

Ppill Cas9-CKO Strategy

Designer:

Reviewer:

Design Date:

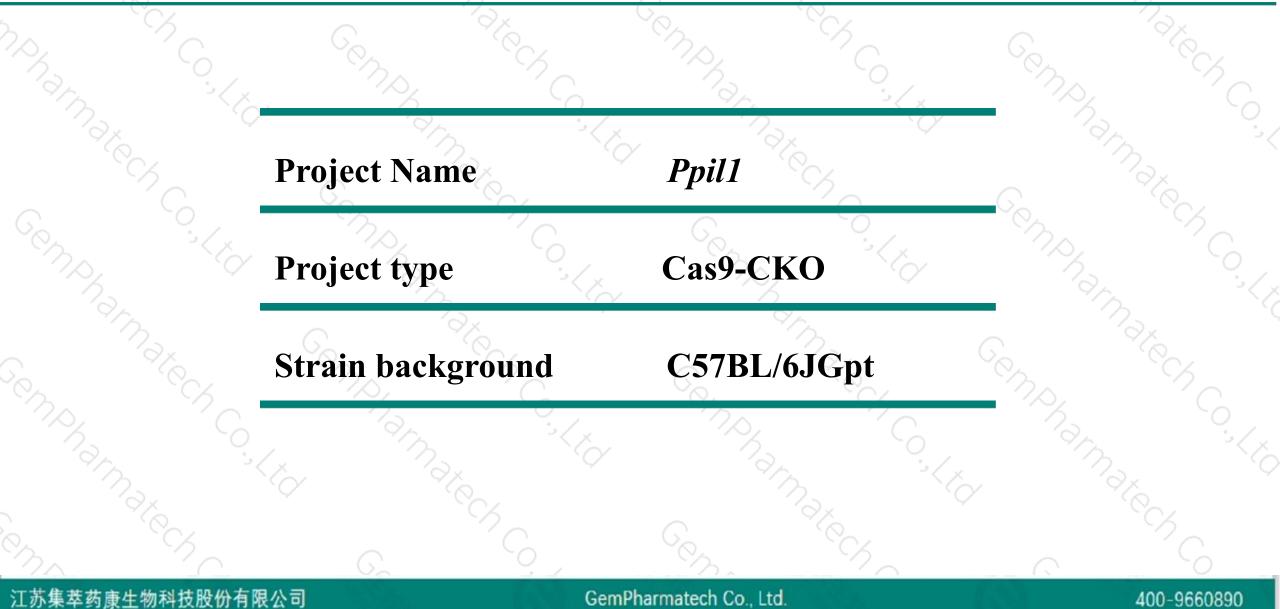
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2020-4-16

Project Overview

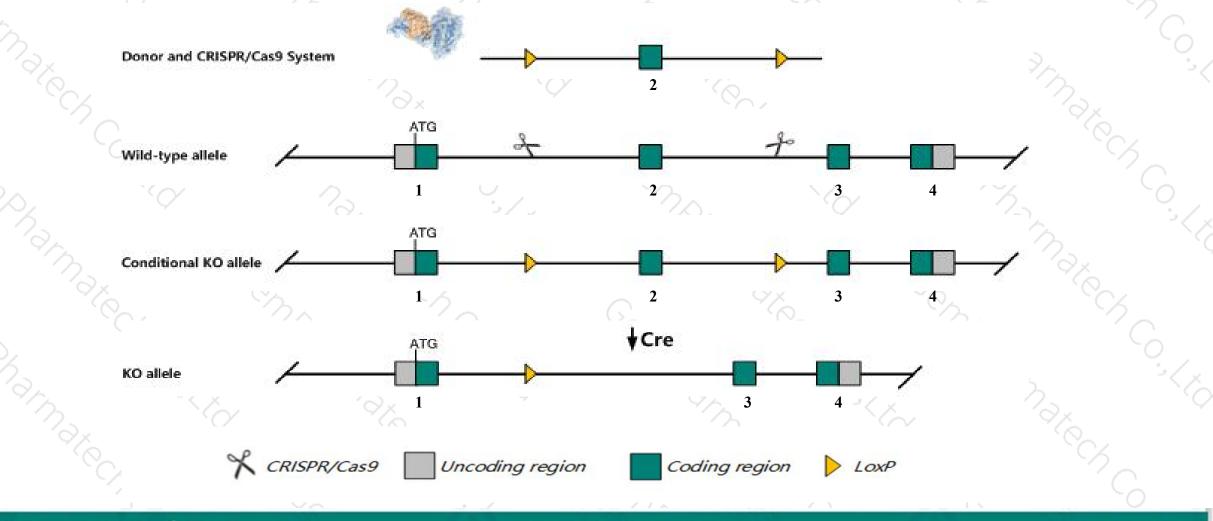




Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Ppil1* gene. The schematic diagram is as follows:



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 The *Ppil1* gene has 7 transcripts. According to the structure of *Ppil1* gene, exon2 of *Ppil1-201* (ENSMUST00000024802.9) transcript is recommended as the knockout region. The region contains 155bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Ppil1* 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



- ➤ Transcript *Ppil1*-204&205&206&207 may not be affected.
- The floxed region is near to the N-terminal of BC004004 gene and Gm50068 gene, this strategy may influence the regulatory function of the N-terminal of these genes.
- The *Ppil1* gene is located on the Chr17. 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.

