

Clpx Cas9-CKO Strategy

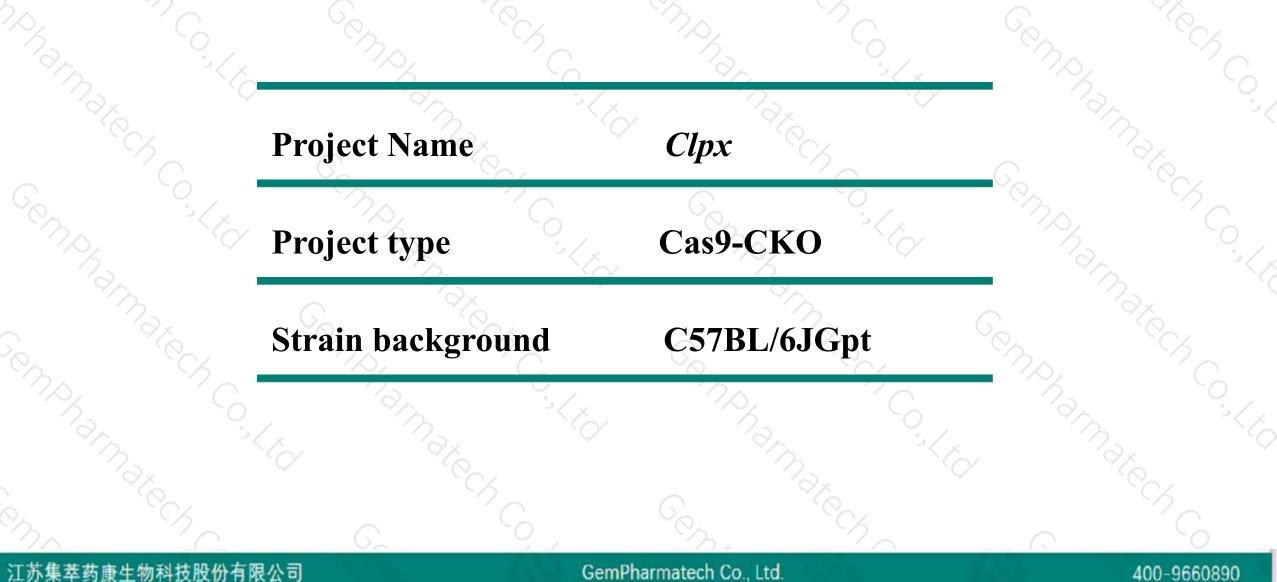
Designer: Design Date:

Q,

Huan Wang 2019-8-3

Project Overview





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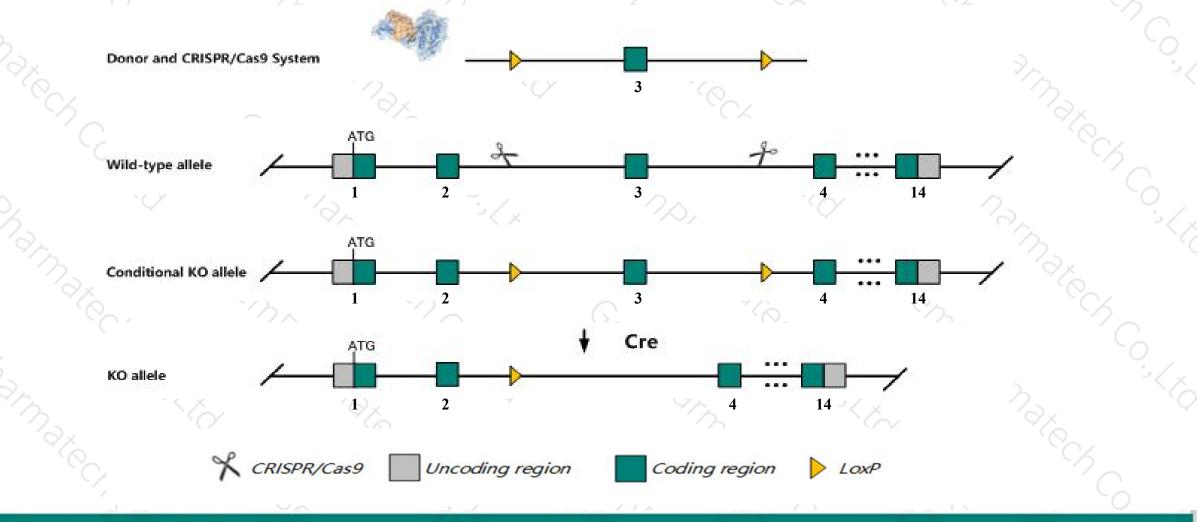
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Conditional Knockout strategy



