

Mecp2 Cas9-CKO Strategy

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Reviewer:

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Design Date:

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Project Overview



Project Name

Mecp2

Project type

Cas9-CKO

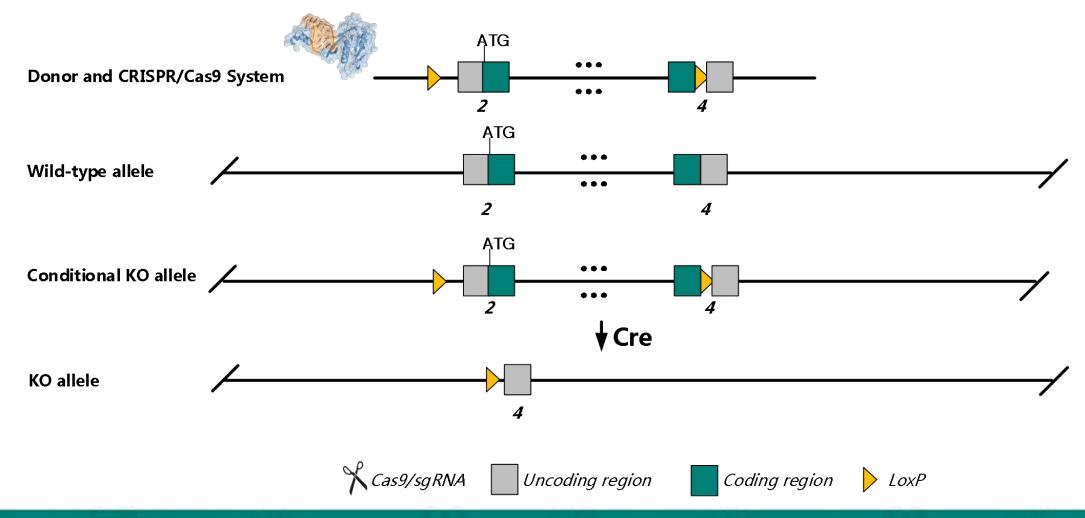
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Mecp2* gene. The schematic diagram is as follows:



Technical routes



- The *Mecp2* gene has 6 transcripts. According to the structure of *Mecp2* gene, exon2-exon4 of *Mecp2-202* (ENSMUST00000100750.9) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Mecp2* 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, Female mice homozygous or male mice hemizygous for a null allele exhibit premature death, behavioral and neurological abnormalities, abnormal nervous system phenotypes, abnormal breathing, and abnormal hearing. Heterozygous mice exhibit similar behavioral and neurological abnormalities.
- > The *Mecp2* gene is located on the ChrX. 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)



Mecp2 methyl CpG binding protein 2 [Mus musculus (house mouse)]

Gene ID: 17257, updated on 9-Apr-2019

Summary

☆ ?

Official Symbol Mecp2 provided by MGI

Official Full Name methyl CpG binding protein 2 provided by MGI

Primary source MGI:MGI:99918

See related Ensembl:ENSMUSG00000031393

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 1500041B07Rik, D630021H01Rik, Mbd5, WBP10

Expression Ubiquitous expression in whole brain E14.5 (RPKM 8.6), CNS E18 (RPKM 7.6) and 26 other tissuesSee more

Orthologs <u>human</u> all

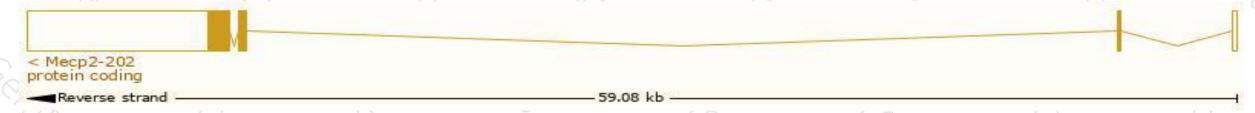
Transcript information (Ensembl)



