

St6galnac6 Cas9-CKO Strategy

Designer:

Reviewer:

Design Date:

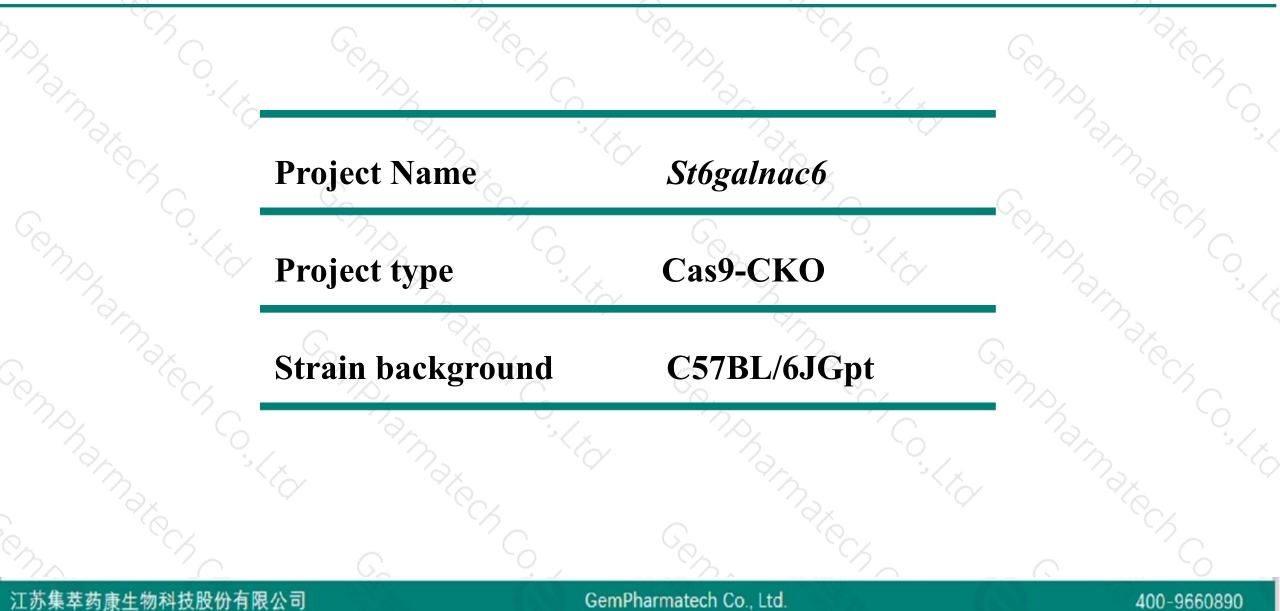
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2020-4-14

