

Etfbkmt Cas9-CKO Strategy

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Project Overview

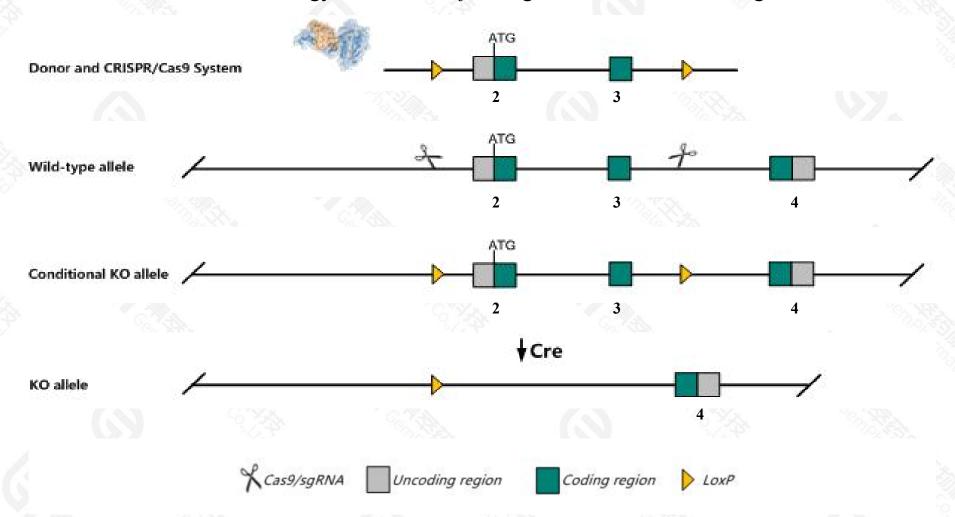


Project Name	Etfbkmt			
Project type	Cas9-CKO			
Strain background	C57BL/6JGpt			

Conditional Knockout strategy



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



Technical routes



- The *Etfbkmt* gene has 9 transcripts. According to the structure of *Etfbkmt* gene, exon2-exon3 of *Etfbkmt*-209(ENSMUST00000179873.8) transcript is recommended as the knockout region. The region contains start codon ATG. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Etfbkmt* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor 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



- > According to the existing MGI data,mice homozygous for a knock-out allele exhibit increased oxygen consumption rate and heat production under ketogenic conditions and enhanced cold tolerance during fasting.
- > The *Etfbkmt* gene is located on the Chr6. 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)



Etfbkmt electron transfer flavoprotein beta subunit lysine methyltransferase [Mus musculus (house mouse)]

Gene ID: 320204, updated on 20-Feb-2021

Summary



Official Symbol Etfbkmt provided by MGI

Official Full Name electron transfer flavoprotein beta subunit lysine methyltransferase provided by MGI

Primary source MGI:MGI:2443575

See related Ensembl:ENSMUSG00000039958

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 4833442J19Rik, Al256744, ETFB-KMT, Mettl2, Mettl20

Expression Ubiquitous expression in bladder adult (RPKM 13.6), heart adult (RPKM 5.1) and 24 other tissuesSee more

Orthologs <u>human</u> all

Transcript information (Ensembl)



The gene has 9 transcripts, all transcripts are shown below:

