

# Abo Cas9-CKO Strategy

Designer: Reviewer:

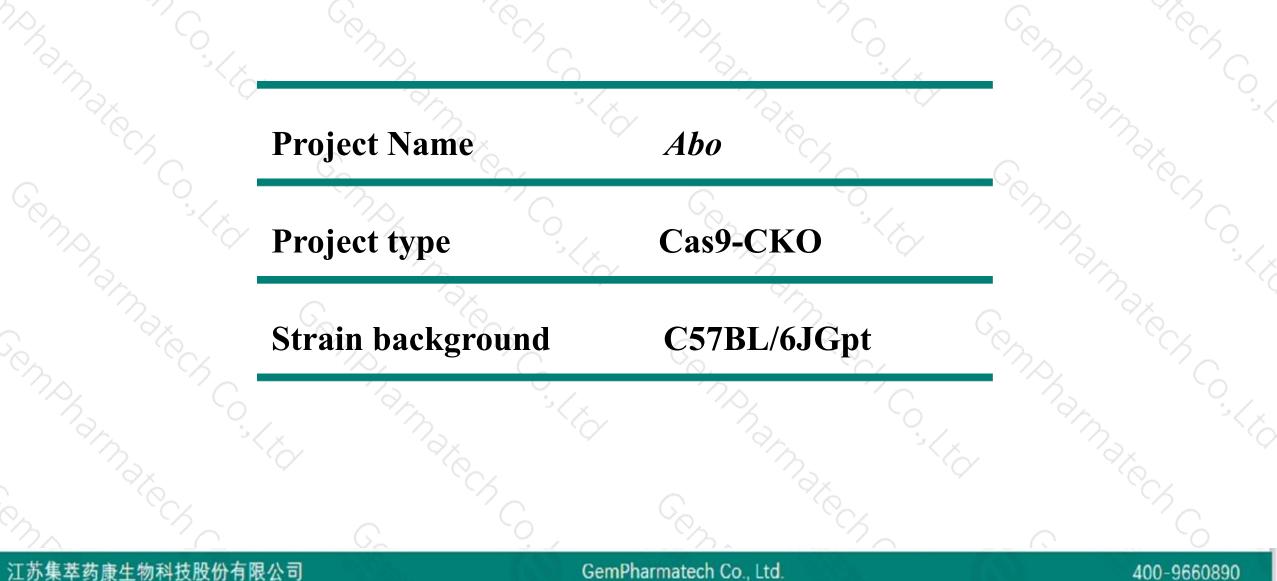
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Design Date:

Huan Fan Huan Wang 2019-12-11

## **Project Overview**





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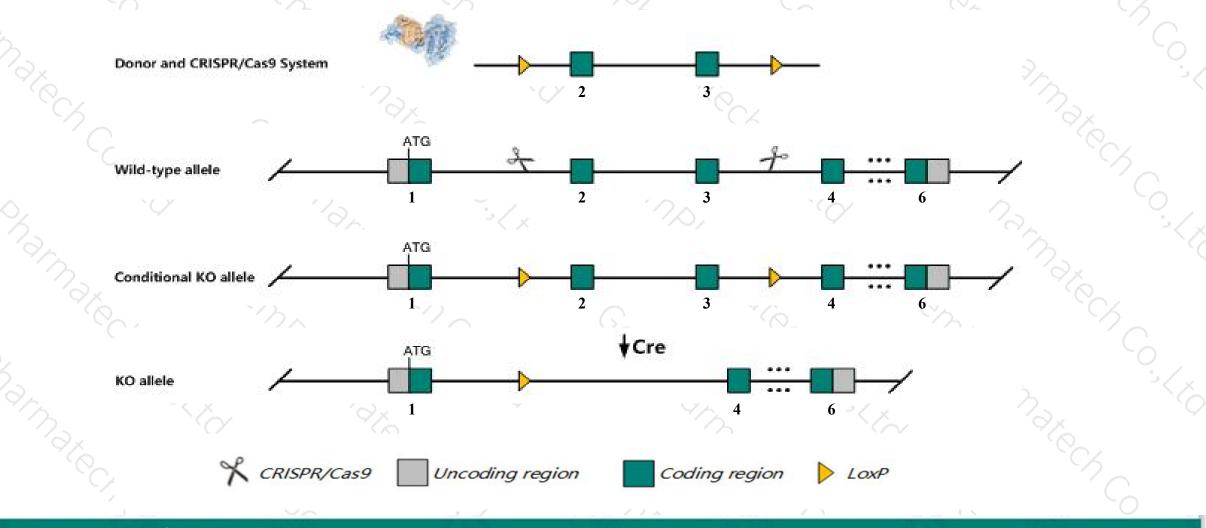
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### **Conditional Knockout strategy**



400-9660890

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



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The Abo gene has 2 transcripts. According to the structure of Abo gene, exon2-exon3 of Abo-201 (ENSMUST00000102900.1) transcript is recommended as the knockout region. The region contains 127bp coding sequence. Knock out the region will result in disruption of protein function.