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

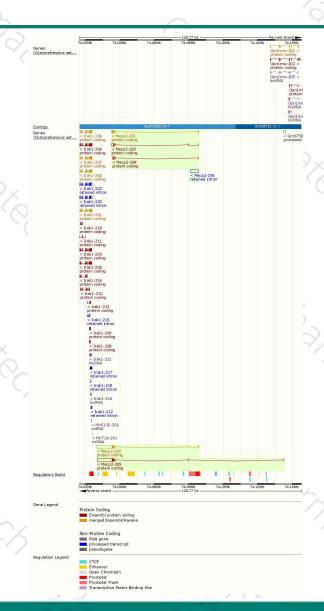
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Mecp2-202	ENSMUST00000100750.9	10538	<u>484aa</u>	Protein coding	CCDS41017	Q9Z2D6	TSL:1 GENCODE basic APPRIS ALT2
Mecp2-205	ENSMUST00000170481.8	10175	<u>484aa</u>	Protein coding	CCDS41017	Q9Z2D6	TSL:5 GENCODE basic APPRIS ALT2
Mecp2-201	ENSMUST00000033770.12	1739	<u>501aa</u>	Protein coding	CCDS41016	Q9Z2D6	TSL:1 GENCODE basic APPRIS P4
Mecp2-203	ENSMUST00000123362.7	1675	<u>172aa</u>	Protein coding	29	D3YY81	TSL:5 GENCODE basic
Mecp2-204	ENSMUST00000140399.2	1168	<u>189aa</u>	Protein coding	- E4	<u>D3Z7U4</u>	TSL:5 GENCODE basic
Mecp2-206	ENSMUST00000198358.1	4563	No protein	Retained intron	-8	-	TSL:NA

The strategy is based on the design of *Mecp2-202* transcript, The transcription is shown below



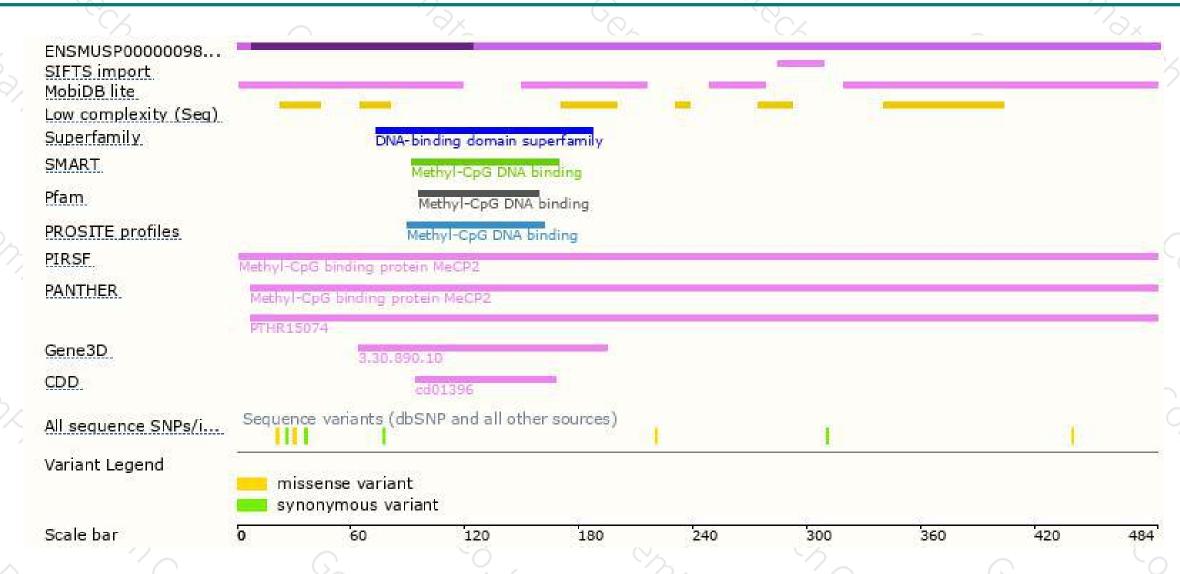
Genomic location distribution





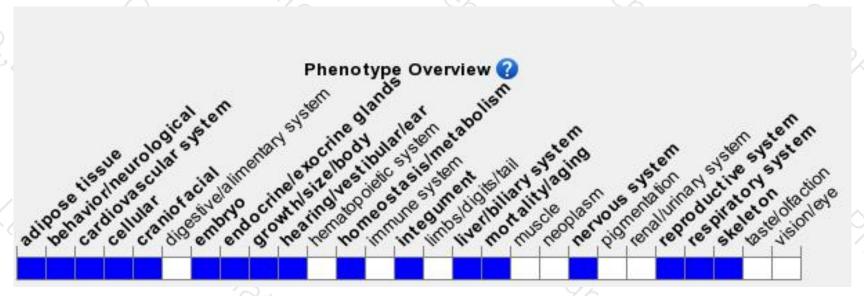
Protein domain





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, Female mice homozygous or male mice hemizygous for a null allele exhibit premature death, behavioral and neurological abnormalities, abnormal nervous system phenotypes, abnormal breathing, and abnormal hearing. Heterozygous mice exhibit similar behavioral and neurological abnormalities.



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





