

# Usp36 Cas9-CKO Strategy

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# **Project Overview**



**Project Name** 

Usp36

**Project type** 

Cas9-CKO

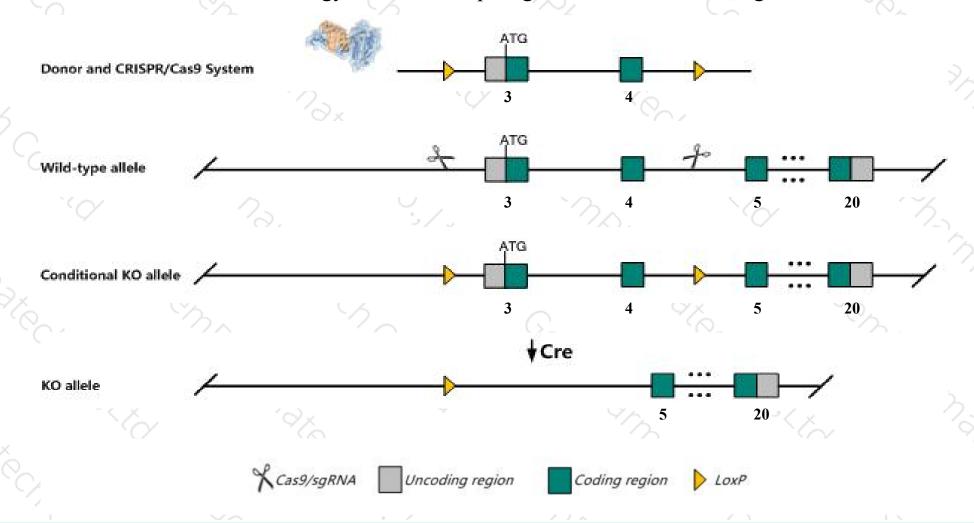
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Usp36* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Usp36* gene has 6 transcripts. According to the structure of *Usp36* gene, exon3-exon4 of *Usp36*-202(ENSMUST00000106296.8) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Usp36* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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**



- > According to the existing MGI data, mice homozygous for a gene trap allele display lethality before implantation and arrest at the morula stage.
- ➤ The *Usp36* gene is located on the Chr11. 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)



#### Usp36 ubiquitin specific peptidase 36 [Mus musculus (house mouse)]

Gene ID: 72344, updated on 13-Mar-2020

#### Summary

☆ ?

Official Symbol Usp36 provided by MGI

Official Full Name ubiquitin specific peptidase 36 provided by MGI

Primary source MGI:MGI:1919594

See related Ensembl:ENSMUSG00000033909

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 2700002L06Rik, mKIAA1453

Expression Ubiquitous expression in ovary adult (RPKM 8.8), limb E14.5 (RPKM 8.2) and 28 other tissuesSee more

Orthologs <u>human all</u>

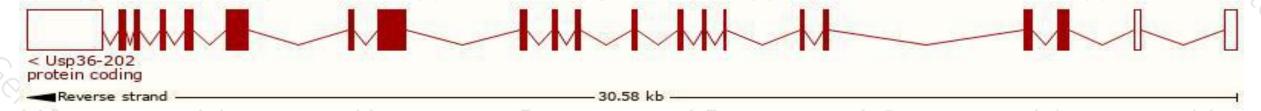
# Transcript information (Ensembl)



The gene has 6 transcripts, all transcripts are shown below:

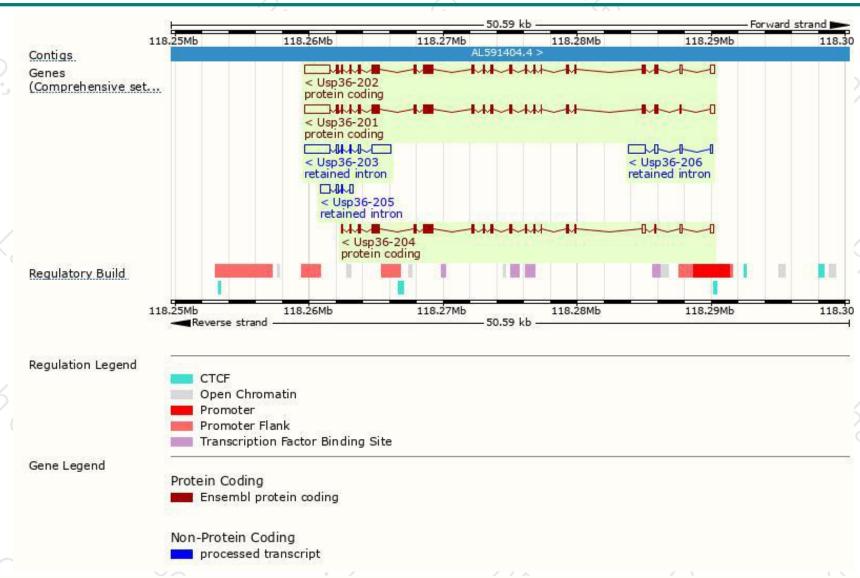
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Usp36-202	ENSMUST00000106296.8	5655	1098aa	Protein coding	CCDS48998	B1AQJ2	TSL:5 GENCODE basic APPRIS P1
Usp36-201	ENSMUST00000092382.9	5609	<u>1098aa</u>	Protein coding	CCDS48998	B1AQJ2	TSL:5 GENCODE basic APPRIS P1
Usp36-204	ENSMUST00000144153.7	3391	<u>890aa</u>	Protein coding		B0QZF8	CDS 3' incomplete TSL:5
Usp36-203	ENSMUST00000141647.7	3815	No protein	Retained intron	6-0	-	TSL:1
Usp36-206	ENSMUST00000151433.1	1821	No protein	Retained intron	(54)	-	TSL:1
Usp36-205	ENSMUST00000148998.1	1212	No protein	Retained intron	450		TSL:1

The strategy is based on the design of *Usp36-202* transcript, the transcription is shown below:



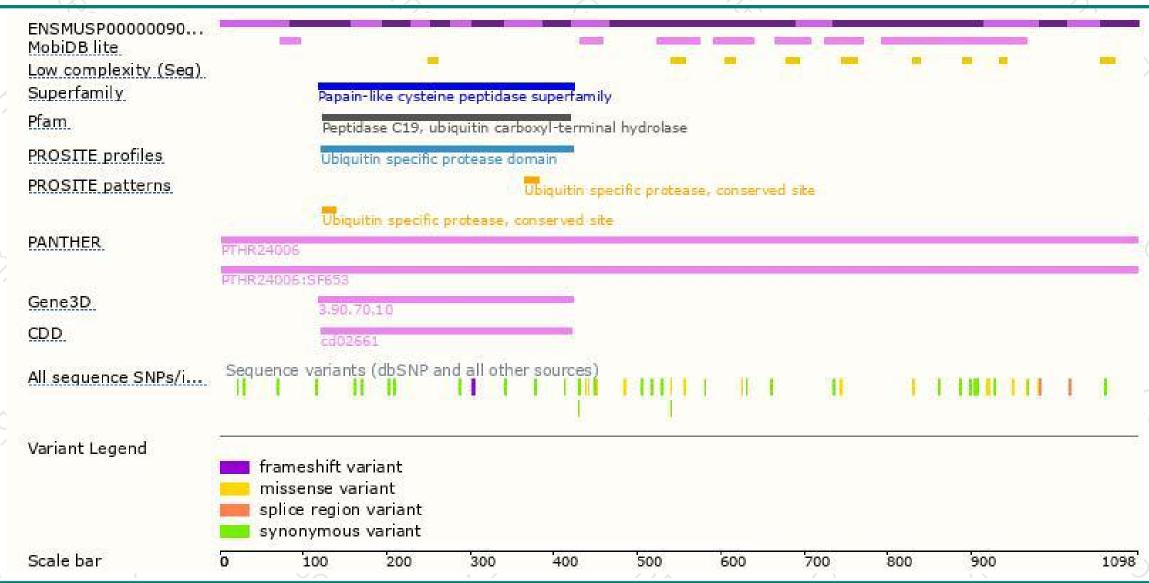
### Genomic location distribution





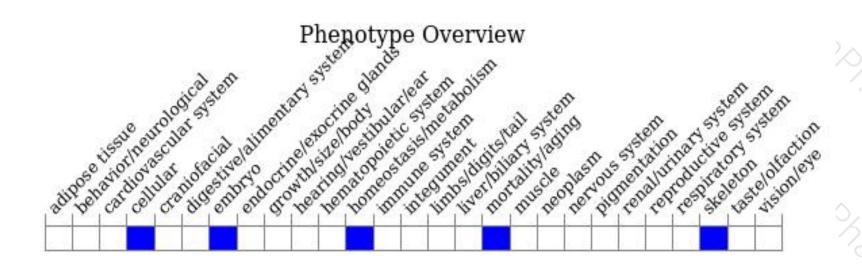
### Protein domain





# 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, mice homozygous for a gene trap allele display lethality before implantation and arrest at the morula stage.



If you have any questions, you are welcome to inquire.

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