

# Piezo2 Cas9-CKO Strategy

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Reviewer:

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# **Project Overview**



**Project Name** 

Piezo2

**Project type** 

Cas9-CKO

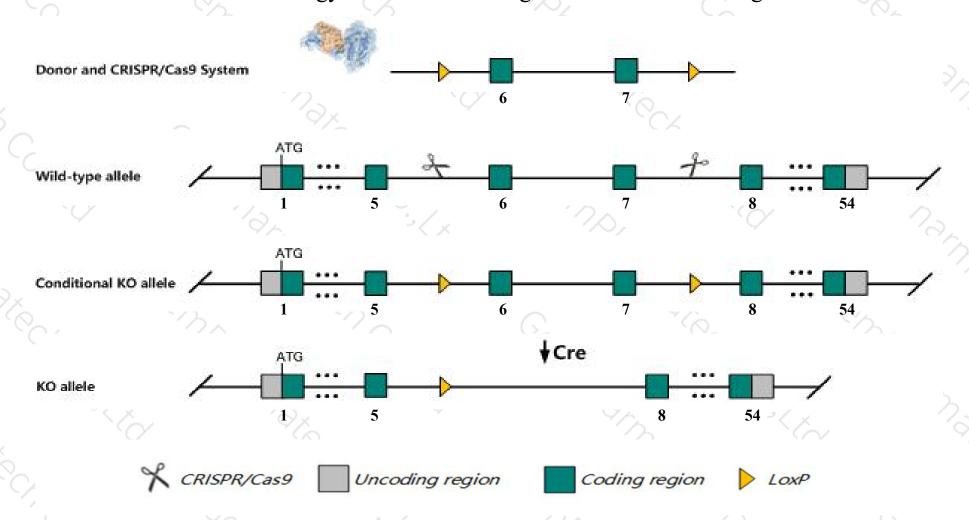
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Piezo2* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Piezo2* gene has 15 transcripts. According to the structure of *Piezo2* gene, exon6-exon7 of *Piezo2-202*(ENSMUST00000047480.12) transcript is recommended as the knockout region. The region contains 425bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Piezo2* 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 *Piezo2* gene is located on the Chr18. 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)



#### Piezo2 piezo-type mechanosensitive ion channel component 2 [Mus musculus (house mouse)]

Gene ID: 667742, updated on 31-Jan-2019

#### Summary



Official Symbol Piezo2 provided by MGI

Official Full Name piezo-type mechanosensitive ion channel component 2 provided by MGI

Primary source MGI:MGI:1918781

See related Ensembl:ENSMUSG00000041482

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 5930434P17, 9030411M15Rik, 9430028L06Rik, Fam38b, Fam38b2

Expression Biased expression in limb E14.5 (RPKM 12.4), lung adult (RPKM 5.9) and 9 other tissuesSee more

Orthologs <u>human all</u>

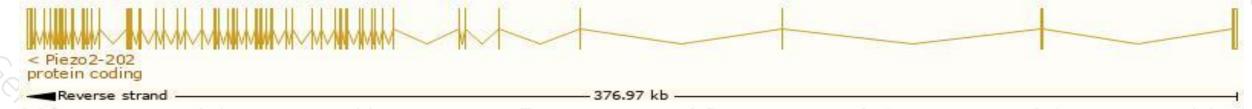
# Transcript information (Ensembl)



The gene has 15 transcripts, all transcripts are shown below:

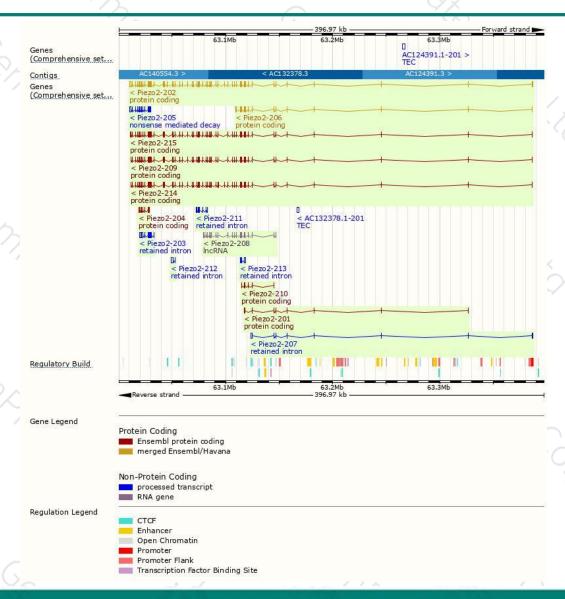
all the							
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Piezo2-202	ENSMUST00000047480.12	10724	2824aa	Protein coding	CCDS29296	E9QNW4	TSL:1 GENCODE basic APPRIS P2
Piezo2-215	ENSMUST00000238156.1	8773	2690aa	Protein coding	10-	-	GENCODE basic
Piezo2-209	ENSMUST00000183217.8	8564	2753aa	Protein coding	-	S4R2S0	TSL:5 GENCODE basic APPRIS ALT2
Piezo2-214	ENSMUST00000238051.1	8337	2778aa	Protein coding	12	2	GENCODE basic APPRIS ALT2
Piezo2-206	ENSMUST00000182166.8	1988	644aa	Protein coding		S4R2R6	TSL:5 GENCODE basic
Piezo2-204	ENSMUST00000132576.8	1318	378aa	Protein coding	19-	-	CDS 5' incomplete TSL:1
Piezo2-201	ENSMUST00000046860.6	1125	<u>375aa</u>	Protein coding	12-	Q8CD54	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:
Piezo2-210	ENSMUST00000235366.1	689	230aa	Protein coding	12	-	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete
Piezo2-205	ENSMUST00000137141.3	3200	409aa	Nonsense mediated decay	0.5	F6YYK4	CDS 5' incomplete TSL:1
Piezo2-203	ENSMUST00000123322.1	3739	No protein	Retained intron		-	TSL:1
Piezo2-207	ENSMUST00000182177.2	2963	No protein	Retained intron	<u> </u>	ū.	TSL:1
Piezo2-213	ENSMUST00000237812.1	1803	No protein	Retained intron	12	2	
Piezo2-212	ENSMUST00000236438.1	1452	No protein	Retained intron	15	5	
Piezo2-211	ENSMUST00000235659.1	889	No protein	Retained intron	-	-	
Piezo2-208	ENSMUST00000182233.8	3459	No protein	IncRNA	ŞI.	9	TSL:5
7 .				7//		/ 1	

The strategy is based on the design of *Piezo2-202* transcript, The transcription is shown below



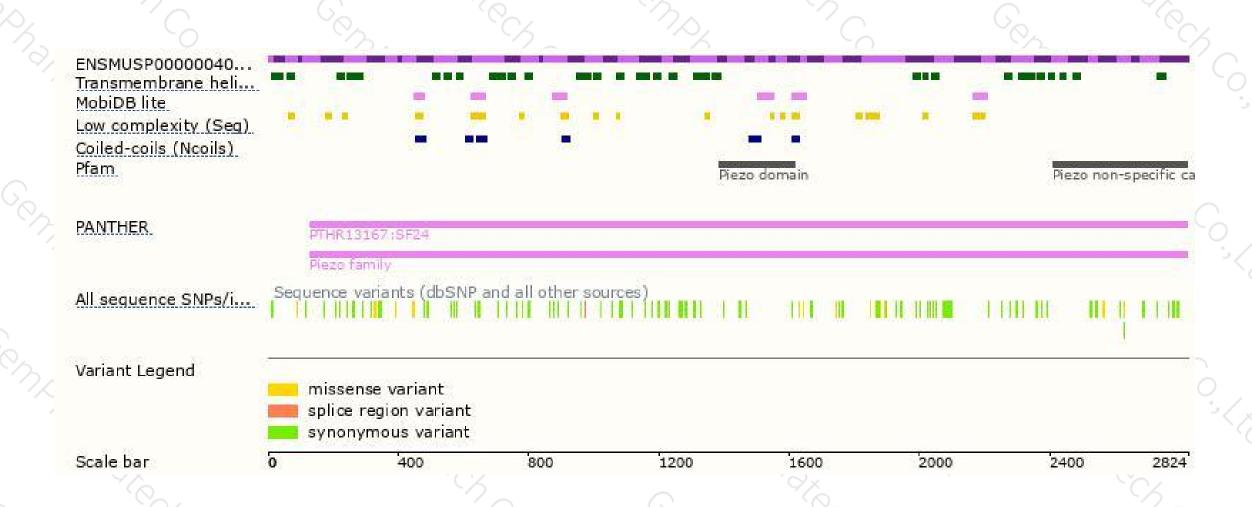
### Genomic location distribution





### Protein domain







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