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



- The Abo 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)**



### Abo ABO blood group (transferase A, alpha 1-3-N-acetylgalactosaminyltransferase, transferase B, alpha 1-3galactosyltransferase) [Mus musculus (house mouse)]

Gene ID: 80908, updated on 31-Jan-2019

Summary	
Official Symbol	Abo provided by MGI
Official Full Name	ABO blood group (transferase A, alpha 1-3-N-acetylgalactosaminyltransferase, transferase B, alpha 1-3-galactosyltransferase) provided
	by <u>MGI</u>
Primary source	MGI:MGI:2135738
See related	Ensembl:ENSMUSG00000015787
Gene type	protein coding
<b>RefSeq status</b>	VALIDATED
Organism	Mus musculus
Lineage	
	Muroidea; Muridae; Murinae; Mus; Mus
Also known as	NAGAT
Expression	Biased expression in genital fat pad adult (RPKM 4.6), colon adult (RPKM 3.3) and 2 other tissues See more
Orthologs	human all

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The gene has 2 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Abo-201	ENSMUST00000102900.1	1779	<u>332aa</u>	Protein coding	CCDS15811	<u>P38649</u>	TSL:1 GENCODE basic APPRIS P2
Abo-202	ENSMUST00000114045.8	1811	<u>334aa</u>	Protein coding	-	Z4YLB0	TSL:5 GENCODE basic APPRIS ALT2

The strategy is based on the design of *Abo-201* transcript, The transcription is shown below

< Abo-201 protein coding

Reverse strand -

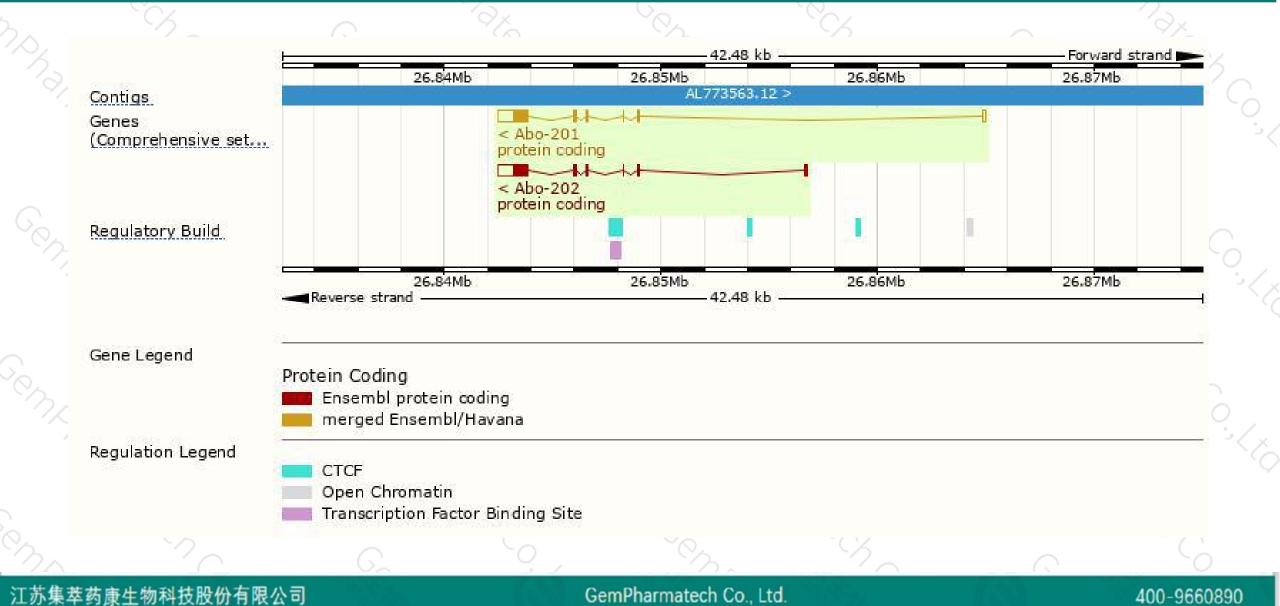
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22.48 kb

### **Genomic location distribution**





### **Protein domain**



No.	Co.	Cho.	Č S					2000 -	°°G	
	ENSMUSP00000099 Transmembrane heli Low complexity (Seg) Superfamily	Nuc	leotide-diphospho-s	ugar transferase	25	• 7	-			
$\sim$	<u>Pfam</u>	Glyco	osyl transferase, far	nily 6						
C.	PANTHER	Glycosyl transferase, family 6								
		PTHR10462;SF	10							
	Gene3D	Nucle	otide-diphospho-su	gar transferases	5					
	CDD		cd02515							
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If you have any questions, you are welcome to inquire. Tel: 400-9660890



