Galnt1 Cas9-KO Strategy

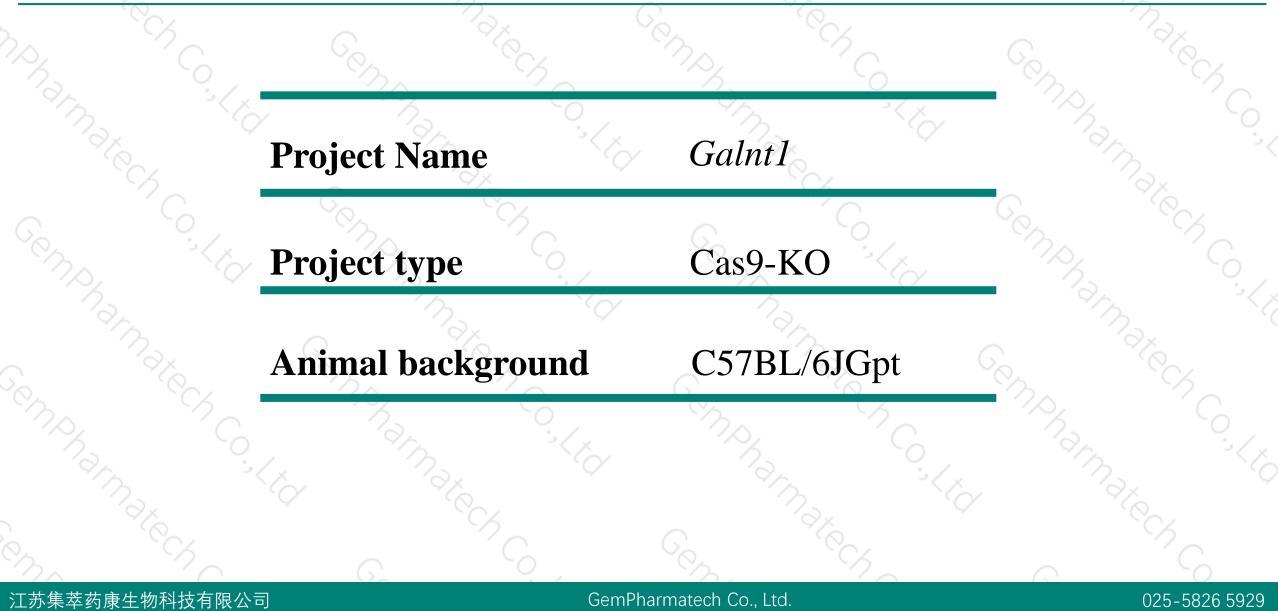
Designer: Reviewer :

Design Date:

Daohua Xu Huimin Su 2019-9-28

Project Overview

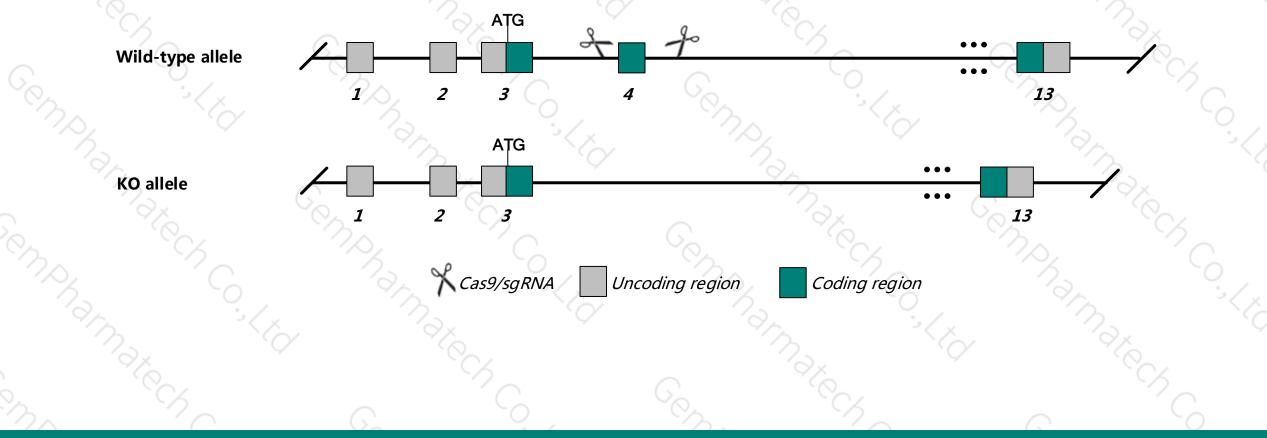




Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Galnt1 gene. The schematic diagram is as follows:



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- The *Galnt1* gene has 8 transcripts, According to the structure of *Galnt1* gene, exon4 of *Galnt1-201* transcript is recommended as the knockout region. The region contains the 175bp coding sequence. Knock out the region, result in destruction of protein.
- This project uses CRISPR/Cas9 technology to modify *Galnt1* gene. The brief process is as follows: sgRNA was transcribed in vitro, Cas9, sgRNA were microinjected into fertilized eggs of C57BL/6JGpt mice and homologous recombination was carried out to obtain F0 mice. A stable and hereditary F1 generation mouse model was obtained by mating F0 generation mice with C57BL/6JGpt mice which were confirmed positive by PCR-sequencing.



- According to the existing MGI data, Mice homozygous for a null allele exhibit some embryonic lethality, increased bleeding time, decreased T and B cells, impaired leukocyte rolling, decreased IgG levels, and hypoalbuminemia.
- The *Galnt1* gene is located in the Chr18. If the knockout mice are mixed with other mice, two target genes are avoided on the same chromosome as possible, otherwise the offspring of mice with double gene positive and homozygous gene knockout can not be obtained.
 - 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)



GaInt1 polypeptide N-acetylgalactosaminyltransferase 1 [Mus musculus (house mouse)]

Gene ID: 14423, updated on 8-Dec-2018

Summary

Official Symbol GaInt1 provided by MGI polypeptide N-acetylgalactosaminyltransferase 1 provided by MGI Official Full Name Primary source MGI:MGI:894693 See related Ensembl:ENSMUSG0000000420 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 Ubiquitous expression in liver E14 (RPKM 19.5), placenta adult (RPKM 18.4) and 28 other tissues See more Orthologs human all

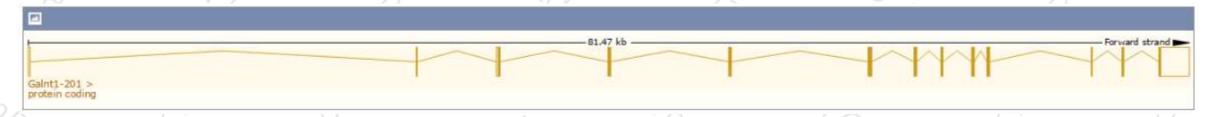
Transcript information (Ensembl)



The gene has 8 transcripts, and all transcripts are shown below :

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	Name 🍦	Transcript ID 🛛 🍦	bp 👙	Protein 🝦	Biotype 🝦	CCDS 🖕	UniProt 🖕	RefSeq 🍦	Flags	-
	Galnt1-206	ENSMUST00000170243.7	4108	<u>559aa</u>	Protein coding	<u>CCDS29100</u> ജ	<u>008912</u> മ	<u>NM 001160404</u> <u>NP 001153876</u>	TSL:1 GENCODE basic APPRIS	21
	Gaint1-201	ENSMUST0000000430.13	3977	<u>559aa</u>	Protein coding	CCDS29100@	<u>008912</u> ജ	<u>NM 001361200</u> <u>NM 013814</u> <u>NP 001348129</u> <u>NP 038842</u> ผ	TSL:1 GENCODE basic APPRIS	21
	GaInt1-208	ENSMUST00000178605.1	3796	<u>559aa</u>	Protein coding	<u>CCDS29100</u> മ	<u>008912</u> മ	-	TSL:1 GENCODE basic APPRIS	21
1	GaInt1-203	ENSMUST00000164998.7	980	<u>142aa</u>	Protein coding	-	<u>E9Q0U0</u> @	-	CDS 3' incomplete TSL:5	
	GaInt1-202	ENSMUST00000164066.1	720	<u>78aa</u>	Nonsense mediated decay	-	<u>F6PYE1</u> @	-	CDS 5' incomplete TSL:3	
	Galnt1-207	ENSMUST00000171583.1	678	<u>62aa</u>	Nonsense mediated decay	-	<u>E9PVK6</u> @	-	TSL:3	
	GaInt1-204	ENSMUST00000169474.1	818	No protein	Processed transcript	-	-	-	TSL:3	
	Galnt1-205	ENSMUST00000170191.1	798	No protein	Processed transcript	-	-	-	TSL:5	

