

# *Clock* Cas9-KO Strategy

Designer:Lixin LYU

# Project Overview

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**Project Name**

***Clock***

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**Project type**

**Cas9-KO**

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**Strain background**

**C57BL/6JGpt**

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# Knockout strategy

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



- The *Clock* gene has 6 transcripts. According to the structure of *Clock* gene, exon7 of *Clock-205* (ENSMUST00000202651.3) transcript is recommended as the knockout region. The region contains 92bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Clock* gene. The brief process is as follows: CRISPR/Cas9 system

- According to the existing MGI data, Mice homozygous for a knock-out allele exhibit abnormal circadian phase. Mice homozygous for a spontaneous mutation exhibit abnormal circadian rhythm, reproduction, behavior, hair cycle, macronutrient absorption, and metabolism.
- The *Clock* gene is located on the Chr5. 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

# Gene information (NCBI)

## Clock circadian locomotor output cycles kaput [Mus musculus (house mouse)]

Gene ID: 12753, updated on 9-Apr-2019

### Summary



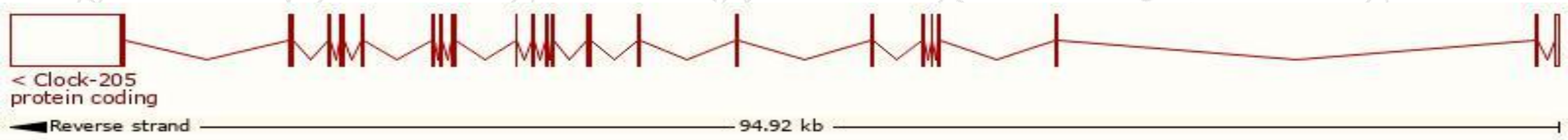
<b>Official Symbol</b>	Clock provided by <a href="#">MGI</a>
<b>Official Full Name</b>	circadian locomotor output cycles kaput provided by <a href="#">MGI</a>
<b>Primary source</b>	<a href="#">MGI:MGI:99698</a>
<b>See related</b>	<a href="#">Ensembl:ENSMUSG00000029238</a>
<b>Gene type</b>	protein coding
<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>	5330400M04Rik, KAT13D
<b>Summary</b>	The protein encoded by this gene plays a central role in the regulation of circadian rhythms. The protein encodes a transcription factor of the basic helix-loop-helix (bHLH) family and contains DNA binding histone acetyltransferase activity. The encoded protein forms a heterodimer with Arntl (Bmal1) that binds E-box enhancer elements upstream of Period (Per1, Per2, Per3) and Cryptochrome (Cry1, Cry2) genes and activates transcription of these genes. Per and Cry proteins heterodimerize and repress their own transcription by interacting in a feedback loop with Clock/Arntl complexes. Polymorphisms in this gene may be associated with behavioral changes, obesity, and metabolic syndrome. Two transcripts encoding the same protein have been found for this gene. [provided by RefSeq, Jan 2014]
<b>Expression</b>	Ubiquitous expression in subcutaneous fat pad adult (RPKM 6.4), cerebellum adult (RPKM 6.2) and 28 other tissues <a href="#">See more</a>
<b>Orthologs</b>	<a href="#">human</a> <a href="#">all</a>

