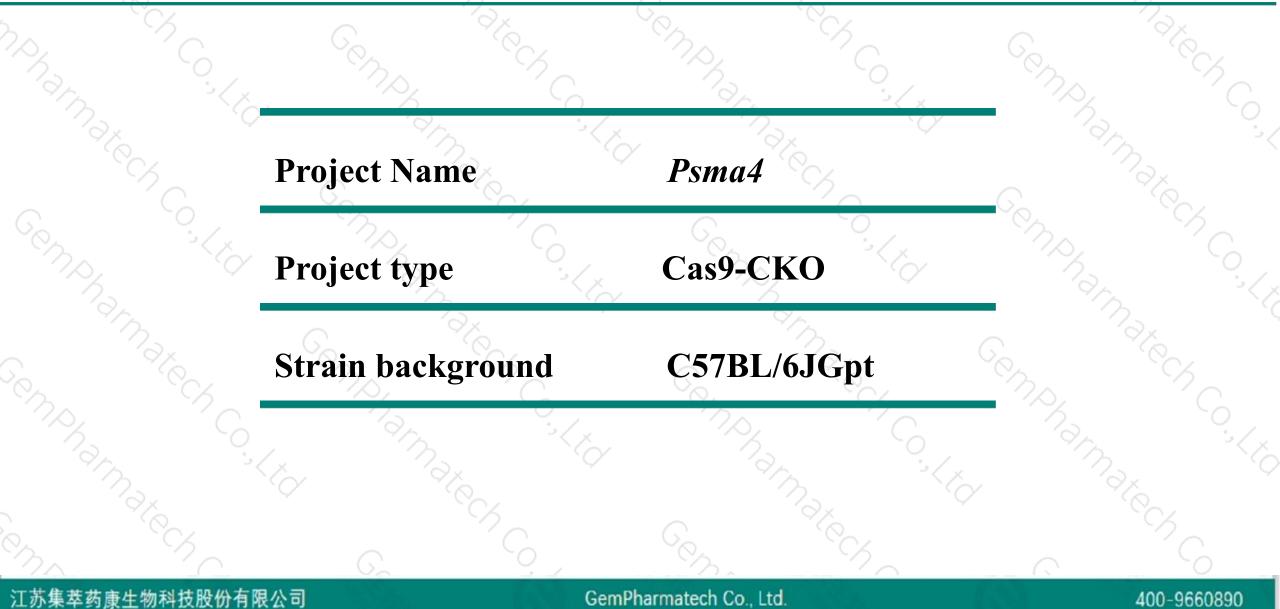


# **Psma4 Cas9-CKO Strategy**

Designer: Reviewer: Design Date: JiaYu Xiaojing Li 2020-3-19

## **Project Overview**

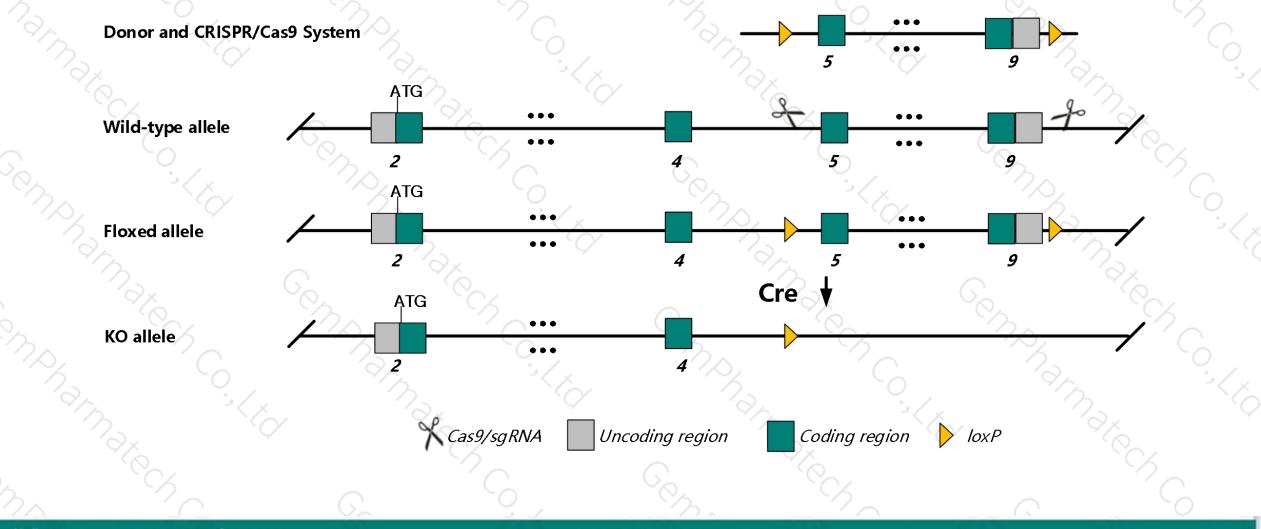




## **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Psma4* gene. The schematic diagram is as follows:



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The Psma4 gene has 7 transcripts. According to the structure of Psma4 gene, exon5-exon9 of Psma4-201 (ENSMUST00000034848.13) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Psma4* 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 flox region is about 3 kb away from the 5th end of the AY074887 gene, which may affect the regulation of this gene.
- The Psma4 gene is located on the Chr9. 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)**



☆ ?

### Psma4 proteasome subunit alpha 4 [ Mus musculus (house mouse) ]

Gene ID: 26441, updated on 13-Mar-2020

Summary

Official Symbol	Psma4 provided by MGI
Official Full Name	proteasome subunit alpha 4 provided by MGI
Primary source	MGI:MGI:1347060
See related	Ensembl:ENSMUSG0000032301
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;
	Myomorpha; Muroidea; Muridae; Mus; Mus
Also known as	C9
Expression	Ubiquitous expression in placenta adult (RPKM 71.5), CNS E11.5 (RPKM 59.3) and 27 other tissues See more
Orthologs	human all
	$\nabla A_{ij} = \frac{1}{2} \left( \frac{1}{2} \right) \left( \frac{1}{2$