400-9660890

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



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The Clpx gene has 4 transcripts. According to the structure of Clpx gene, exon3 of Clpx-201 (ENSMUST00000015501.10) transcript is recommended as the knockout region. The region contains 118bp coding sequence. Knock out the region will result in disruption of protein function.

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



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Clpx caseinolytic mitochondrial matrix peptidase chaperone subunit [Mus musculus (house mouse)]

Gene ID: 270166, updated on 7-Apr-2019

Summary

Official Symbol	Clpx provided by MGI
•	caseinolytic mitochondrial matrix peptidase chaperone subunit provided by <u>MGI</u>
Primary source	MGI:MGI:1346017
See related	Ensembl:ENSMUSG0000015357
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	AU014732, E330029l21
Expression	Ubiquitous expression in liver E18 (RPKM 23.0), testis adult (RPKM 18.8) and 28 other tissues See more
Orthologs	human all

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Transcript information (Ensembl)



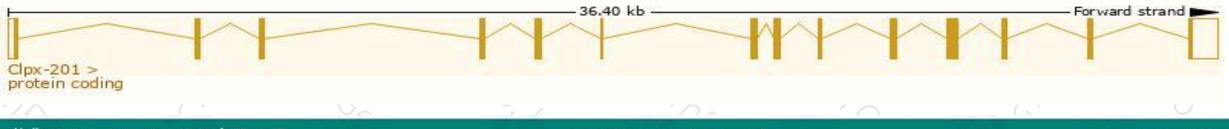
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The gene has 4 transcripts, all transcripts are shown below:

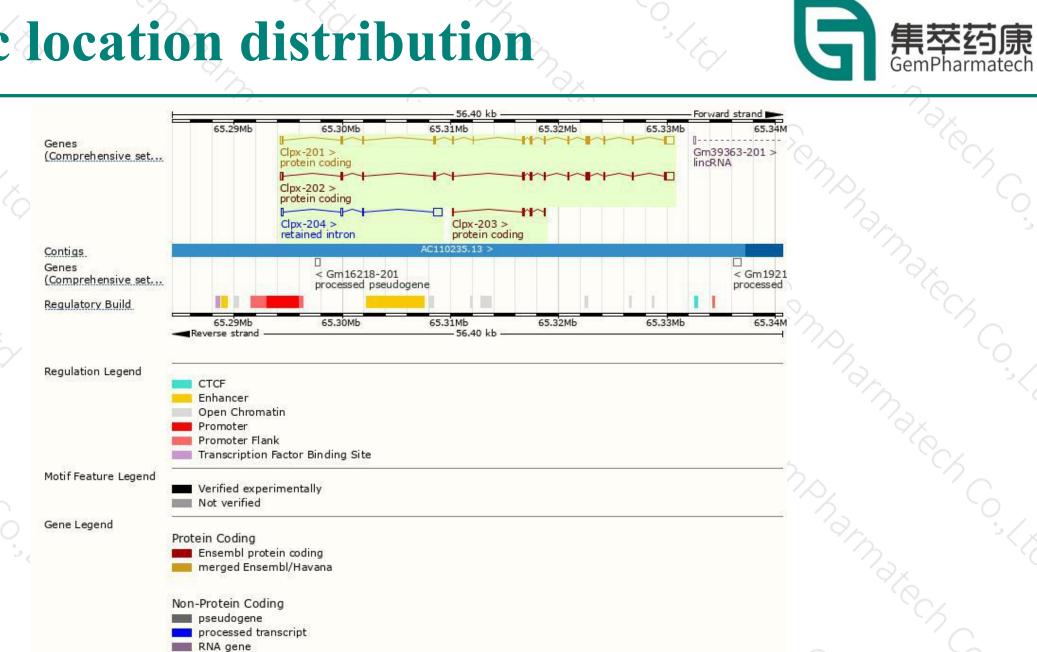
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	bp	Protein 4	Biotype 🍦	CCDS 🖕	UniProt 🍦	Flags
	1279	No protein	Details and interest			
2 ENIONUOT000004470704		no protoni	Retained intron	-	-	TSL:1
03 ENSMUST00000147279.1	577	<u>192aa</u>	Protein coding		<u>F7BB92</u> ₽	CDS 5' and 3' incomplete TSL:2
02 ENSMUST00000113824.7	2809	<u>620aa</u>	Protein coding	<u>CCDS40669</u> &	<u>Q6P8N8</u> &	TSL:1 GENCODE basic APPRIS ALT
01 ENSMUST0000015501.10	2885	<u>634aa</u>	Protein coding	<u>CCDS23288</u> 교	<u>Q9JHS4</u> 교	TSL:1 GENCODE basic APPRIS P3
° CY		5.		0		3. SA
[×] Q ^µ		- ?~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~		~		$\gamma_{\mathcal{S}_{\mathcal{L}}}$

