

Abca7 Cas9-CKO Strategy

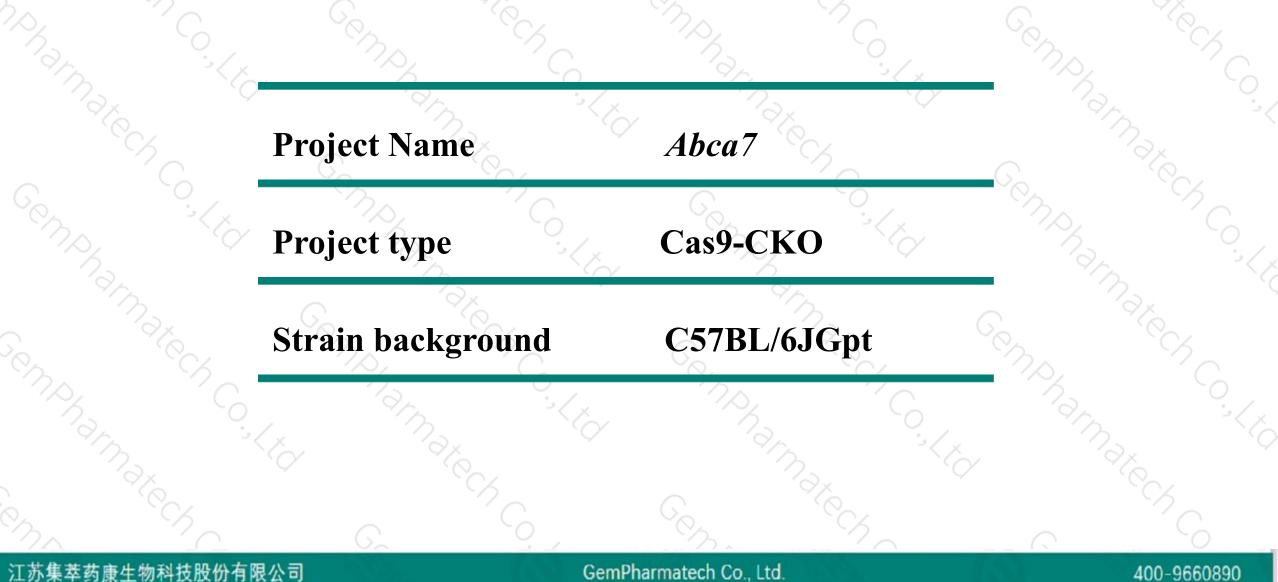
Designer: Reviewer:

Design Date:

Daohua Xu Huimin Su 2020-2-14

Project Overview





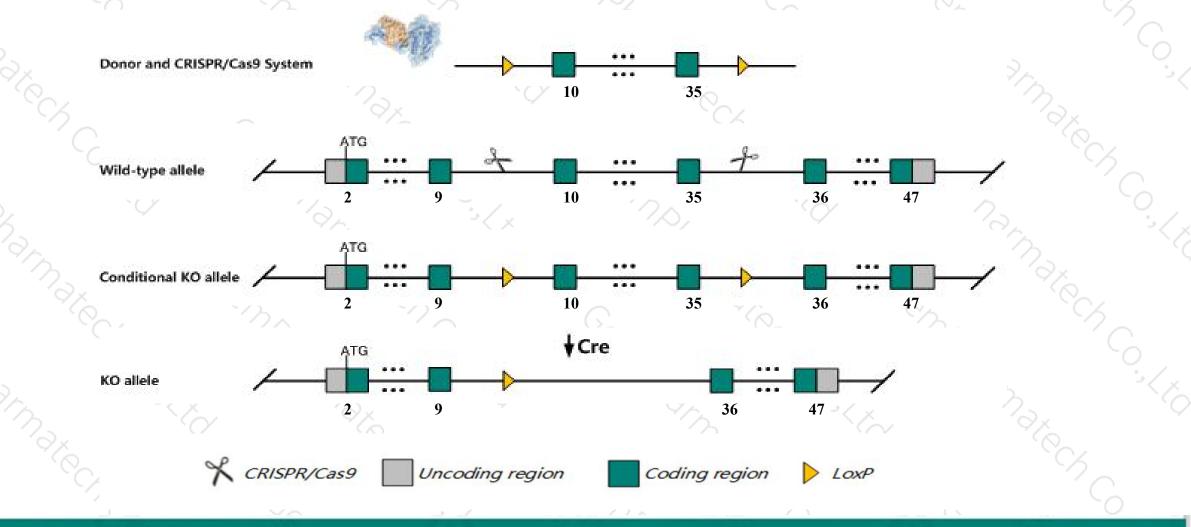
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Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Abca7* gene. The schematic diagram is as follows:



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The Abca7 gene has 3 transcripts. According to the structure of Abca7 gene, exon10-exon35 of Abca7-202 (ENSMUST00000132517.7) transcript is recommended as the knockout region. The region contains 4001bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Abca7* 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, Homozygous mutant females, but not males, have less white fat and lower total serum and HDL cholesterol levels. Males exhibit a 10% reduction in kidney size.
- The Abca7 gene is located on the Chr10. 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)



Abca7 ATP-binding cassette, sub-family A (ABC1), member 7 [Mus musculus (house mouse)]

Gene ID: 27403, updated on 3-Feb-2019

Summary

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Official Symbol	Abca7 provided by MGI
Official Full Name	
Primary source	MGI:MGI:1351646
See related	Ensembl:ENSMUSG0000035722
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
	Muroidea; Muridae; Murinae; Mus; Mus
Also known as	ABCX, Abc51
Summary	The protein encoded by this gene is a member of the superfamily of ATP-binding cassette (ABC) transporters. ABC proteins transport various molecules across extra- and intracellular membranes. ABC genes are divided into seven distinct subfamilies (ABC1, MDR/TAP, MRP, ALD, OABP, GCN20, White). This protein is a member of the ABC1 subfamily. Members of the ABC1 subfamily comprise the only major ABC subfamily found exclusively in multicellular eukaryotes. This protein is widely expressed with highest detection in spleen and hematopoietic tissues. Defects in this gene cause an increase in amyloid-beta deposits in a mouse model of Alzheimer's disease, and a related human protein is thought to play a role in lipid homeostasis in cells of the immune system. [provided by RefSeq, Jan 2017]
Expression	Ubiquitous expression in spleen adult (RPKM 30.2), thymus adult (RPKM 29.6) and 26 other tissues See more
Orthologs	human all

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Transcript information (Ensembl)



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

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Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Abca7-202	ENSMUST00000132517.7	6696	<u>2159aa</u>	Protein coding	CCDS24004	<u>Q91V24</u>	TSL:5 GENCODE basic APPRIS P3
Abca7-203	ENSMUST00000171637.7	6638	<u>2167aa</u>	Protein coding	CCDS83724	<u>E9Q6G4</u>	TSL:1 GENCODE basic APPRIS ALT2
Abca7-201	ENSMUST0000043866.7	6590	<u>2159aa</u>	Protein coding	CCDS24004	<u>Q91V24</u>	TSL:1 GENCODE basic APPRIS P3

The strategy is based on the design of Abca7-202 transcript, The transcription is shown below