# Transcript information (Ensembl)

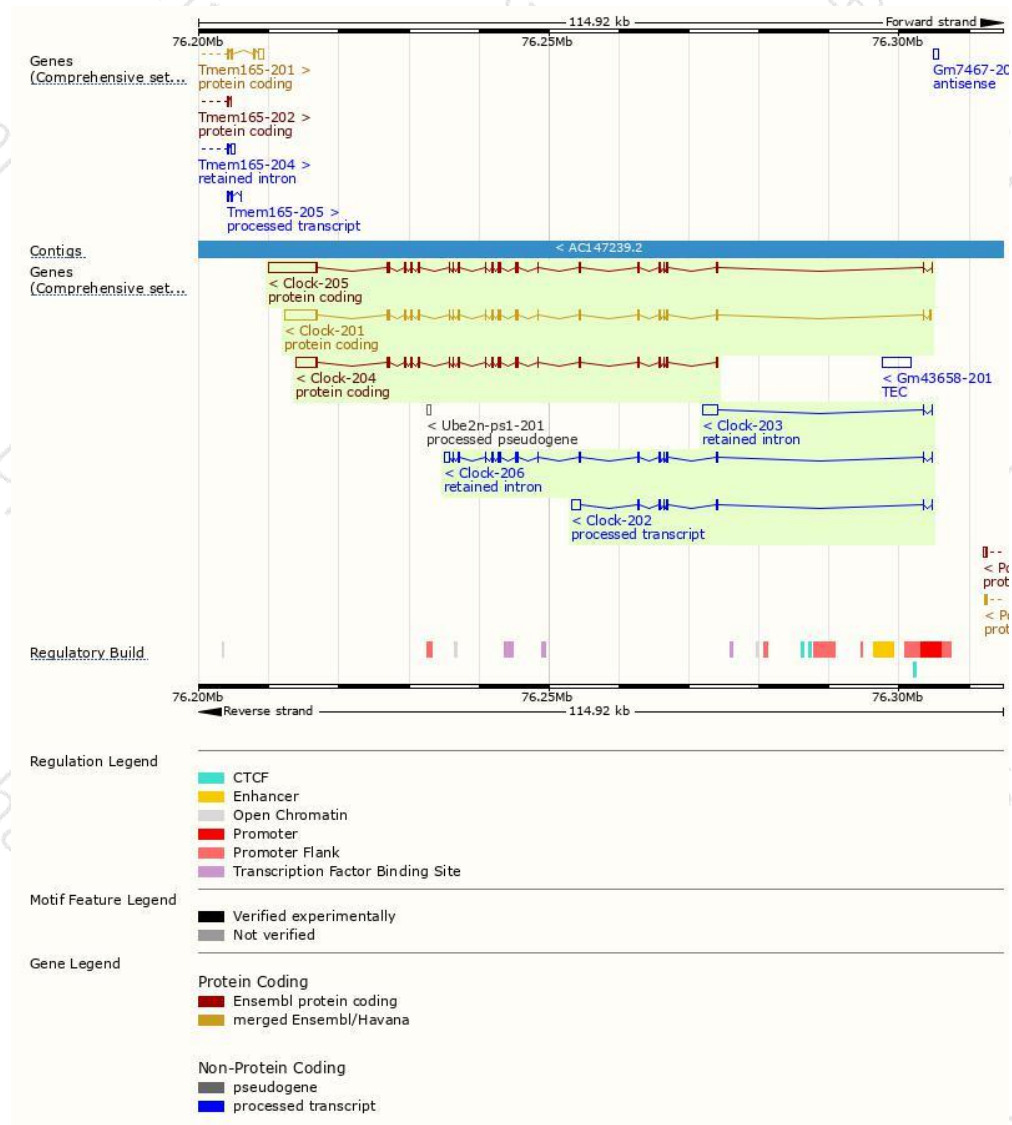
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Clock-205	<a href="#">ENSMUST00000202651.3</a>	9744	<a href="#">855aa</a>	Protein coding	<a href="#">CCDS19360</a>	<a href="#">Q08785</a>	TSL:2 GENCODE basic APPRIS P3
Clock-201	<a href="#">ENSMUST00000075159.4</a>	7478	<a href="#">855aa</a>	Protein coding	<a href="#">CCDS19360</a>	<a href="#">Q08785</a>	TSL:1 GENCODE basic APPRIS P3
Clock-204	<a href="#">ENSMUST00000202122.1</a>	5508	<a href="#">854aa</a>	Protein coding	<a href="#">CCDS84889</a>	<a href="#">A0A0J9YU61</a>	TSL:1 GENCODE basic APPRIS ALT2
Clock-202	<a href="#">ENSMUST00000200957.3</a>	1889	No protein	Processed transcript	-	-	TSL:2
Clock-206	<a href="#">ENSMUST00000202857.3</a>	2473	No protein	Retained intron	-	-	TSL:2
Clock-203	<a href="#">ENSMUST00000201052.1</a>	2287	No protein	Retained intron	-	-	TSL:1

The strategy is based on the design of *Clock-205* transcript,The transcription is shown below



# Genomic location distribution



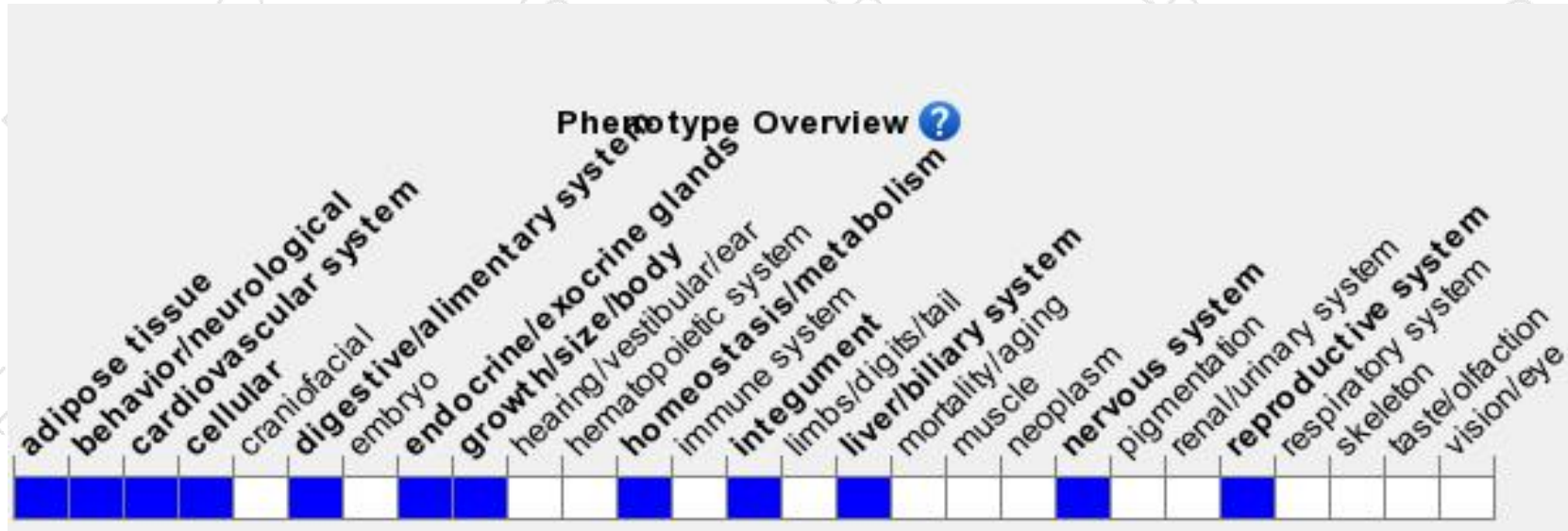
# Protein domain



集萃药康  
GemPharmatech



# 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 abnormal circadian phase. Mice homozygous for a spontaneous mutation exhibit abnormal circadian rhythm, reproduction, behavior, hair cycle, macronutrient absorption, and metabolism.

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

Tel: 400-9660890

