

Ube2i Cas9-CKO Strategy

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Design Date: 2019-8-15

Project Overview

Project Name

Ube2i

Project type

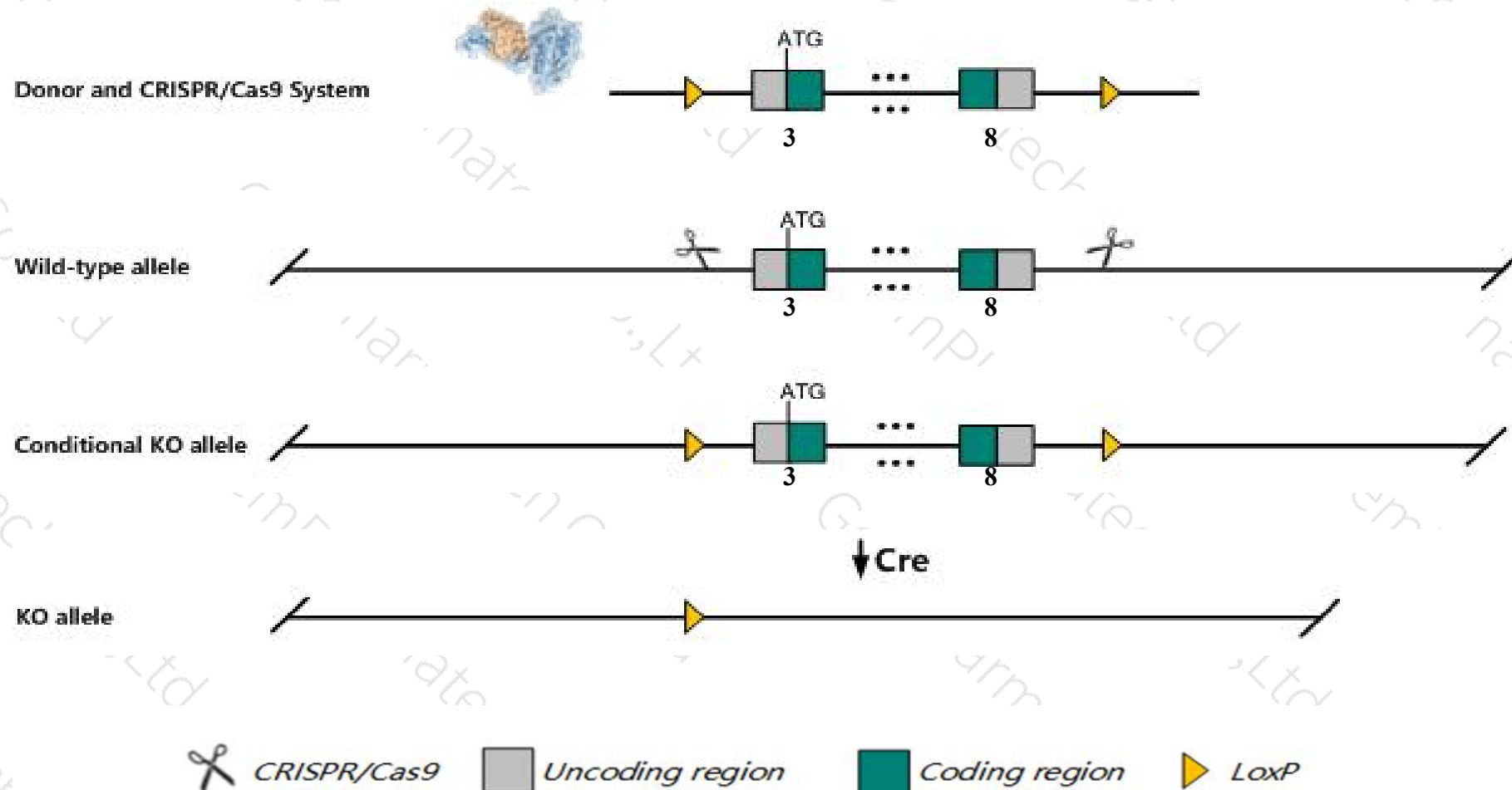
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ube2i* gene. The schematic diagram is as follows:



- The *Ube2i* gene has 14 transcripts. According to the structure of *Ube2i* gene, exon3-exon8 of *Ube2i*-206 (ENSMUST00000172618.7) transcript is recommended as the knockout region. The region contains all 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 *Ube2i* 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.

