

# *Wwp2* Cas9-CKO Strategy

**Designer**

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**Reviewer**

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**Design Date**

**2018-6-12**

# Project Overview

**Project Name**

*Wwp2*

**Project type**

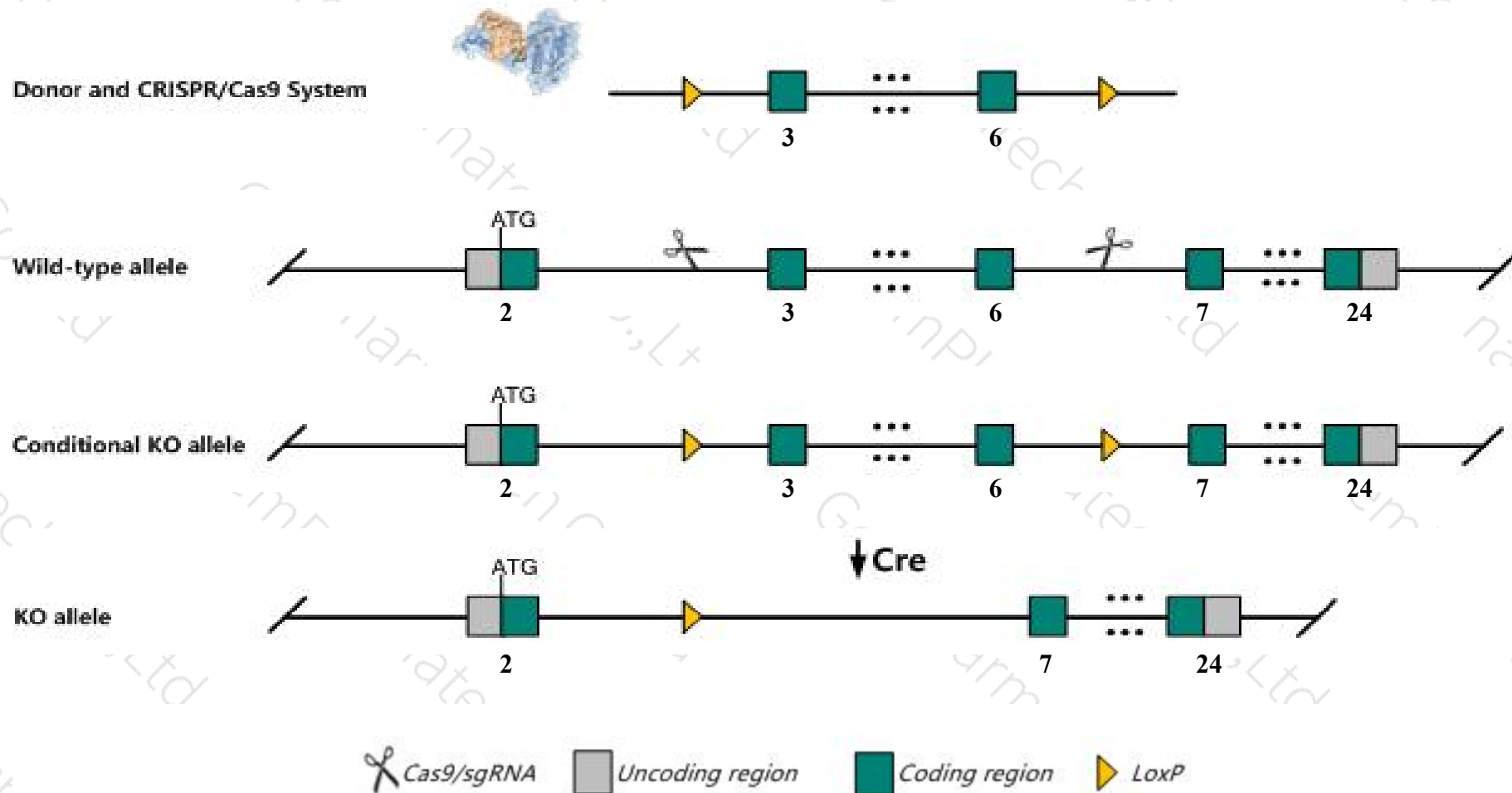
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Wwp2* gene. The schematic diagram is as follows:



- The *Wwp2* gene has 11 transcripts. According to the structure of *Wwp2* gene, exon3-exon6 of *Wwp2-201* (ENSMUST00000166615.2) transcript is recommended as the knockout region. The region contains 505bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Wwp2* 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, Mice homozygous for a gene trapped allele exhibit decreased body size, domed skull, short snout, twisted snout and overgrown mandibular incisors. Mice homozygous for a different knock-out allele exhibit increased sensitivity to pIpC-treatment.
- Transcript *Wwp2-202/205/206/208/210* may not be affected.
- The *Wwp2* gene is located on the Chr8. 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)

