

Hecw2 Cas9-CKO Strategy

Designer

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Reviewer

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2018-6-11

Project Overview

Project Name

Hecw2

Project type

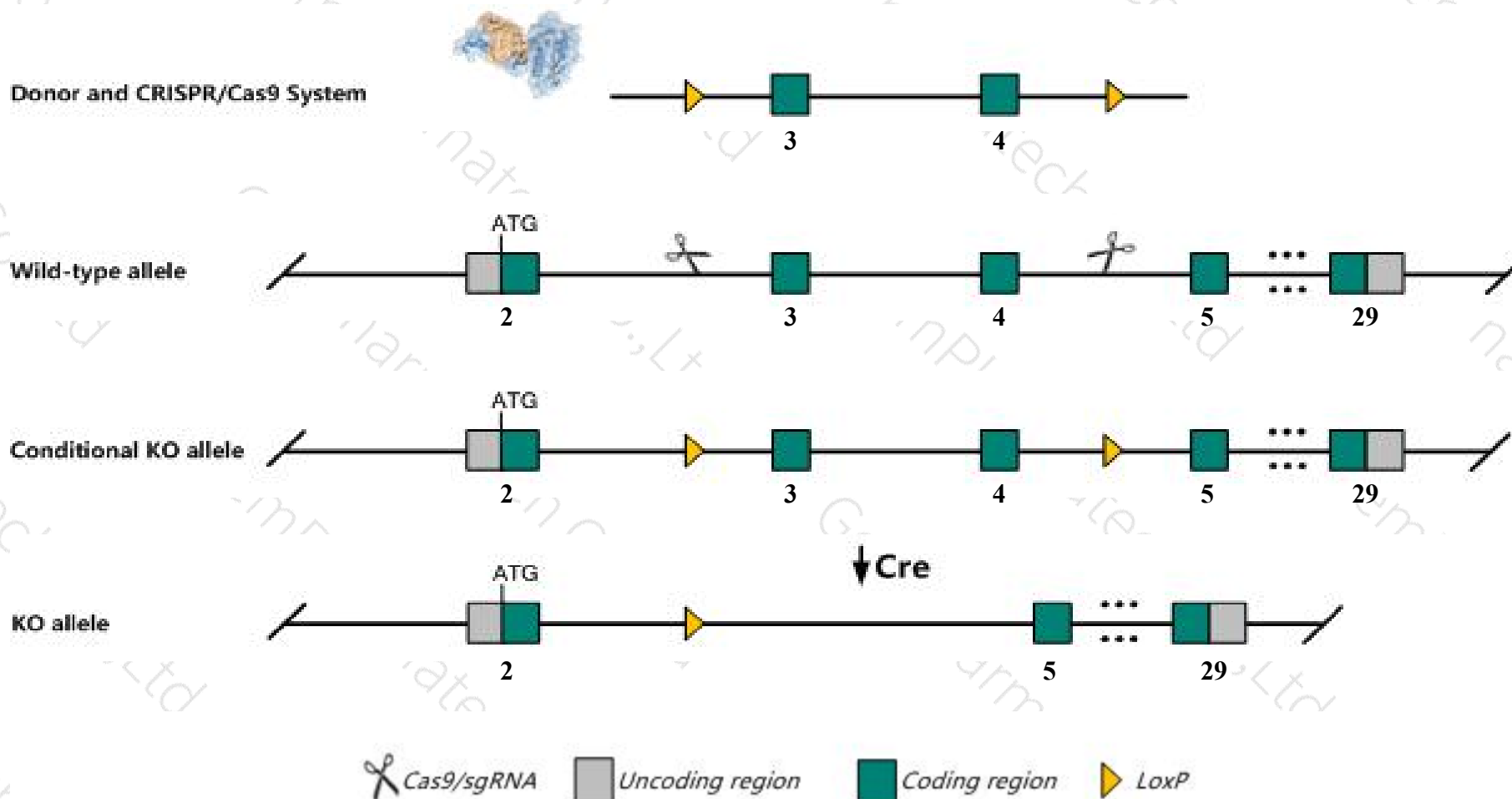
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Hecw2* gene. The schematic diagram is as follows:



- The *Hecw2* gene has 6 transcripts. According to the structure of *Hecw2* gene, exon3-exon4 of *Hecw2*-201 (ENSMUST00000087659.10) transcript is recommended as the knockout region. The region contains 203bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Hecw2* 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

- Transcript *Hecw2*-205/206 may not be affected.
- The *Hecw2* gene is located on the Chr1. 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)

Hecw2 HECT, C2 and WW domain containing E3 ubiquitin protein ligase 2 [Mus musculus (house mouse)]