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Gene information (NCBI)



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Ppil1 peptidylprolyl isomerase (cyclophilin)-like 1 [Mus musculus (house mouse)]

Gene ID: 68816, updated on 13-Mar-2020

Summary

Official Symbol	Ppil1 provided by MGI
Official Full Name	peptidylprolyl isomerase (cyclophilin)-like 1 provided by <u>MGI</u>
Primary source	MGI:MGI:1916066
See related	Ensembl:ENSMUSG0000024007
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; Murinae; Mus; Mus
Also known as	1110060O10Rik, Al327391, Cypl1
Expression	Ubiquitous expression in liver E14.5 (RPKM 35.3), CNS E11.5 (RPKM 34.7) and 28 other tissues See more
Orthologs	human all

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Transcript information (Ensembl)



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

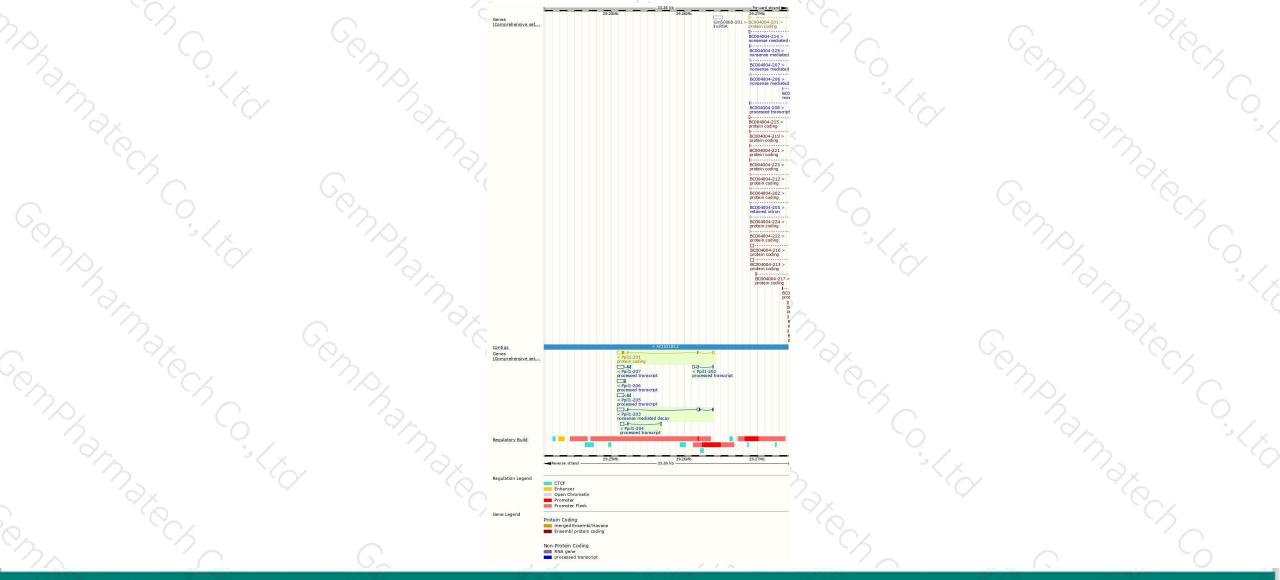
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ppil1-201	ENSMUST0000024802.9	1462	<u>166aa</u>	Protein coding	CCDS28593	B9EJX8 Q9D0W5	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS F
Ppil1-203	ENSMUST00000135123.2	1476	<u>64aa</u>	Nonsense mediated decay	(F6SSV9	TSL:5
Ppil1-207	ENSMUST00000234880.1	1117	No protein	Processed transcript	1943	1946	
Ppil1-205	ENSMUST00000234139.1	1101	No protein	Processed transcript	1021	121	
Ppil1-206	ENSMUST00000234315.1	1050	No protein	Processed transcript		(5)	
Ppil1-204	ENSMUST00000136811.7	734	No protein	Processed transcript	()	(8 1	TSL:3
Ppil1-202	ENSMUST00000129157.2	508	No protein	Processed transcript	12421	144	TSL:3
	No.	100 c		N			

The strategy is based on the design of *Ppil1-201* transcript, the transcription is shown below



Genomic location distribution





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Protein domain



ENSMUSP00000024 Low complexity (Seg)			9	3							
Superfamily	Cyclophilin-like o	lomain superfamily						1	Q		
Prints		Cyclophilin-type pep	tidyl-prolyl cis-t	rans isomerase	e domain	(6 -100	19 A				
Pfam	Cyclophilir	n-type peptidyl-proly	cis-trans isom	erase domain							
PROSITE profiles	Cycloph	ilin-type peptidyl-pro	olyl cis-trans iso	merase domaii	n			-			
PROSITE patterns		Cyc	lophilin-type pe	ptidyl-prolyl ci	s-trans isomerase,	conserved site					
PIRSF	Cyclophilin-type (eptidyl-prolyl cis-tra	ins isomerase								
PANTHER	PTHR45625(SF7										
	PTHR45625										
Gene3D	Cyclophilin-like domi	ain superfamily									
CDD.	cd01922	i and a star star star star star star star st						-			
All sequence SNPs/i	Sequence variants		her sources)				1) 57,		
Variant Legend	synonymous	variant	10				(14 C				
Scale bar	0 20	40	60	80	100	120	140	166			
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	62	9.2		25.				<u> </u>			

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



