

# Ube2e2 Cas9-CKO Strategy

Designer: Xiaojing Li

Design Date: 2019-8-15

# **Project Overview**



**Project Name** 

Ube2e2

**Project type** 

Cas9-CKO

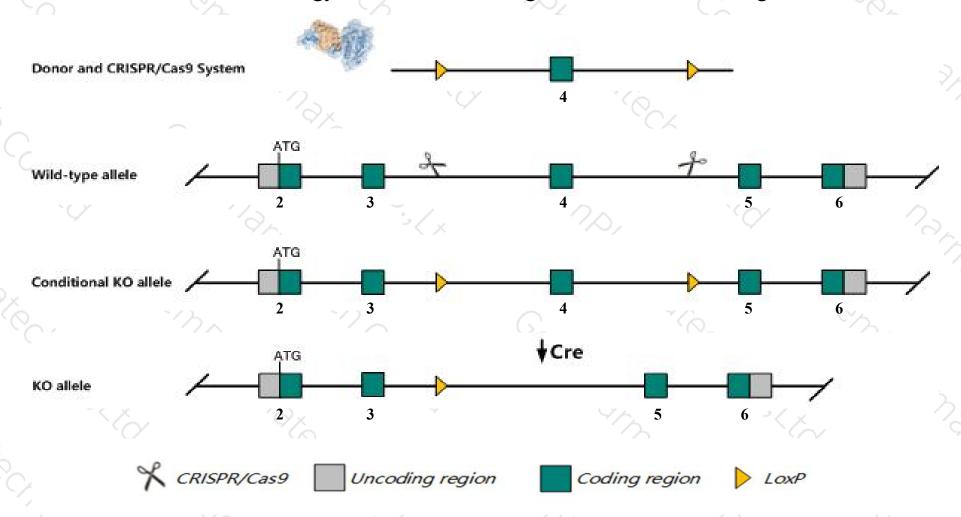
Strain background

C57BL/6JGpt

# Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Ube2e2* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Ube2e2* gene has 14 transcripts. According to the structure of *Ube2e2* gene, exon4 of *Ube2e2-205*(ENSMUST00000150727.7) transcript is recommended as the knockout region. The region contains 133bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Ube2e2* 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**



- ➤ The *Ube2e2* gene is located on the Chr14. 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)



#### Ube2e2 ubiquitin-conjugating enzyme E2E 2 [Mus musculus (house mouse)]

Gene ID: 218793, updated on 31-Jan-2019

#### Summary

☆ ?

Official Symbol Ube2e2 provided by MGI

Official Full Name ubiquitin-conjugating enzyme E2E 2 provided by MGI

Primary source MGI:MGI:2384997

See related Ensembl:ENSMUSG00000058317

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 BC016265

Expression Ubiquitous expression in CNS E18 (RPKM 15.9), cortex adult (RPKM 15.2) and 28 other tissuesSee more

Orthologs <u>human</u> all

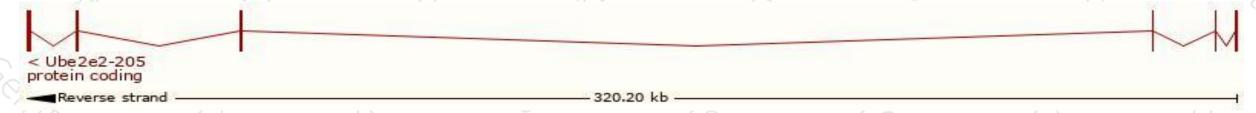
# Transcript information (Ensembl)



The gene has 14 transcripts, all transcripts are shown below:

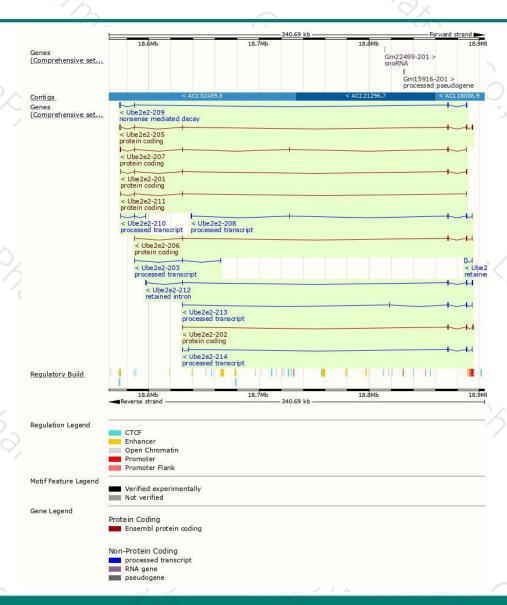
Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
ENSMUST00000150727.7	1523	201aa	Protein coding	CCDS26838	Q91W82	TSL:1 GENCODE basic APPRIS P1
ENSMUST00000076133.9	606	201aa	Protein coding	CCDS26838	Q91W82	TSL:3 GENCODE basic APPRIS P1
ENSMUST00000175643.7	757	<u>167aa</u>	Protein coding	2	H3BL69	CDS 5' incomplete TSL:3
ENSMUST00000151926.7	666	<u>181aa</u>	Protein coding	2	D3YXD1	CDS 3' incomplete TSL:3
ENSMUST00000124353.1	445	108aa	Protein coding	5	D3YW10	CDS 3' incomplete TSL:3
ENSMUST00000176843.7	438	146aa	Protein coding	-	H3BL23	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
ENSMUST00000176555.7	1200	<u>92aa</u>	Nonsense mediated decay	2	H3BJB5	TSL:5
ENSMUST00000176062.2	647	No protein	Processed transcript	2	556	TSL:3
ENSMUST00000177398.7	514	No protein	Processed transcript	-	15	TSL:3
ENSMUST00000177259.7	423	No protein	Processed transcript	-	35#3	TSL:3
ENSMUST00000135381.7	335	No protein	Processed transcript	2	020	TSL:3
ENSMUST00000176672.1	328	No protein	Processed transcript	-	3523	TSL:5
ENSMUST00000144873.1	2135	No protein	Retained intron		1871	TSL:1
ENSMUST00000176931.7	772	No protein	Retained intron	-	-	TSL:3
	ENSMUST00000150727.7 ENSMUST00000175643.7 ENSMUST00000175643.7 ENSMUST00000151926.7 ENSMUST00000124353.1 ENSMUST00000176843.7 ENSMUST00000176555.7 ENSMUST00000177398.7 ENSMUST00000177259.7 ENSMUST00000176672.1 ENSMUST00000176672.1 ENSMUST00000144873.1	ENSMUST00000150727.7 1523 ENSMUST00000076133.9 606 ENSMUST00000175643.7 757 ENSMUST00000151926.7 666 ENSMUST00000124353.1 445 ENSMUST00000176843.7 438 ENSMUST00000176555.7 1200 ENSMUST00000176062.2 647 ENSMUST00000177398.7 514 ENSMUST00000177259.7 423 ENSMUST00000177259.7 423 ENSMUST00000135381.7 335 ENSMUST00000176672.1 328 ENSMUST00000144873.1 2135	ENSMUST00000150727.7 1523 201aa  ENSMUST00000076133.9 606 201aa  ENSMUST00000175643.7 757 167aa  ENSMUST00000151926.7 666 181aa  ENSMUST00000124353.1 445 108aa  ENSMUST00000176843.7 438 146aa  ENSMUST00000176555.7 1200 92aa  ENSMUST00000176062.2 647 No protein  ENSMUST00000177259.7 423 No protein  ENSMUST00000135381.7 335 No protein  ENSMUST00000176672.1 328 No protein  ENSMUST00000176672.1 328 No protein	ENSMUST00000150727.7         1523         201aa         Protein coding           ENSMUST00000076133.9         606         201aa         Protein coding           ENSMUST00000175643.7         757         167aa         Protein coding           ENSMUST00000151926.7         666         181aa         Protein coding           ENSMUST00000124353.1         445         108aa         Protein coding           ENSMUST00000176843.7         438         146aa         Protein coding           ENSMUST00000176555.7         1200         92aa         Nonsense mediated decay           ENSMUST00000176062.2         647         No protein         Processed transcript           ENSMUST00000177398.7         514         No protein         Processed transcript           ENSMUST00000177259.7         423         No protein         Processed transcript           ENSMUST00000135381.7         335         No protein         Processed transcript           ENSMUST00000176672.1         328         No protein         Processed transcript           ENSMUST00000144873.1         2135         No protein         Processed transcript	ENSMUST00000150727.7         1523         201aa         Protein coding         CCDS26838           ENSMUST00000076133.9         606         201aa         Protein coding         CCDS26838           ENSMUST00000175643.7         757         167aa         Protein coding         -           ENSMUST00000151926.7         666         181aa         Protein coding         -           ENSMUST00000124353.1         445         108aa         Protein coding         -           ENSMUST00000176843.7         438         146aa         Protein coding         -           ENSMUST00000176555.7         1200         92aa         Nonsense mediated decay         -           ENSMUST00000176062.2         647         No protein         Processed transcript         -           ENSMUST00000177398.7         514         No protein         Processed transcript         -           ENSMUST00000177259.7         423         No protein         Processed transcript         -           ENSMUST00000135381.7         335         No protein         Processed transcript         -           ENSMUST00000176672.1         328         No protein         Processed transcript         -           ENSMUST00000144873.1         2135         No protein         Processed transcript	ENSMUST00000150727.7         1523         201aa         Protein coding         CCDS26838         Q91W82           ENSMUST00000076133.9         606         201aa         Protein coding         CCDS26838         Q91W82           ENSMUST00000175643.7         757         167aa         Protein coding         -         H3BL69           ENSMUST00000151926.7         666         181aa         Protein coding         -         D3YW10           ENSMUST00000124353.1         445         108aa         Protein coding         -         D3YW10           ENSMUST00000176643.7         438         146aa         Protein coding         -         H3BL23           ENSMUST00000176555.7         1200         92aa         Nonsense mediated decay         -         H3BJB5           ENSMUST00000176062.2         647         No protein         Processed transcript         -         -           ENSMUST00000177398.7         514         No protein         Processed transcript         -         -           ENSMUST00000135381.7         335         No protein         Processed transcript         -         -           ENSMUST00000176672.1         328         No protein         Processed transcript         -         -           ENSMUST000001448873.1 <t< td=""></t<>

