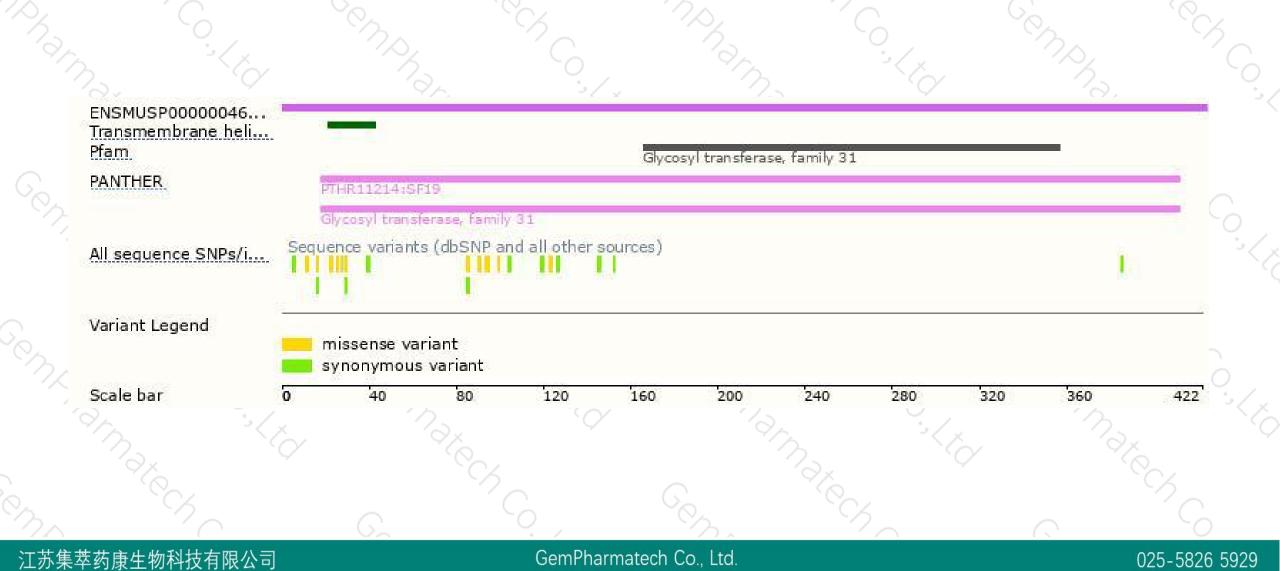


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Protein domain

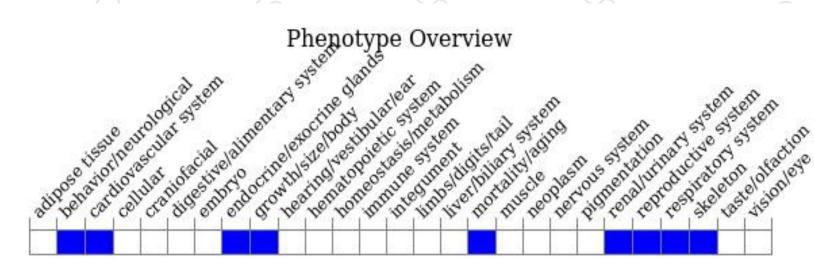




Mouse phenotype description(MGI)



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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 mice for a targeted mutation display hyperactivity, impaired motor coordination, decreased anxiety, increased startle reflexes, and decreased coping response.



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