- According to the existing MGI data, Embryos homozygous for a targeted null mutation die prior to E7.5. In culture, mutant blastocysts are viable up to 2 days but show subsequent apoptosis of the inner cell mass. Mutant cells exhibit major chromosome condensation and segregation defects as well as gross defects in nuclear organization.
- The *Ube2i* 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.

Gene information (NCBI)

Ube2i ubiquitin-conjugating enzyme E2I [Mus musculus (house mouse)]

Gene ID: 22196, updated on 2-Apr-2019

Summary



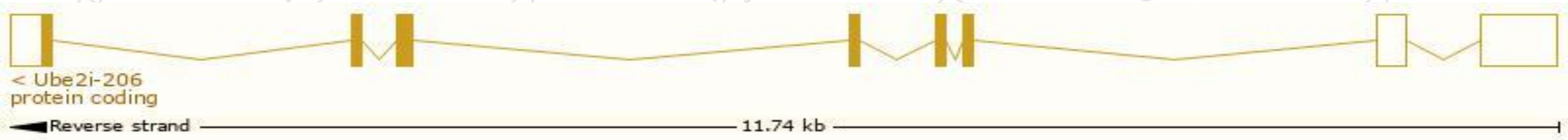
Official Symbol	Ube2i provided by MGI
Official Full Name	ubiquitin-conjugating enzyme E2I provided by MGI
Primary source	MGI:MGI:107365
See related	Ensembl:ENSMUSG00000015120
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	5830467E05Rik, UBC9, Ubce2i, Ubce9
Expression	Ubiquitous expression in CNS E11.5 (RPKM 68.8), whole brain E14.5 (RPKM 68.0) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