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



Genomic location distribution



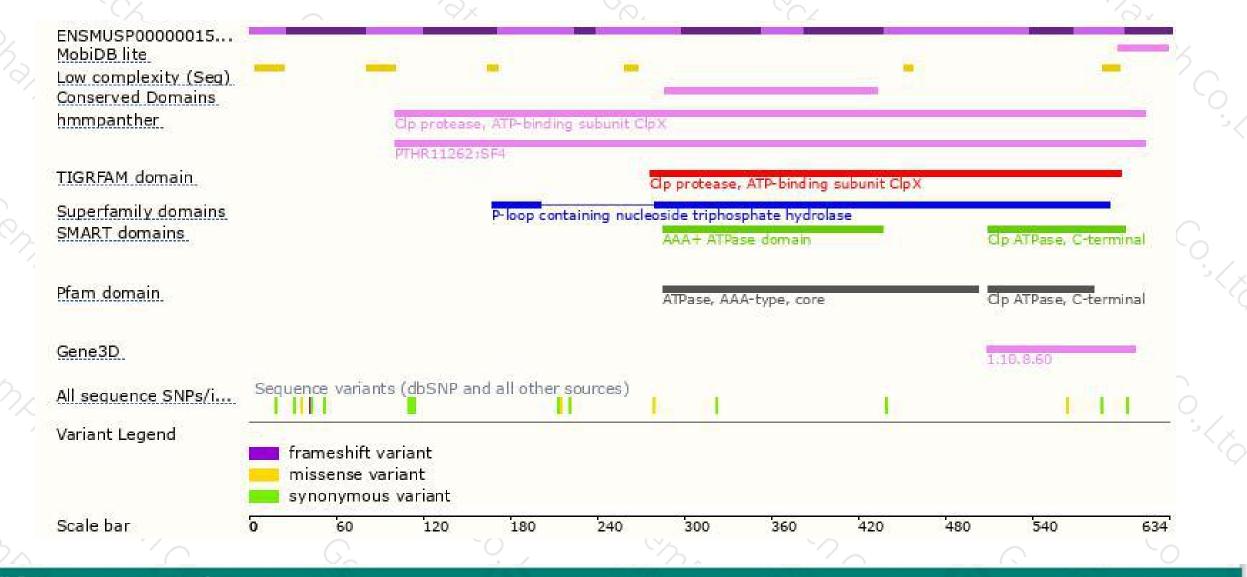
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Protein domain





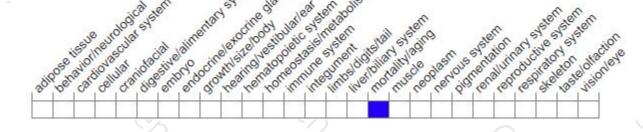
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Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).



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