## Wwp2 WW domain containing E3 ubiquitin protein ligase 2 [Mus musculus (house mouse)]

Gene ID: 66894, updated on 19-Mar-2019

### Summary



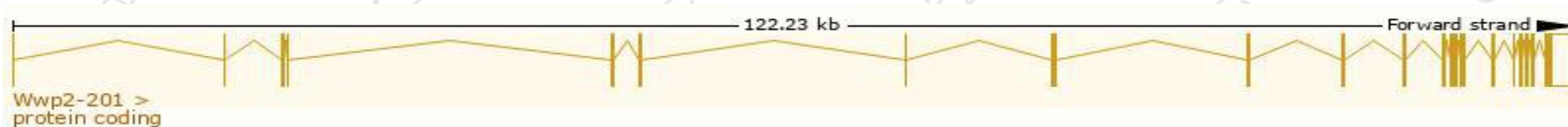
<b>Official Symbol</b>	Wwp2 provided by <a href="#">MGI</a>
<b>Official Full Name</b>	WW domain containing E3 ubiquitin protein ligase 2 provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:1914144</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000031930</a>
<b>Gene type</b>	protein coding
<b>RefSeq status</b>	VALIDATED
<b>Organism</b>	<a href="#">Mus musculus</a>
<b>Lineage</b>	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
<b>Also known as</b>	1300010O06Rik, AA690238, AIP2, AW554328
<b>Expression</b>	Ubiquitous expression in limb E14.5 (RPKM 69.1), testis adult (RPKM 34.2) and 28 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

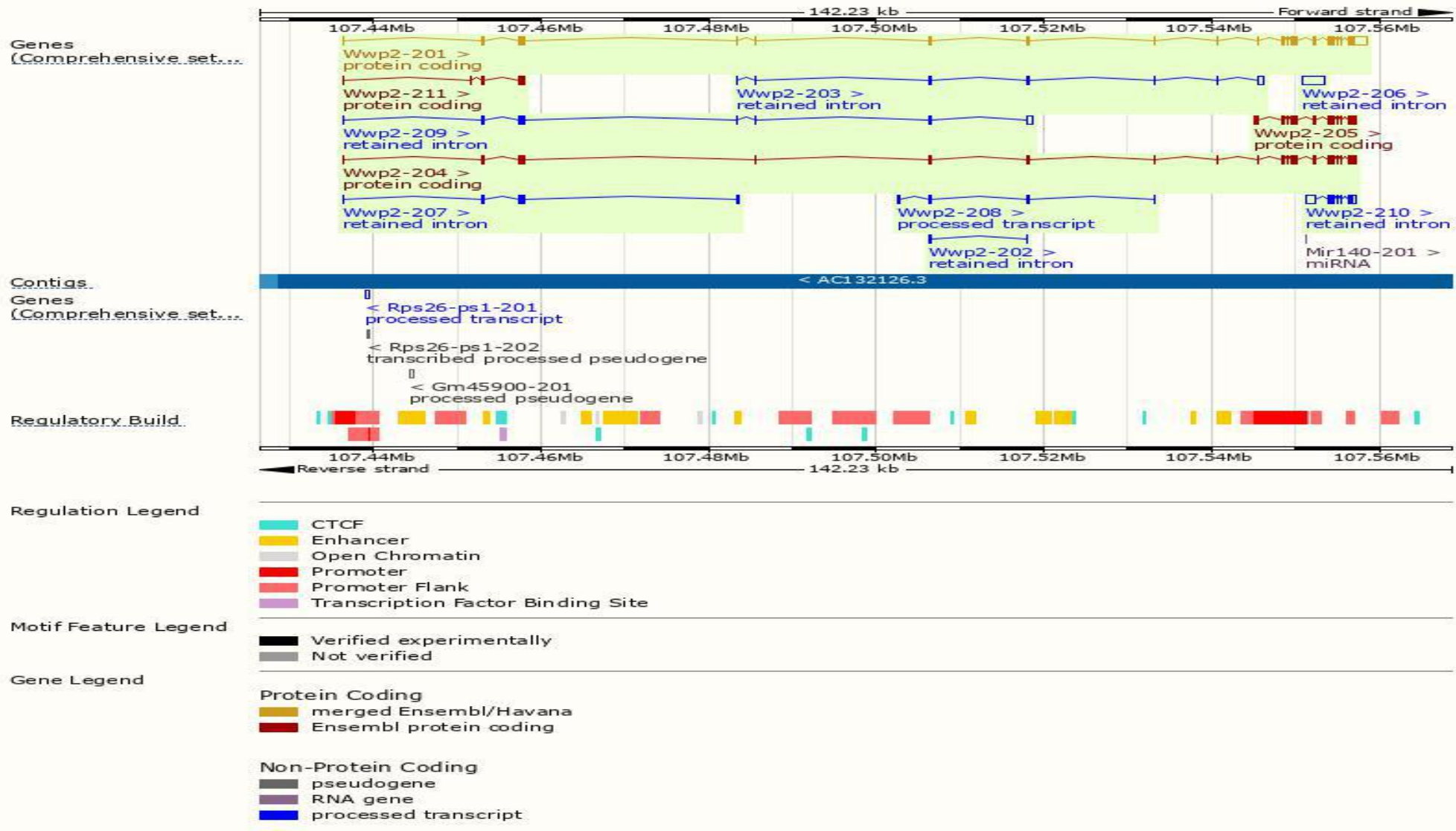
The gene has 11 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Wwp2-201	<a href="#">ENSMUST00000166615.2</a>	4314	<a href="#">870aa</a>	Protein coding	<a href="#">CCDS40467</a>	<a href="#">I3RSH5_Q9DBH0</a>	TSL:1 GENCODE basic APPRIS P1
Wwp2-204	<a href="#">ENSMUST00000212205.1</a>	2758	<a href="#">824aa</a>	Protein coding	-	<a href="#">A0A1D5RM92</a>	TSL:1 GENCODE basic
Wwp2-205	<a href="#">ENSMUST00000212543.1</a>	1996	<a href="#">431aa</a>	Protein coding	-	<a href="#">Q3TXI7</a>	TSL:1 GENCODE basic
Wwp2-211	<a href="#">ENSMUST00000213097.1</a>	425	<a href="#">104aa</a>	Protein coding	-	<a href="#">A0A1D5RLH0</a>	CDS 3' incomplete TSL:3
Wwp2-208	<a href="#">ENSMUST00000212737.1</a>	623	No protein	Processed transcript	-	-	TSL:2
Wwp2-206	<a href="#">ENSMUST00000212559.1</a>	2653	No protein	Retained intron	-	-	TSL:NA
Wwp2-210	<a href="#">ENSMUST00000212993.1</a>	2098	No protein	Retained intron	-	-	TSL:1
Wwp2-209	<a href="#">ENSMUST00000212906.1</a>	1579	No protein	Retained intron	-	-	TSL:1
Wwp2-203	<a href="#">ENSMUST00000212063.1</a>	1548	No protein	Retained intron	-	-	TSL:1
Wwp2-207	<a href="#">ENSMUST00000212645.1</a>	640	No protein	Retained intron	-	-	TSL:2
Wwp2-202	<a href="#">ENSMUST00000212012.1</a>	329	No protein	Retained intron	-	-	TSL:1

