Mgat4a Cas9-CKO Strategy

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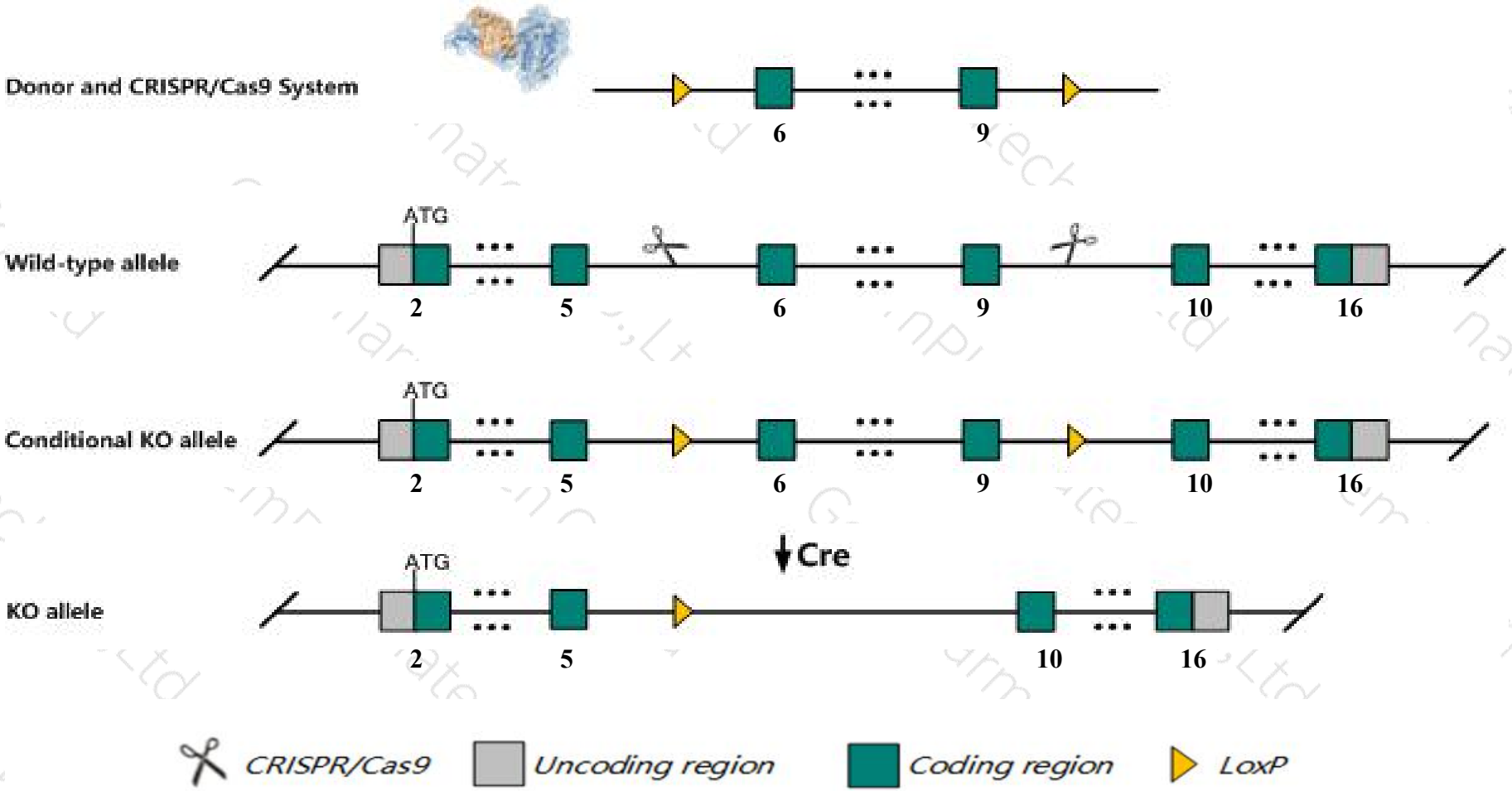
Project Overview

Project Name	***Mgat4a***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Mgat4a* gene. The schematic diagram is as follows:



Technical routes

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- The *Mgat4a* gene has 8 transcripts. According to the structure of *Mgat4a* gene, exon6-exon9 of *Mgat4a-205* (ENSMUST00000151952.7) transcript is recommended as the knockout region.The region contains 352bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Mgat4a* 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.