Gene ID: 329152, updated on 25-Mar-2019

Summary



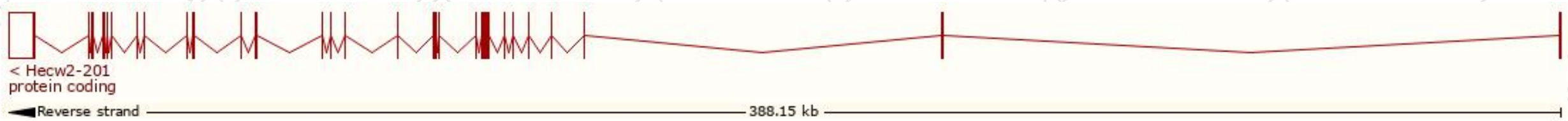
Official Symbol	Hecw2 provided by MGI
Official Full Name	HECT, C2 and WW domain containing E3 ubiquitin protein ligase 2 provided by MGI
Primary source	MGI:MGI:2685817
See related	Ensembl:ENSMUSG00000042807
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	A730039N16Rik, D030049F17Rik, Gm971, Nedl2, mKIAA1301
Expression	Broad expression in cortex adult (RPKM 2.4), lung adult (RPKM 2.2) and 19 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

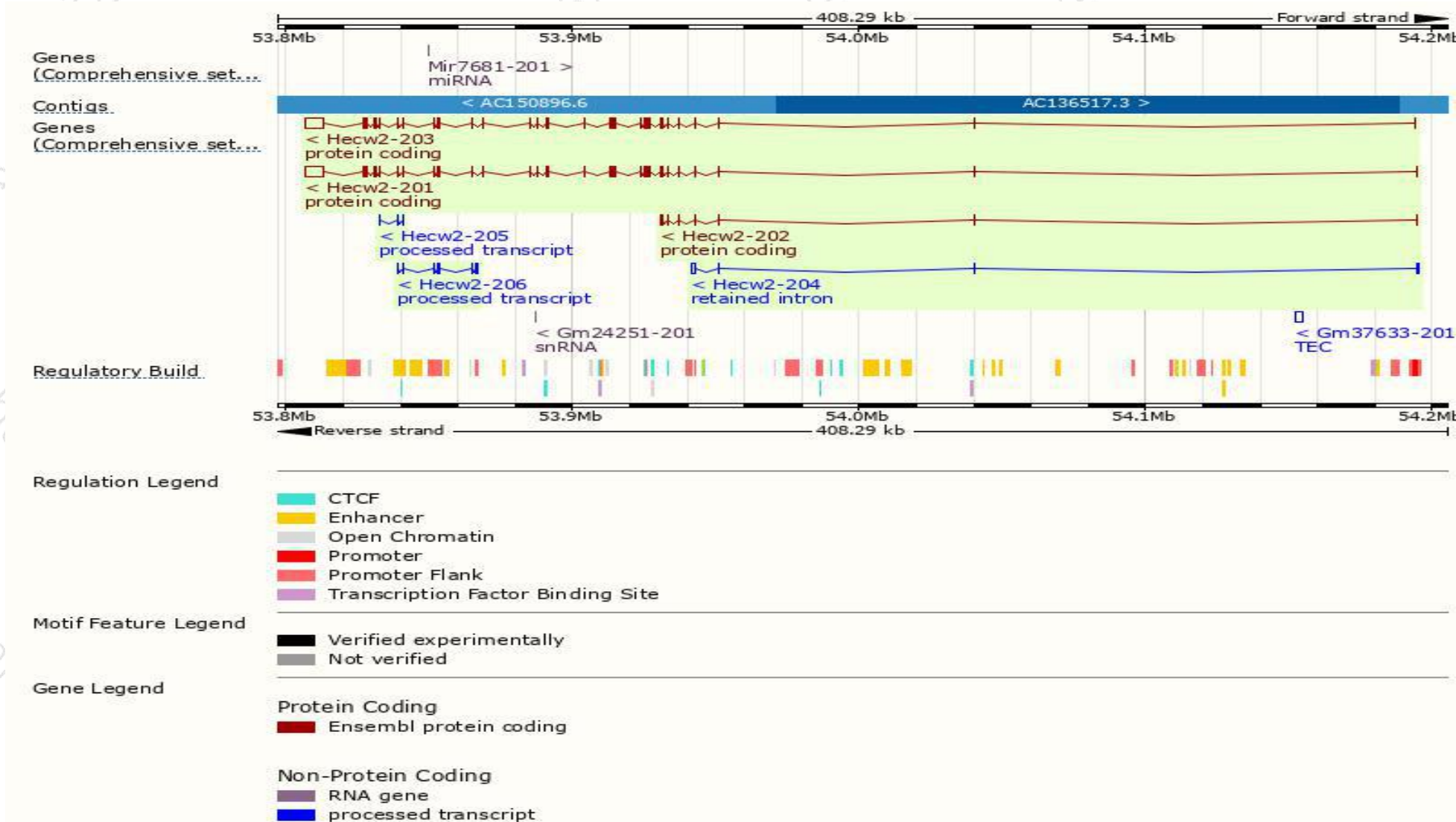
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Hecw2-203	ENSMUST00000120904.7	11263	1578aa	Protein coding	CCDS14956	Q6I6G8	TSL:5 GENCODE basic APPRIS P1
Hecw2-201	ENSMUST00000087659.10	11171	1578aa	Protein coding	CCDS14956	Q6I6G8	TSL:5 GENCODE basic APPRIS P1
Hecw2-202	ENSMUST00000097741.2	1132	294aa	Protein coding	CCDS14957	A3KPB7 Q6I6G8	TSL:1 GENCODE basic
Hecw2-206	ENSMUST00000152870.1	665	No protein	Processed transcript	-	-	TSL:2
Hecw2-205	ENSMUST00000150677.7	363	No protein	Processed transcript	-	-	TSL:3
Hecw2-204	ENSMUST00000146850.1	1933	No protein	Retained intron	-	-	TSL:1

