

***Ubr5* Cas9-CKO Strategy**

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Design Date: 2019-08-07

Project Overview

Project Name

Ubr5

Project type

Cas9-CKO

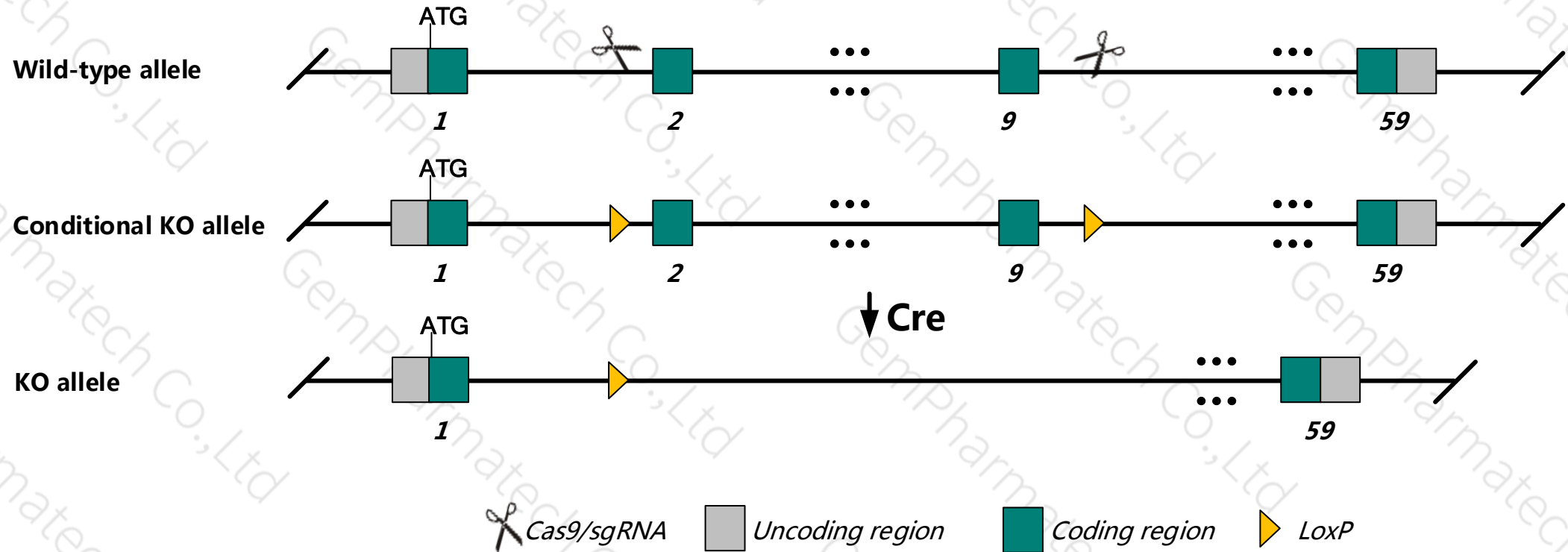
Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ubr5* gene. The schematic diagram is as follows:

Donor and CRISPR/Cas9 System



Technical routes

- The *Ubr5* gene has 14 transcripts. According to the structure of *Ubr5* gene, exon2-exon9 of *Ubr5-201* (ENSMUST00000110336.3) transcript is recommended as the knockout region. The region contains 1018bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ubr5* 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.

- According to the existing MGI data, Homozygous null mice display embryonic lethality during organogenesis, impaired growth of the allantois, failure or impairment of chorioallantoic fusion, impaired angiogenesis in the yolk sac and allantois, decreased cell proliferation, and increased apoptosis.
- The *Ubr5* gene is located on the Chr15. 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)

Ubr5 ubiquitin protein ligase E3 component n-recognin 5 [Mus musculus (house mouse)]

Gene ID: 70790, updated on 3-Feb-2019

Summary



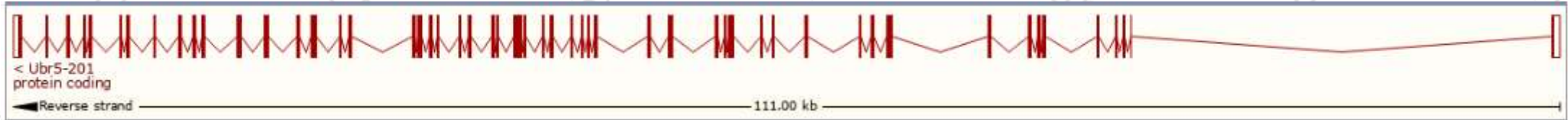
Official Symbol	Ubr5 provided by MGI
Official Full Name	ubiquitin protein ligase E3 component n-recognin 5 provided by MGI
Primary source	MGI:MGI:1918040
See related	Ensembl:ENSMUSG00000037487
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Edd, Edd1
Expression	Ubiquitous expression in testis adult (RPKM 22.8), CNS E11.5 (RPKM 15.9) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