Notice

- According to the existing MGI data,Mice homozygous for a knock-out allele show defects in glucose-stimulated insulin secretion, impaired cellular glucose import, increased susceptibility to weight gain, hyperglycemia, impaired glucose tolerance, insulin resistance, high free fatty acid and triglyceride levels, and hepatic steatosis.
- Some amino acids will remain at the N-terminus and some functions may be retained.
- Transcripts 207,208 may not be affected. The effect of transcript 203 is unknown.
- The *Mgat4a* 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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Mgat4a mannoside acetylglucosaminyltransferase 4, isoenzyme A [*Mus musculus* (house mouse)]

Gene ID: 269181, updated on 10-Oct-2019

Summary

Official Symbol	Mgat4a provided by MGI
Official Full Name	mannoside acetylglucosaminyltransferase 4, isoenzyme A provided by MGI
Primary source	MGI:MGI:2662992
See related	Ensembl:ENSMUSG00000026110
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	GnT-IVa; glcNAc-T-IVa; 9530018I07Rik
Expression	Broad expression in large intestine adult (RPKM 27.4), colon adult (RPKM 23.5) and 20 other tissues See more
Orthologs	human all

Genomic context

Location: 1; 1 B

See Mgat4a in Genome Data Viewer

Exon count: 17

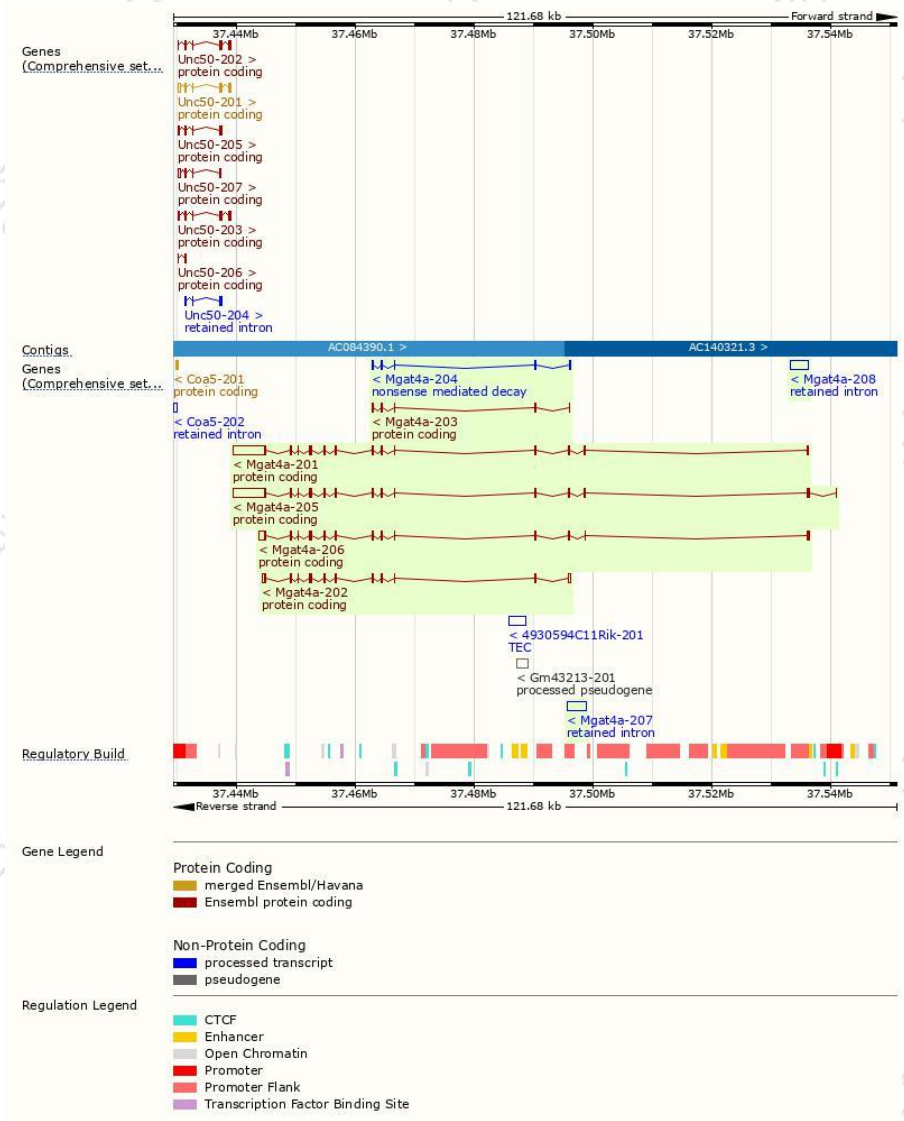
Transcript information（Ensembl）

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

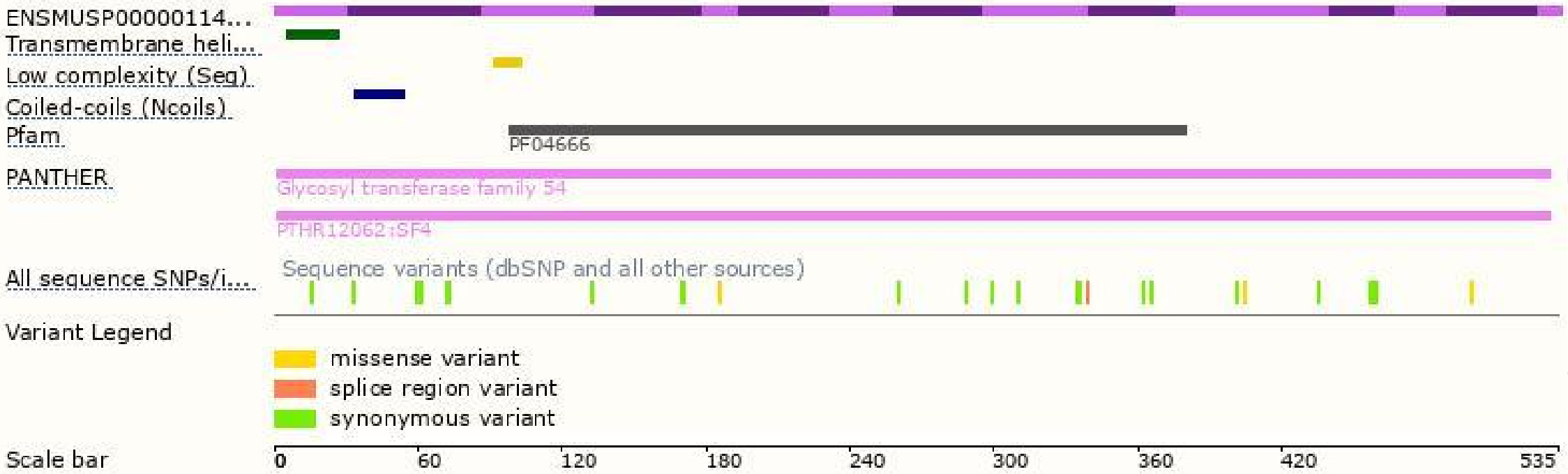