The strategy is based on the design of *Ube2e2-205* transcript, The transcription is shown below



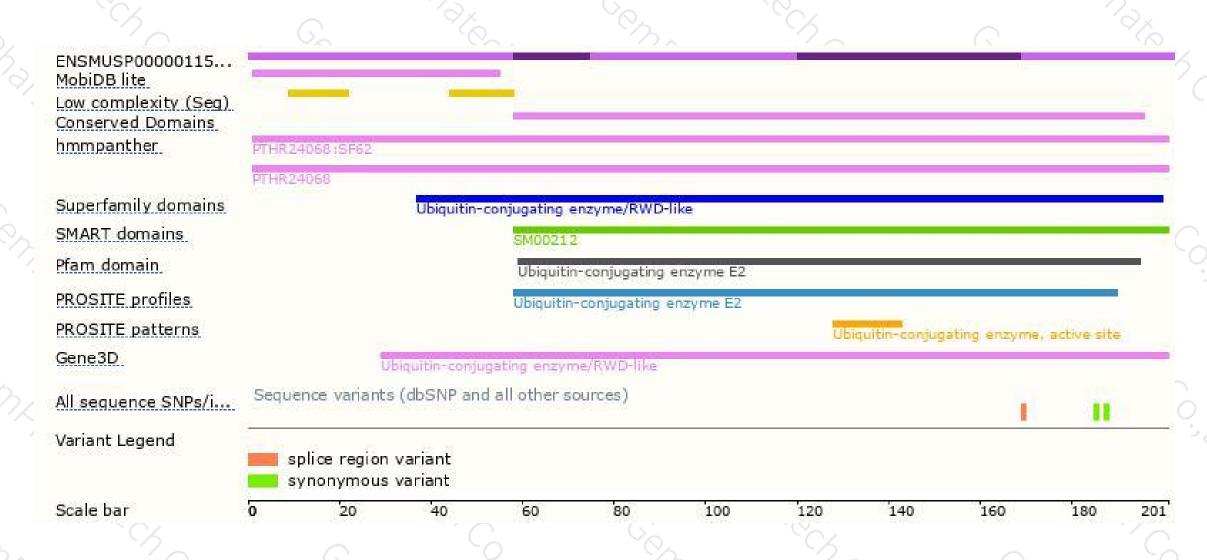
### Genomic location distribution





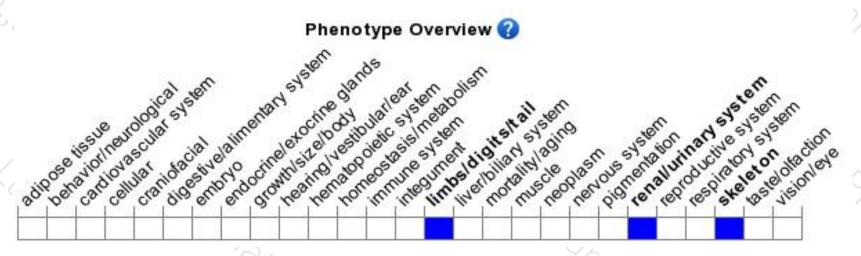
### Protein domain





# Mouse phenotype description(MGI)





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



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