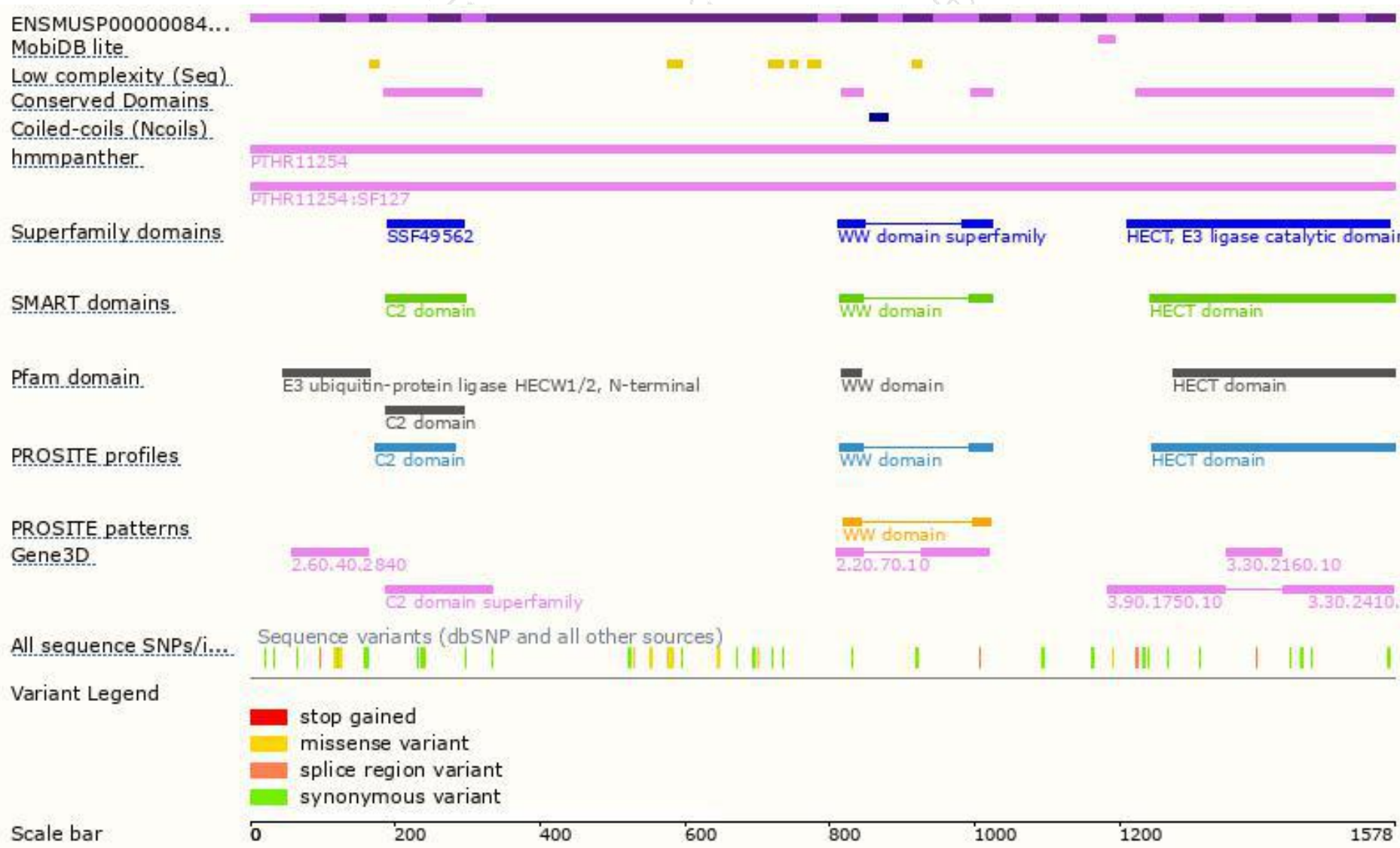
The strategy is based on the design of *Hecw2-201* transcript,The transcription is shown below



Genomic location distribution



Protein domain



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

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