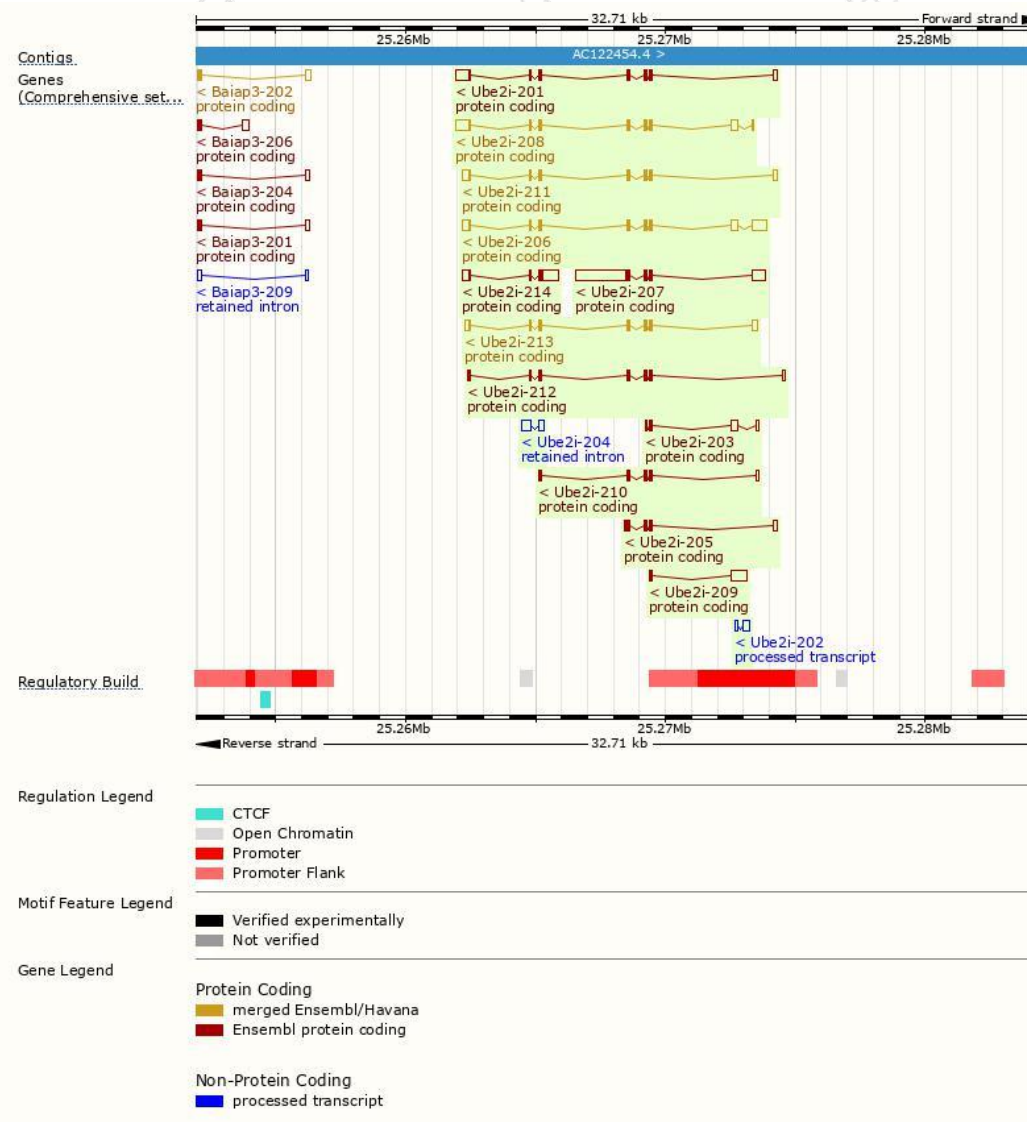
The gene has 14 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ube2i-206	ENSMUST00000172618.7	1559	158aa	Protein coding	CCDS28513	P63280	TSL:1 GENCODE basic APPRIS P1
Ube2i-208	ENSMUST00000173084.7	1286	158aa	Protein coding	CCDS28513	P63280	TSL:1 GENCODE basic APPRIS P1
Ube2i-201	ENSMUST00000049911.15	1138	158aa	Protein coding	CCDS28513	P63280	TSL:3 GENCODE basic APPRIS P1
Ube2i-211	ENSMUST00000173713.7	890	158aa	Protein coding	CCDS28513	P63280	TSL:1 GENCODE basic APPRIS P1
Ube2i-213	ENSMUST00000174031.7	824	158aa	Protein coding	CCDS28513	P63280	TSL:1 GENCODE basic APPRIS P1
Ube2i-212	ENSMUST00000174001.7	623	158aa	Protein coding	CCDS28513	P63280	TSL:3 GENCODE basic APPRIS P1
Ube2i-207	ENSMUST00000172868.7	2755	101aa	Protein coding	-	Q8CFZ0	TSL:1 GENCODE basic
Ube2i-214	ENSMUST00000174216.7	1144	101aa	Protein coding	-	G3UZL6	TSL:2 GENCODE basic
Ube2i-209	ENSMUST00000173231.1	696	22aa	Protein coding	-	G3UWL6	CDS 3' incomplete TSL:2
Ube2i-205	ENSMUST00000172587.7	508	101aa	Protein coding	-	Q8CFZ0	TSL:2 GENCODE basic
Ube2i-203	ENSMUST00000172520.1	447	29aa	Protein coding	-	G3UWJ1	CDS 3' incomplete TSL:3
Ube2i-210	ENSMUST00000173621.7	429	103aa	Protein coding	-	G3UYP0	CDS 3' incomplete TSL:3
Ube2i-202	ENSMUST00000172462.1	366	No protein	Processed transcript	-	-	TSL:3
Ube2i-204	ENSMUST00000172569.1	594	No protein	Retained intron	-	-	TSL:2