The strategy is based on the design of *Wwp2-201* transcript, The transcription is shown below



# Genomic location distribution

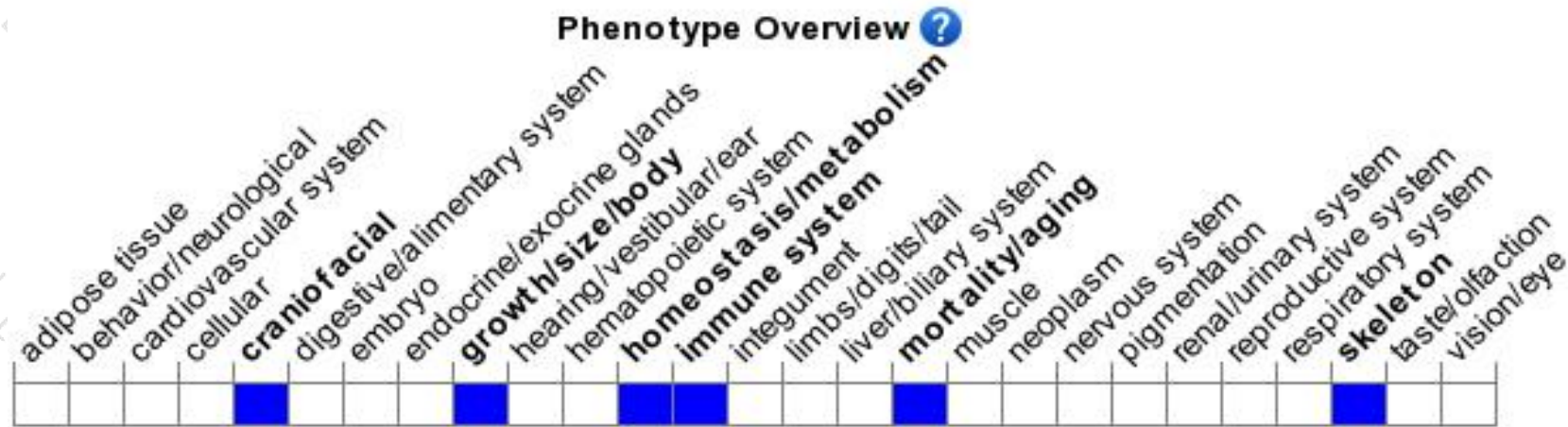




# Protein domain



# 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, Mice homozygous for a gene trapped allele exhibit decreased body size, domed skull, short snout, twisted snout and overgrown mandibular incisors. Mice homozygous for a different knock-out allele exhibit increased sensitivity to pIpC-treatment.

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

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