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Mgat4a-205	ENSMUST00000151952.7	7251	535aa	Protein coding	CCDS14893	Q812G0	TSL:1 GENCODE basic APPRIS P1
Mgat4a-201	ENSMUST00000042161.14	7030	535aa	Protein coding	CCDS14893	Q812G0	TSL:1 GENCODE basic APPRIS P1
Mgat4a-206	ENSMUST00000154819.7	2796	526aa	Protein coding	CCDS69882	Q812G0	TSL:1 GENCODE basic
Mgat4a-202	ENSMUST00000143636.7	1802	397aa	Protein coding	-	D3Z166	TSL:1 GENCODE basic
Mgat4a-203	ENSMUST00000148047.1	387	112aa	Protein coding	-	D3YZH7	CDS 3' incomplete TSL:3
Mgat4a-204	ENSMUST00000149791.7	424	67aa	Nonsense mediated decay	-	D6RG46	TSL:5
Mgat4a-207	ENSMUST00000194660.1	3113	No protein	Retained intron	-	-	TSL:NA
Mgat4a-208	ENSMUST00000195598.1	3039	No protein	Retained intron	-	-	TSL:NA

The strategy is based on the design of *Mgat4a-205* transcript,The transcription is shown below

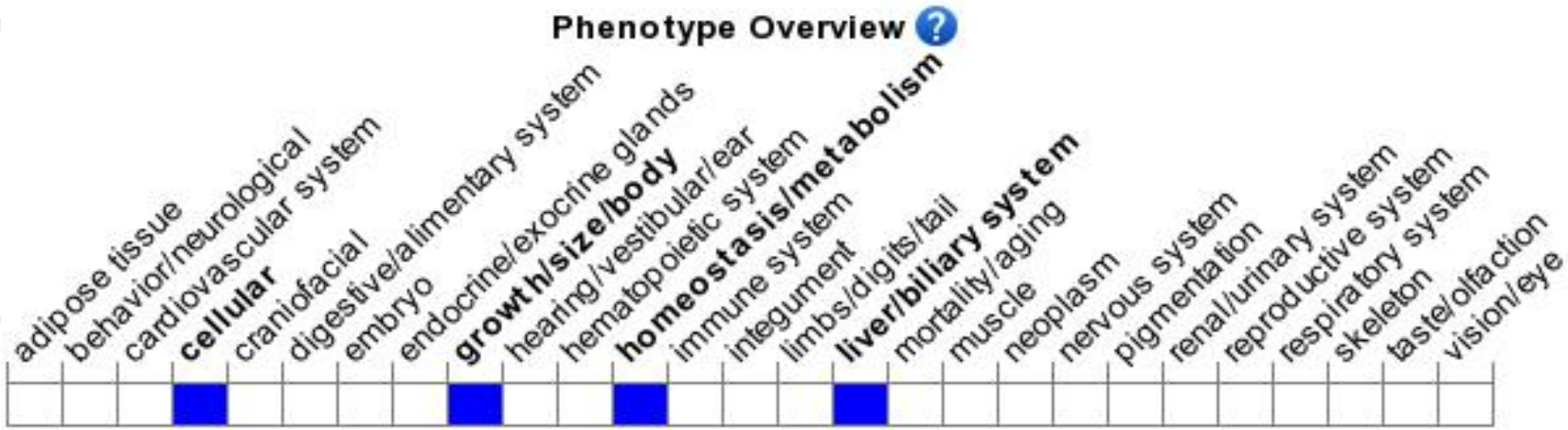
Genomic location distribution



Protein domain



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 defects in glucose-stimulated insulin secretion, impaired cellular glucose import, increased susceptibility to weight gain, hyperglycemia, impaired glucose tolerance, insulin resistance, high free fatty acid and triglyceride levels, and hepatic steatosis.

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If you have any questions, you are welcome to inquire.

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