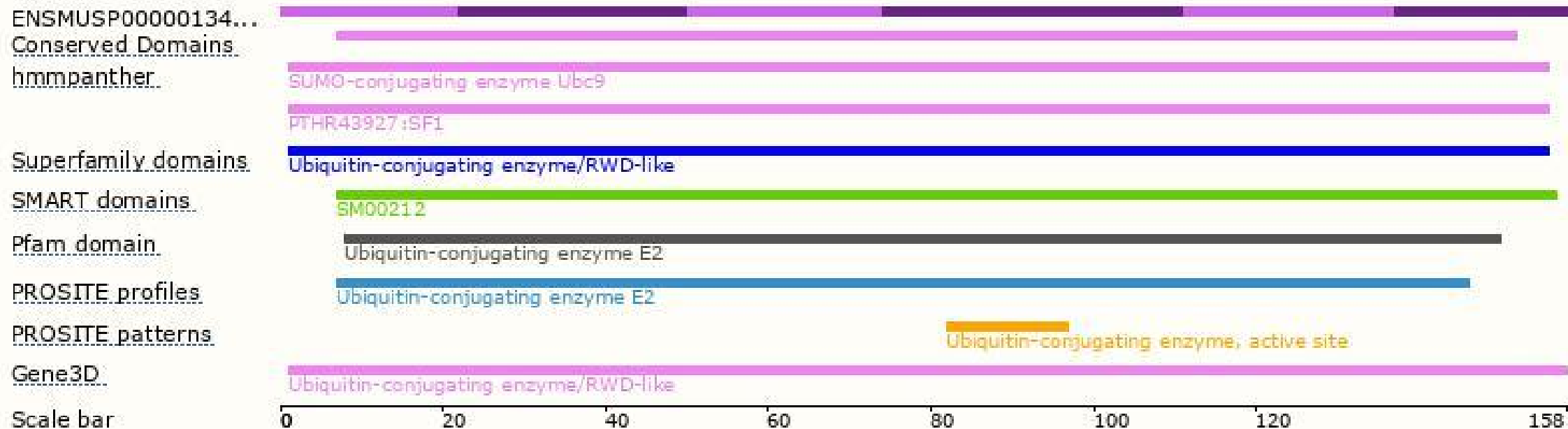
The strategy is based on the design of *Ube2i-206* transcript,The transcription is shown below



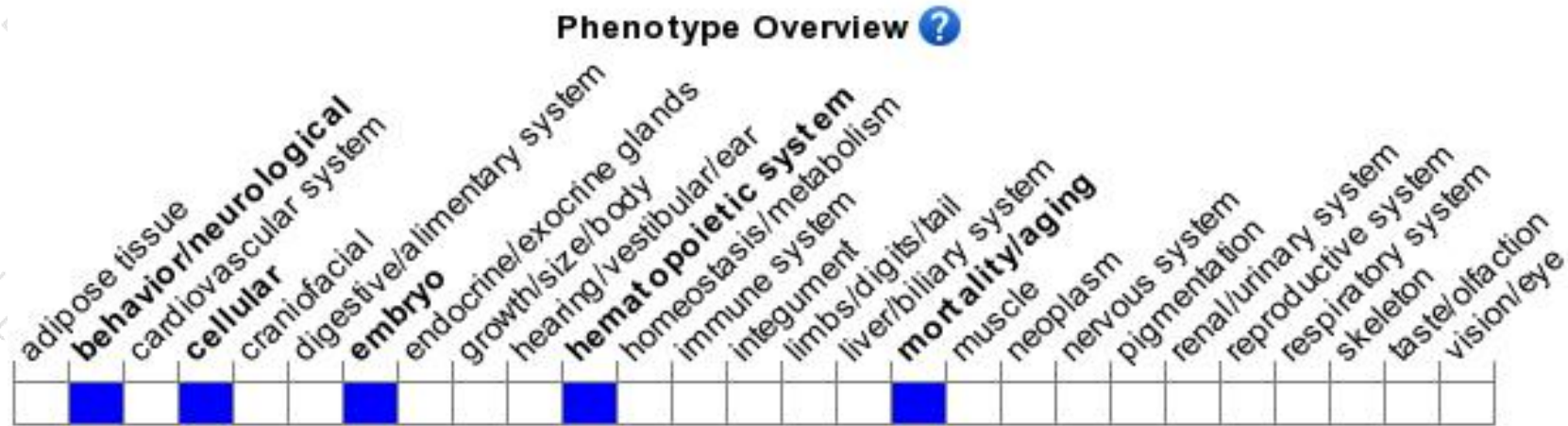
Genomic location distribution



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, Embryos homozygous for a targeted null mutation die prior to E7.5. In culture, mutant blastocysts are viable up to 2 days but show subsequent apoptosis of the inner cell mass. Mutant cells exhibit major chromosome condensation and segregation defects as well as gross defects in nuclear organization.

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

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