

# Macf1 Cas9-CKO Strategy

**Designer:** 

Reviewer:

**Design Date:** 

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2019-9-19

## **Project Overview**



**Project Name** 

Macf1

**Project type** 

Cas9-CKO

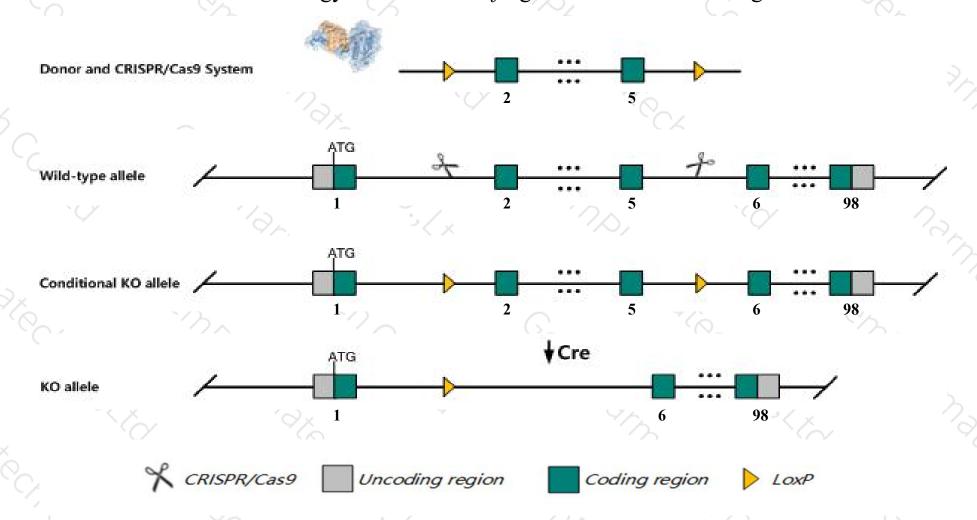
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Macf1* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Macf1* gene has 18 transcripts. According to the structure of *Macf1* gene, exon2-exon5 of *Macf1-203*(ENSMUST00000097897.10) transcript is recommended as the knockout region. The region contains 323bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Macf1* 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**



- > According to the existing MGI data, Mice homozygous for a null allele exhibit lethality before somitogenesis with failure of the primitive streak to form. Mice heterozygous for a knock-out and floxed allele activated in neurons exhibit impaired cortical neuron migration, respiratory distress, and early postnatal lethality.
- Transcript *Macf1-204,207,208,209,210,214,216* may not be affected.
- The *Macf1* gene is located on the Chr4. 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)



#### Macf1 microtubule-actin crosslinking factor 1 [ Mus musculus (house mouse) ]

Gene ID: 11426, updated on 12-Aug-2019

#### Summary



Official Symbol Macf1 provided by MGI

Official Full Name microtubule-actin crosslinking factor 1 provided by MGI

Primary source MGI:MGI:108559

See related Ensembl:ENSMUSG00000028649

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 Acf7; MACF; Aclp7; ABP620; R74989; mKIAA0465

Expression Ubiquitous expression in lung adult (RPKM 15.9), CNS E18 (RPKM 10.5) and 28 other tissues See more

Orthologs <u>human</u> all

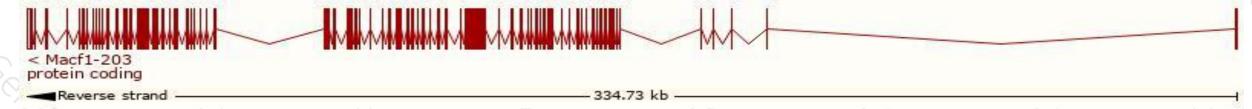
## Transcript information (Ensembl)