Project Overview



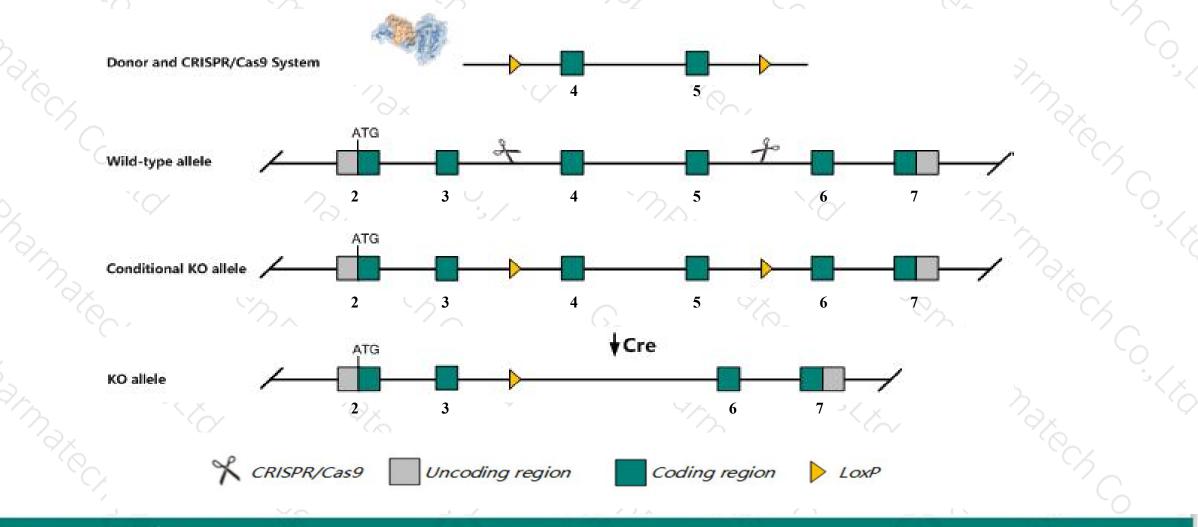


Conditional Knockout strategy



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This model will use CRISPR/Cas9 technology to edit the *St6galnac6* gene. The schematic diagram is as follows:



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- The St6galnac6 gene has 13 transcripts. According to the structure of St6galnac6 gene, exon4-exon5 of St6galnac6-203 (ENSMUST00000095044.9) transcript is recommended as the knockout region. The region contains 587bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *St6galnac6* 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.



- > Some amino acids will remain at the N-terminus and some functions may be retained.
- ➤ The effect of transcripts 206,207,208,210,211 is unknown.
- The *St6galnac6* gene is located on the Chr2. 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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St6galnac6 ST6 (alpha-N-acetyl-neuraminyl-2,3-beta-galactosyl-1,3)-N-acetylgalactosaminide alpha-2,6sialyltransferase 6 [Mus musculus (house mouse)]

Gene ID: 50935, updated on 13-Mar-2020

Summary

| Official Symbol | St6galnac6 provided by MGI |
|--------------------|--|
| Official Full Name | ST6 (alpha-N-acetyl-neuraminyl-2,3-beta-galactosyl-1,3)-N-acetylgalactosaminide alpha-2,6-sialyltransferase 6 provided by MGI |
| Primary source | MGI:MGI:1355316 |
| See related | Ensembl:ENSMUSG0000026811 |
| 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 | ST6GalNAcVI, Siat7f |
| Expression | Biased expression in colon adult (RPKM 299.0), adrenal adult (RPKM 26.2) and 6 other tissues See more |
| Orthologs | human all |

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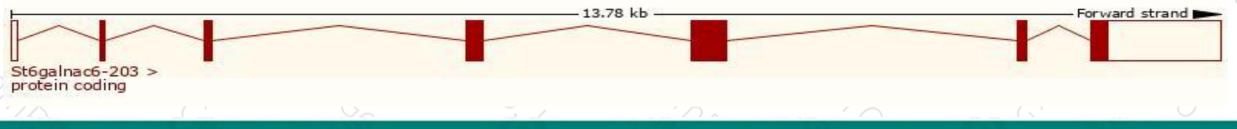
Transcript information (Ensembl)



The gene has 13 transcripts, all transcripts are shown below:

| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|----------------|----------------------|------|--------------|-------------------------|-----------|------------|--|
| St6galnac6-204 | ENSMUST0000095045.8 | 2536 | <u>299aa</u> | Protein coding | CCDS38105 | E9PUI0 | TSL1 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 ALT |
| St6galnac6-205 | ENSMUST00000113290.7 | 2402 | <u>299aa</u> | Protein coding | CCDS38105 | E9PUI0 | TSL:5 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 ALT |
| St6galnac6-203 | ENSMUST0000095044.9 | 2394 | <u>333aa</u> | Protein coding | CCDS15923 | Q9JM95 | 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 P3 |
| St6galnac6-201 | ENSMUST00000072111.7 | 2277 | <u>333aa</u> | Protein coding | CCDS15923 | Q9JM95 | TSL:5 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 P3 |
| St6galnac6-209 | ENSMUST00000131229.7 | 1013 | <u>313aa</u> | Protein coding | CCDS15922 | Z4YLR0 | TSL:2 GENCODE basic |
| St6galnac6-202 | ENSMUST0000081879.11 | 2345 | <u>335aa</u> | Protein coding | - | Q9JM95 | TSL:1 GENCODE basic |
| St6galnac6-210 | ENSMUST00000140983.7 | 875 | <u>229aa</u> | Protein coding | 2 | Z4YLN2 | CDS 3' incomplete TSL:5 |
| St6galnac6-207 | ENSMUST00000128811.1 | 584 | <u>108aa</u> | Protein coding | 2 | Z4YMI1 | CDS 3' incomplete TSL:2 |
| St6galnac6-206 | ENSMUST00000126636.7 | 438 | <u>28aa</u> | Protein coding | | A0A0A6YW64 | CDS 3' incomplete TSL:2 |
| St6galnac6-211 | ENSMUST00000143625.1 | 414 | <u>41aa</u> | Protein coding | - | A0A0A6YW84 | CDS 3' incomplete TSL:2 |
| St6galnac6-208 | ENSMUST00000129165.1 | 355 | <u>39aa</u> | Protein coding | 2 | Z4YN31 | CDS 3' incomplete TSL:2 |
| St6galnac6-213 | ENSMUST00000183538.7 | 2361 | <u>235aa</u> | Nonsense mediated decay | - | Q9JM95 | TSL:1 |
| St6galnac6-212 | ENSMUST00000149220.1 | 2322 | No protein | Retained intron | | | TSL:1 |

The strategy is based on the design of *St6galnac6-203* transcript, The transcription is shown below



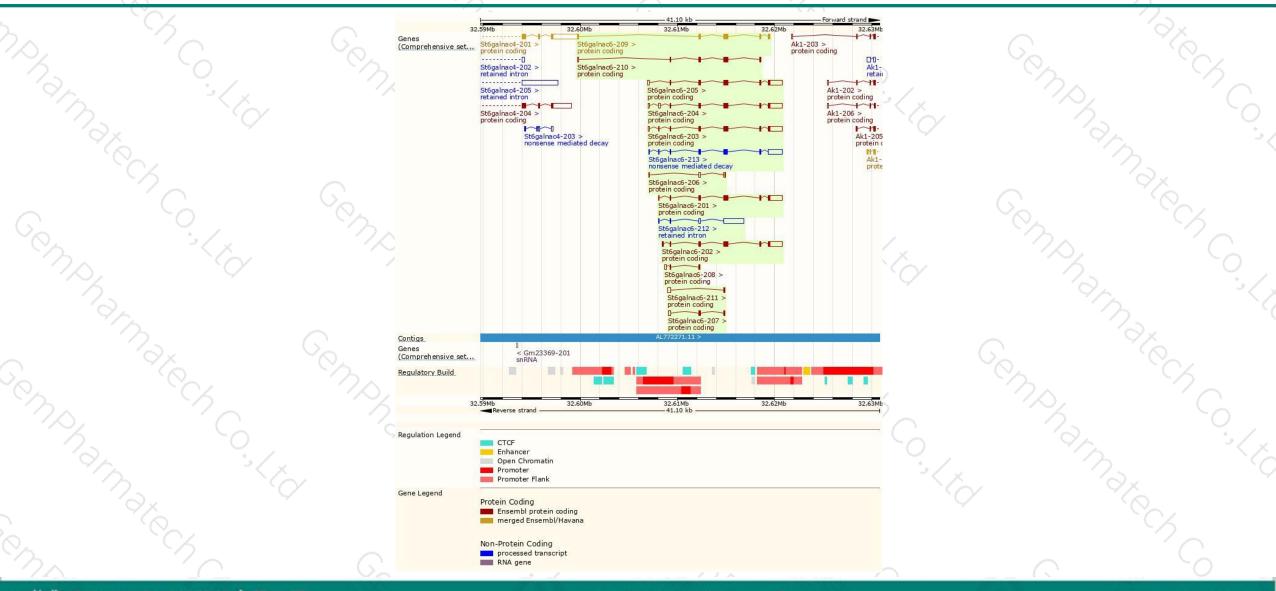
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Genomic location distribution





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Protein domain

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