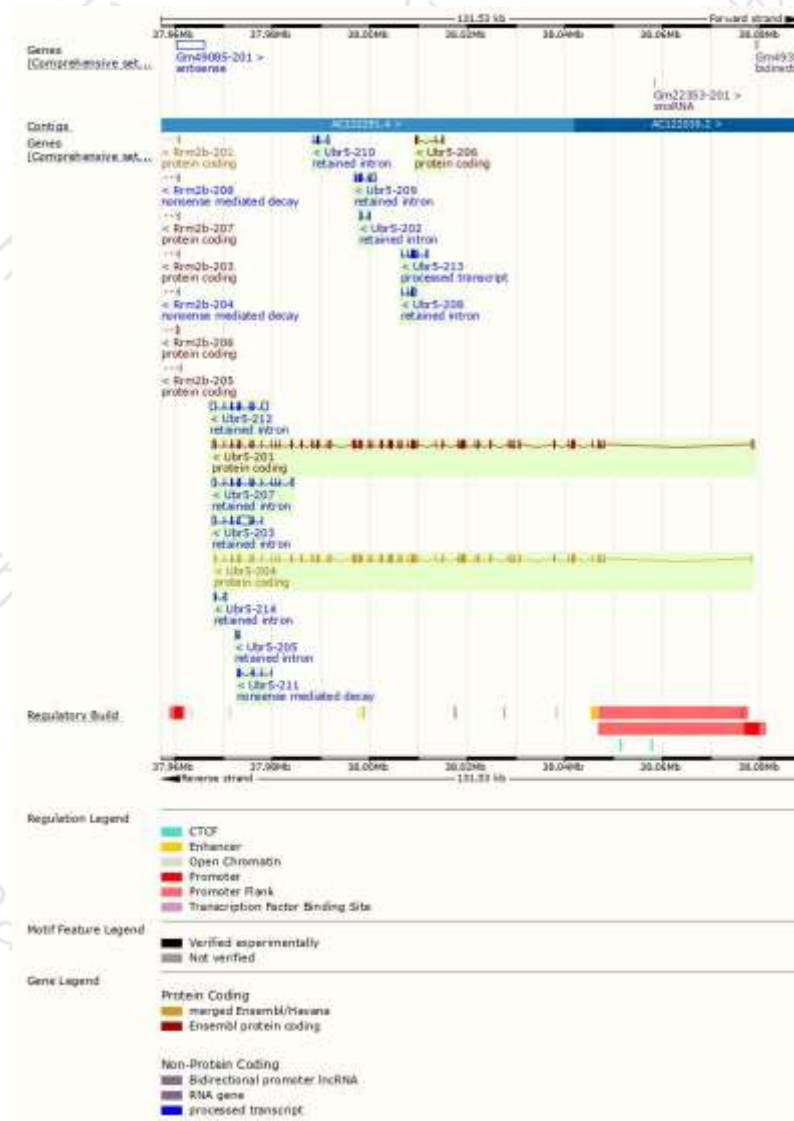
The gene has 14 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ubr5-204	ENSMUST00000226414.1	8397	2798aa	Protein coding	CCDS49595	A0A2I3BQS6	GENCODE basic APPRIS P2
Ubr5-201	ENSMUST00000110336.3	9242	2792aa	Protein coding	-	E9Q2H1	TSL:5 GENCODE basic APPRIS ALT 1
Ubr5-206	ENSMUST00000227143.1	596	142aa	Protein coding	-	A0A2I3BPG8	CDS 5' incomplete
Ubr5-211	ENSMUST00000228333.1	507	101aa	Nonsense mediated decay	-	A0A2I3BQC1	CDS 5' incomplete
Ubr5-213	ENSMUST00000228504.1	654	No protein	Processed transcript	-	-	
Ubr5-203	ENSMUST00000226369.1	3562	No protein	Retained intron	-	-	
Ubr5-212	ENSMUST00000228368.1	2953	No protein	Retained intron	-	-	
Ubr5-207	ENSMUST00000228029.1	2279	No protein	Retained intron	-	-	
Ubr5-209	ENSMUST00000228174.1	1360	No protein	Retained intron	-	-	
Ubr5-210	ENSMUST00000228292.1	707	No protein	Retained intron	-	-	
Ubr5-202	ENSMUST00000226137.1	664	No protein	Retained intron	-	-	
Ubr5-208	ENSMUST00000228101.1	634	No protein	Retained intron	-	-	
Ubr5-214	ENSMUST00000228804.1	522	No protein	Retained intron	-	-	
Ubr5-205	ENSMUST00000226629.1	457	No protein	Retained intron	-	-	