The strategy is based on the design of Galnt1-201 transcript, The transcription is shown below :

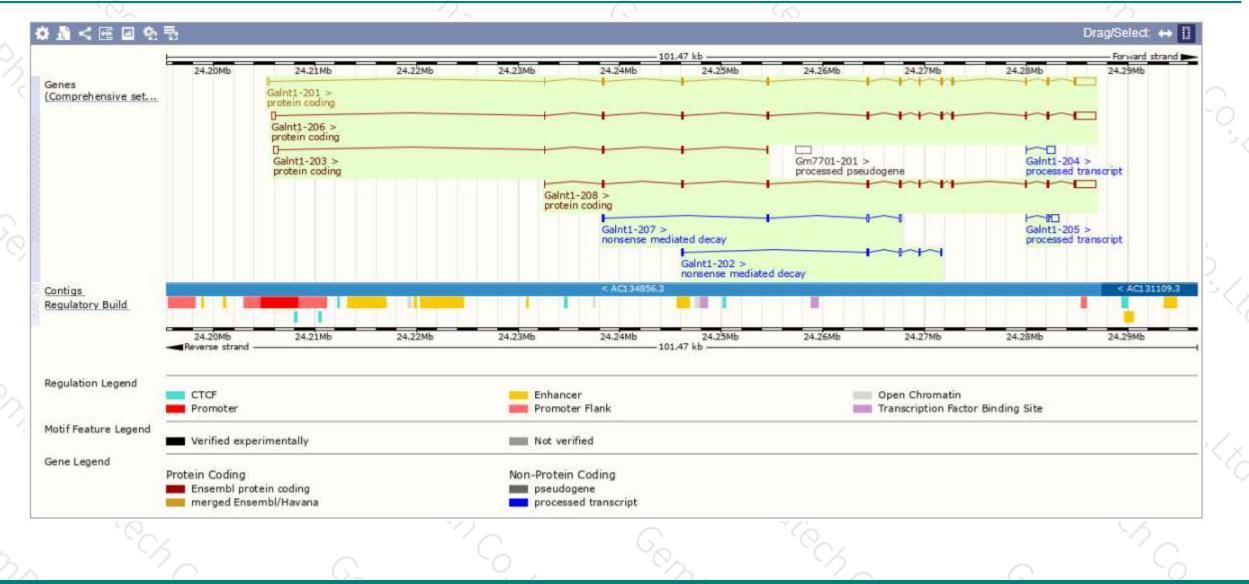


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Genomic location distribution





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Protein domain



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ART domains						Ricin B, lectin domain	n :	-
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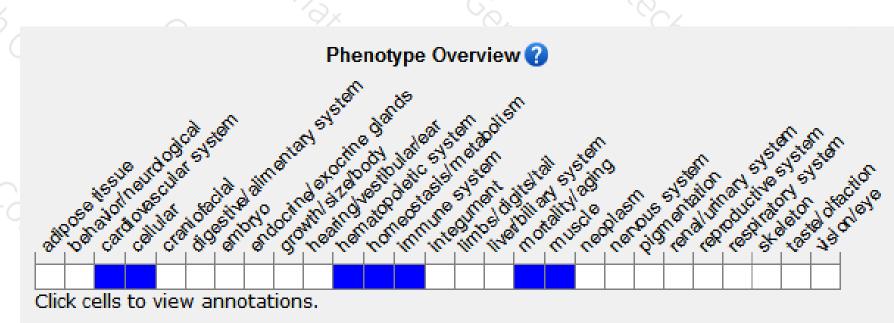
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Mouse phenotype description(MGI)



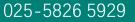


Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data, Mice homozygous for a null allele exhibit some embryonic lethality, increased bleeding time, decreased T and B cells, impaired leukocyte rolling, decreased IgG levels, and hypoalbuminemia.

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