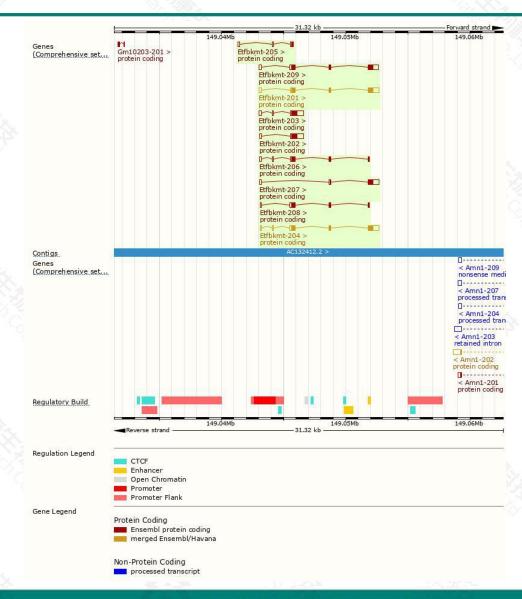
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Etfbkmt-209	ENSMUST00000179873.8	1576	255aa	Protein coding	CCDS20715		TSL:3 , GENCODE basic , APPRIS P1 ,
Etfbkmt-201	ENSMUST00000047531.16	1562	255aa	Protein coding	CCDS20715		TSL:1 , GENCODE basic , APPRIS P1 ,
Etfbkmt-204	ENSMUST00000111551.2	1558	255aa	Protein coding	CCDS20715		TSL:1 , GENCODE basic , APPRIS P1 ,
Etfbkmt-207	ENSMUST00000147934.4	1092	128aa	Protein coding	CCDS85191		TSL:1 , GENCODE basic ,
Etfbkmt-203	ENSMUST00000111548.8	1398	150aa	Protein coding	725		TSL:1 , GENCODE basic ,
Etfbkmt-202	ENSMUST00000111547.2	1250	<u>150aa</u>	Protein coding	(EU		TSL:1 , GENCODE basic ,
Etfbkmt-206	ENSMUST00000134306.8	848	<u>160aa</u>	Protein coding			CDS 3' incomplete , TSL:5 ,
Etfbkmt-208	ENSMUST00000166416.8	698	<u>165aa</u>	Protein coding	(20		CDS 3' incomplete , TSL:3 ,
Etfbkmt-205	ENSMUST00000126406.8	478	45aa	Protein coding	(30		CDS 3' incomplete , TSL:3 ,

The strategy is based on the design of *Etfbkmt-209* transcript, the transcription is shown below:



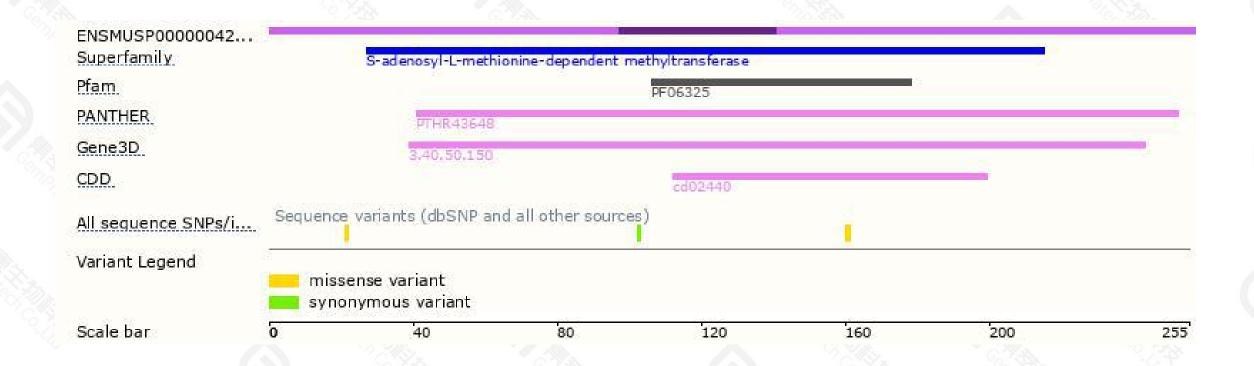
Genomic location distribution





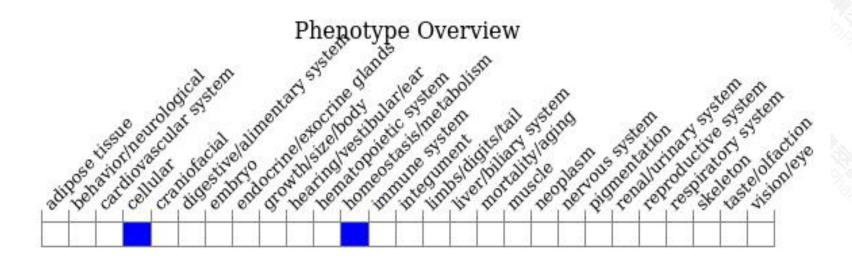
Protein domain





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 knock-out allele exhibit increased oxygen consumption rate and heat production under ketogenic conditions and enhanced cold tolerance during fasting.



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

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