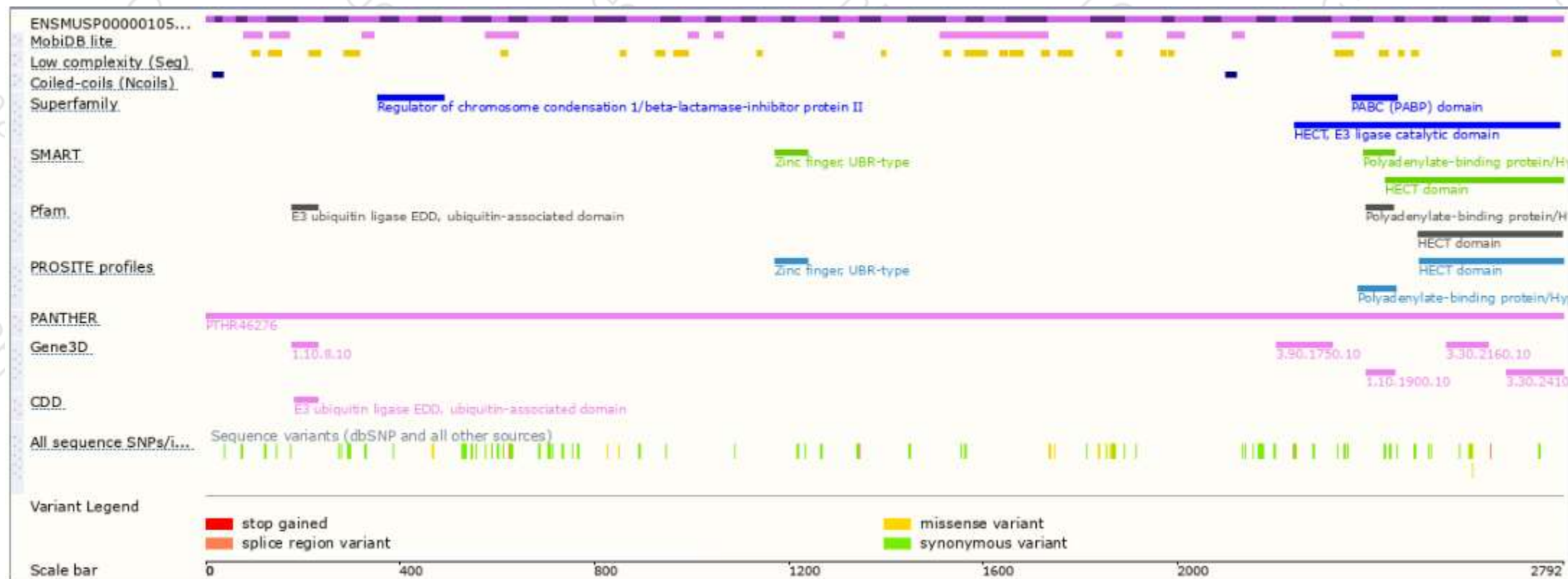
The strategy is based on the design of *Ubr5-201* transcript,The transcription is shown below



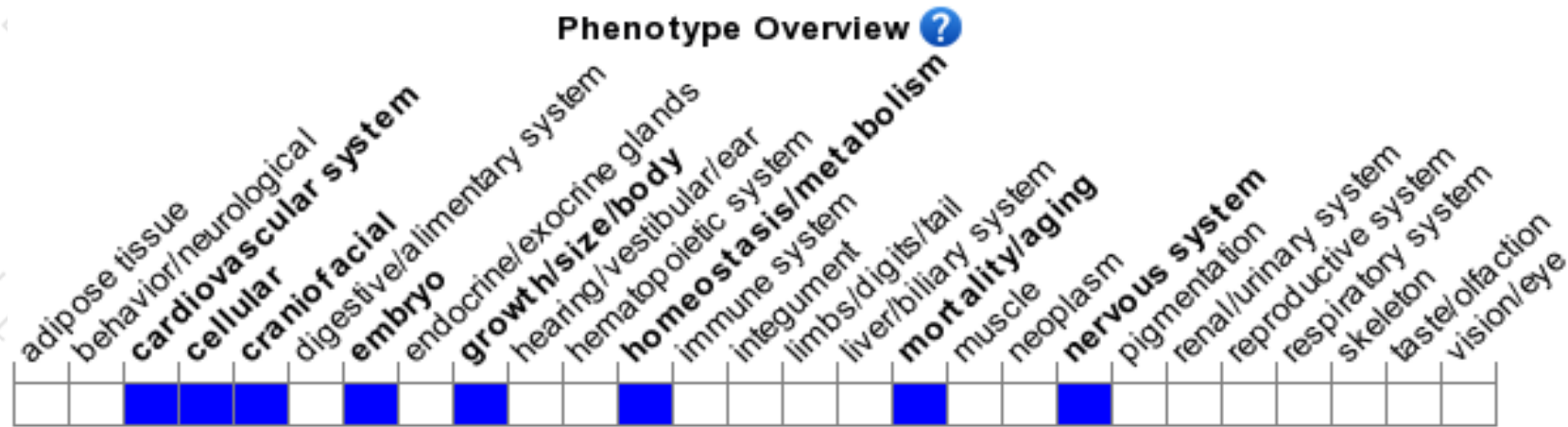
Genomic location distribution



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, Homozygous null mice display embryonic lethality during organogenesis, impaired growth of the allantois, failure or impairment of chorioallantoic fusion, impaired angiogenesis in the yolk sac and allantois, decreased cell proliferation, and increased apoptosis.

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

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