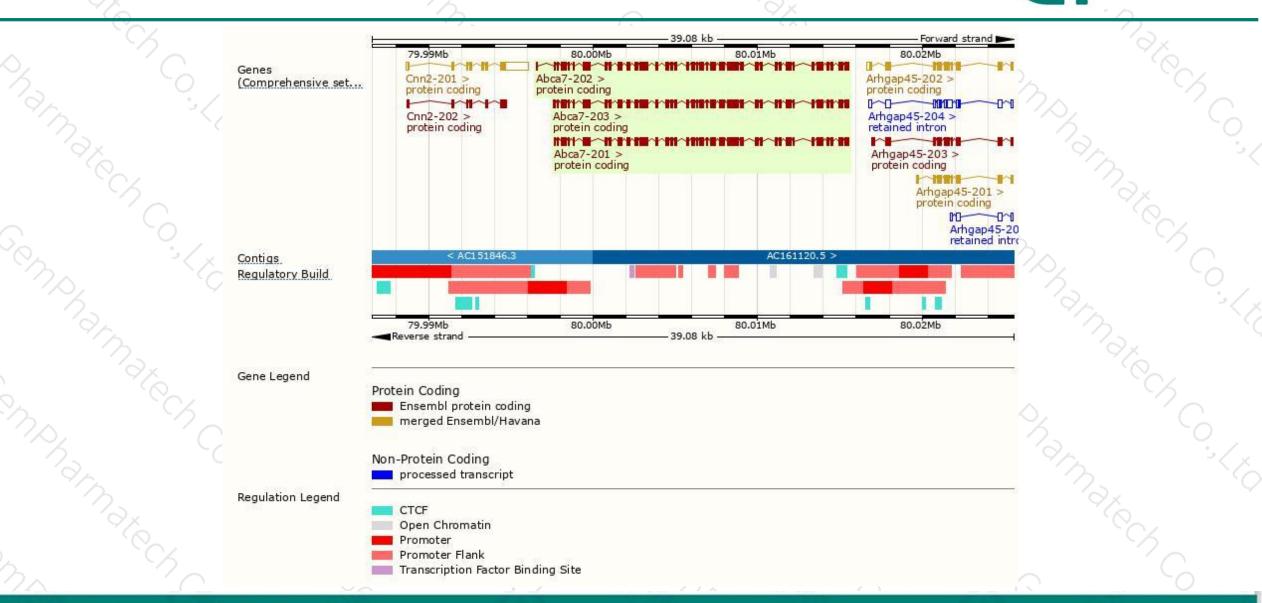
Abca7-202 > protein coding

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Genomic location distribution



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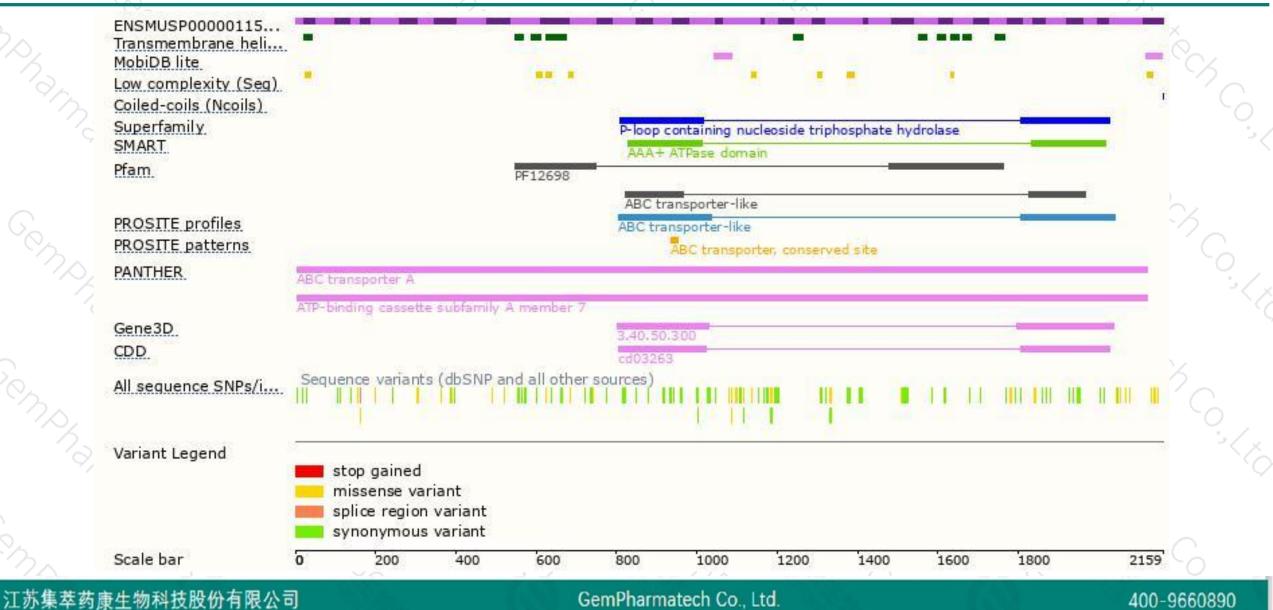
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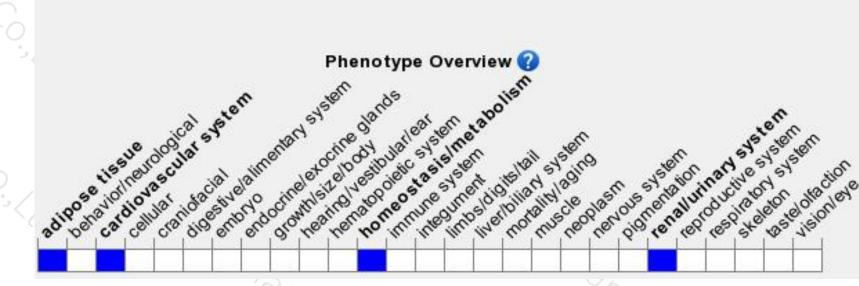
Protein domain





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, Homozygous mutant females, but not males, have less white fat and lower total serum and HDL cholesterol levels. Males exhibit a 10% reduction in kidney size.



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