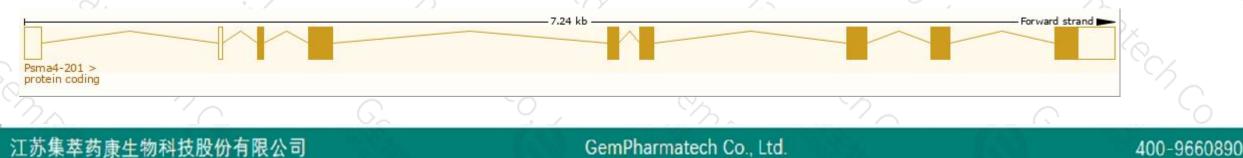
## **Transcript information (Ensembl)**



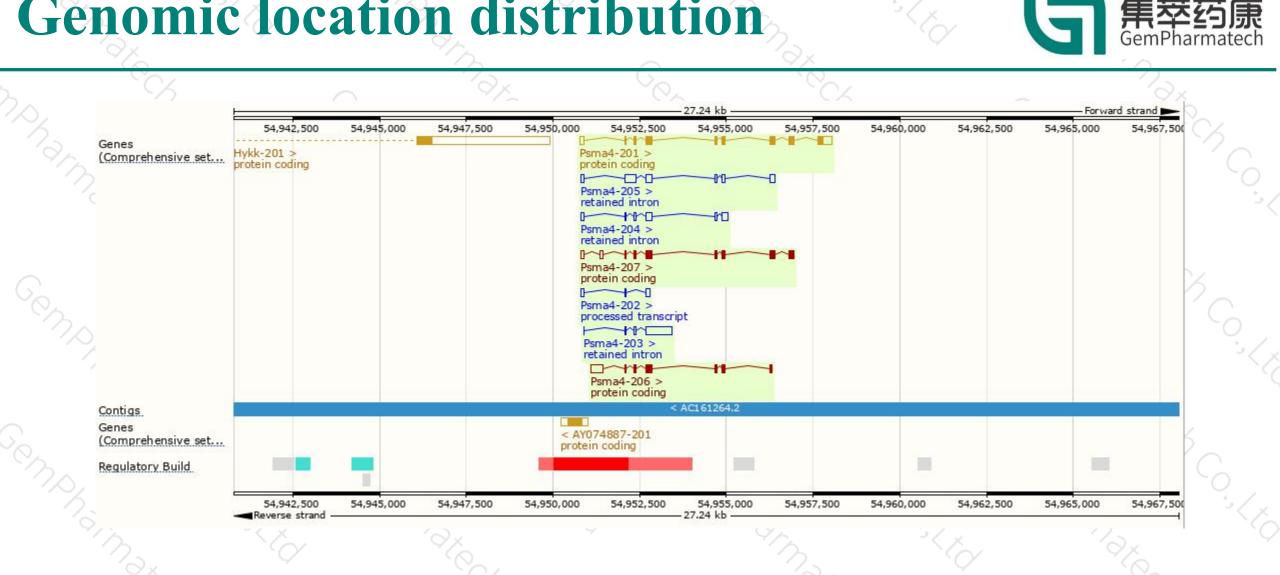
### The gene has 7 transcripts, all transcripts are shown below:

Name 🖕	Transcript ID	bp 🖕	Protein 🖕	Biotype 🖕	CCDS 🍦	UniProt 🖕	Flags
Psma4-201	ENSMUST0000034848.13	1165	<u>261aa</u>	Protein coding	<u>CCDS40643</u> &	<u>Q9R1P0</u> &	TSL:1 GENCODE basic APPRIS P1
Psma4-207	ENSMUST00000172407.7	828	<u>210aa</u>	Protein coding		<u>E9PW69</u> ₽	CDS 3' incomplete TSL:3
Psma4-206	ENSMUST00000171900.1	768	<u>139aa</u>	Protein coding		<u>E9Q0X0</u> &	CDS 3' incomplete TSL:3
Psma4-202	ENSMUST00000163960.7	244	No protein	Processed transcript		23	TSL:5
Psma4-205	ENSMUST00000171578.7	868	No protein	Retained intron	1.00		TSL:3
Psma4-203	ENSMUST00000164679.1	832	No protein	Retained intron	1.52		TSL:2
Psma4-204	ENSMUST00000169008.7	562	No protein	Retained intron			TSL:2

The strategy is based on the design of *Psma4-201* transcript, The transcription is shown below



### **Genomic location distribution**



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## **Protein domain**



ENSMUSP00000034 PDB-ENSP mappings							
MobiDB lite							
Low complexity (Seg)					_		-
Coiled-coils (Ncoils)	<b>1</b>						
Superfamily	Nucleophile aminohydrolases, N-t	erminal					
SMART	Proteasome alpha-subunit, N-te	rminal domain					
Pfam.	Proteasome, s	subunit alpha/beta					
	Proteasome alpha-subunit, N-te	rminal domain					
PROSITE profiles	Proteasome alpha-ty	pe subunit				18 177	
PROSITE patterns	Proteason	e beta-type subunit, conserved site					
	Proteasome alpha-subunit, N-te	rminal domain					2
PANTHER	PTHR11599						
	PTHR11599:SF13						
Gene3D	Nucleophile aminohydrolases, N-ter	minal					
CDD	cd03752						
CDD All sequence SNPs/i	cd03752 Sequence variants (dbSNP and	all other sources)		0.00			
All sequence SNPs/i		all other sources)		3.82	3		
		all other sources)			a -		
All sequence SNPs/i	Sequence variants (dbSNP and	all other sources)	120	160	200		261
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All sequence SNPs/i Variant Legend	Sequence variants (dbSNP and		120	160	200	nam.	261
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If you have any questions, you are welcome to inquire. Tel: 400-9660890