#### The gene has 18 transcripts, all transcripts are shown below:

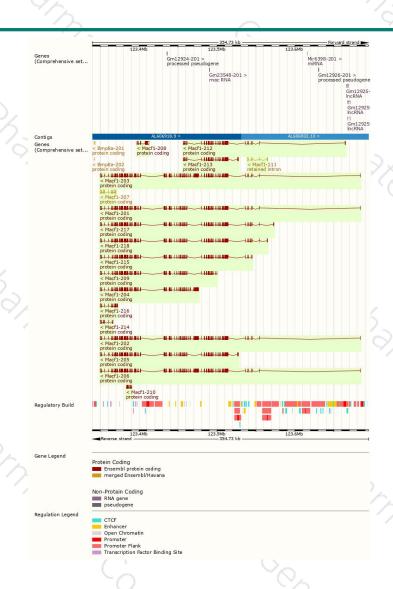
Name 🍦	Transcript ID	bp 🛊	Protein 🍦	Biotype	CCDS .	UniProt #	Flags
Macf1-203	ENSMUST00000097897.10	23495	<u>7355aa</u>	Protein coding	CCDS57295 €	E9PVY8 €	TSL:1 GENCODE basic APPRIS P2
Macf1-201	ENSMUST00000082108.11	17331	<u>5328aa</u>	Protein coding	CCDS57294₽	A0A0A0MQA6@	TSL:1 GENCODE basic
Macf1-206	ENSMUST00000106224.7	23402	7353aa	Protein coding	-	B1ARU4₽	TSL:5 GENCODE basic APPRIS ALT2
Macf1-202	ENSMUST00000084301.11	23398	<u>7351aa</u>	Protein coding	29	E9QA63 €	TSL:5 GENCODE basic APPRIS ALT2
Macf1-204	ENSMUST00000106213.7	18957	<u>5895aa</u>	Protein coding	120	B1ARU1₽	TSL:5 GENCODE basic
Macf1-205	ENSMUST00000106220.8	17608	<u>5478aa</u>	Protein coding	. 22	E9QNP1 ₽	TSL:5 GENCODE basic
Macf1-217	ENSMUST00000238555.1	17480	<u>5323aa</u>	Protein coding	32.4	-	GENCODE basic
Macf1-218	ENSMUST00000238731.1	17268	5309aa	Protein coding		-	GENCODE basic
Macf1-215	ENSMUST00000151346.7	17175	<u>5333aa</u>	Protein coding	-	F7ACR9₽	CDS 5' incomplete TSL:5
Macf1-209	ENSMUST00000134458.7	14463	4429aa	Protein coding	-	F6Q750 ₺	CDS 5' incomplete TSL:5
Macf1-213	ENSMUST00000147228.7	6281	2030aa	Protein coding	-	F6SHS0 €	CDS 3' incomplete TSL:1
Macf1-212	ENSMUST00000147030.1	5706	<u>1837aa</u>	Protein coding	29	F6XCT0₽	CDS 3' incomplete TSL:1
Macf1-208	ENSMUST00000125447.2	5597	<u>1358aa</u>	Protein coding	120	A0A286YD76₺	CDS 3' incomplete TSL:5
Macf1-216	ENSMUST00000154824.7	2808	<u>544aa</u>	Protein coding	20	F6YKN8&	CDS 5' incomplete TSL:1
Macf1-207	ENSMUST00000123765.7	2613	<u>452aa</u>	Protein coding	32.4	F6RCJ3₺	CDS 5' incomplete TSL:1
Macf1-214	ENSMUST00000149022.7	2222	<u>349aa</u>	Protein coding	350	F6RL59₽	CDS 5' incomplete TSL:5
Macf1-210	ENSMUST00000140596.1	1109	<u>369aa</u>	Protein coding	(-)	A0A0A0MQH5@	CDS 5' and 3' incomplete TSL:5
Macf1-211	ENSMUST00000146000.1	1378	No protein	Retained intron	6-7	-	TSL:1

The strategy is based on the design of Macf1-203 transcript, The transcription is shown below



### Genomic location distribution





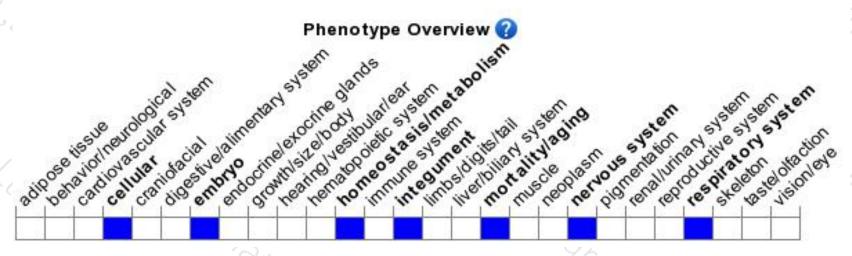
### Protein domain





## 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 null allele exhibit lethality before somitogenesis with failure of the primitive streak to form. Mice heterozygous for a knock-out and floxed allele activated in neurons exhibit impaired cortical neuron migration, respiratory distress, and early postnatal lethality.



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





