

# Crhr1 Cas9-KO Strategy

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# **Project Overview**



**Project Name** 

Crhr1

**Project type** 

Cas9-KO

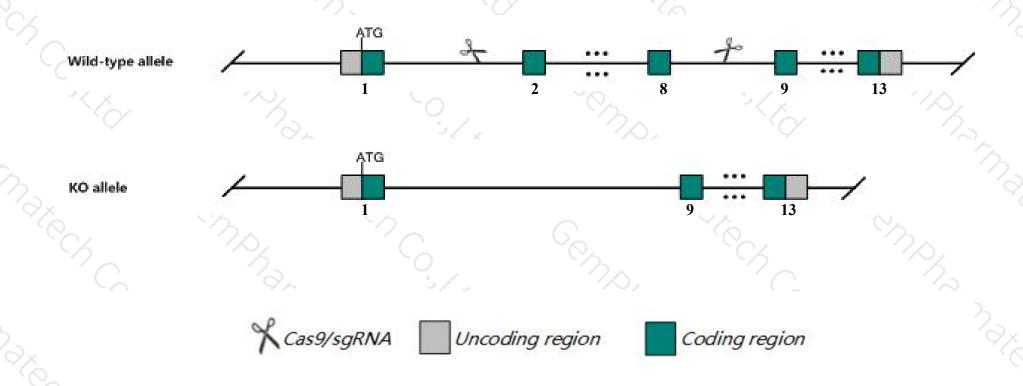
Strain background

C57BL/6JGpt

# **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Crhr1* gene. The schematic diagram is as follows:



### **Technical routes**



- ➤ The *Crhr1* gene has 1 transcript. According to the structure of *Crhr1* gene, exon2-exon8 of *Crhr1-201*(ENSMUST00000093925.4) transcript is recommended as the knockout region. The region contains 737bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Crhr1* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

### **Notice**



- ➤ According to the existing MGI data, Homozygotes for targeted null mutations exhibit agenesis of the adrenal zona fasciculata, low adrenocorticotropic hormone production, and reduced anxiety-related behaviors. Progeny of homozygote matings die from lung dysplasia within 48 hours of birth.
- > The *Crhr1* gene is located on the Chr11. 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)



#### Crhr1 corticotropin releasing hormone receptor 1 [Mus musculus (house mouse)]

Gene ID: 12921, updated on 19-Mar-2019

#### Summary

☆ ?

Official Symbol Crhr1 provided by MGI

Official Full Name corticotropin releasing hormone receptor 1 provided by MGI

Primary source MGI:MGI:88498

See related Ensembl: ENSMUSG00000018634

Gene type protein coding
RefSeq status REVIEWED

Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;

Muroidea; Muridae; Murinae; Mus; Mus

Also known as CRF1R, CRFR1, Crhr

Summary The protein encoded by this gene is a G-protein coupled receptor that binds corticotropin-releasing factor (CRH) and urocortin (UCN) with

high affinity. The encoded protein upregulates adenylate cyclase activity and intracellular cAMP levels. This protein is essential for the activation of signal transduction pathways that regulate diverse physiological processes including stress, reproduction, immune response,

and obesity. Several transcript variants encoding different isoforms have been found for this gene. [provided by RefSeq, Sep 2015]

Expression Biased expression in cerebellum adult (RPKM 7.7), frontal lobe adult (RPKM 4.7) and 6 other tissues See more

Orthologs <u>human</u> all

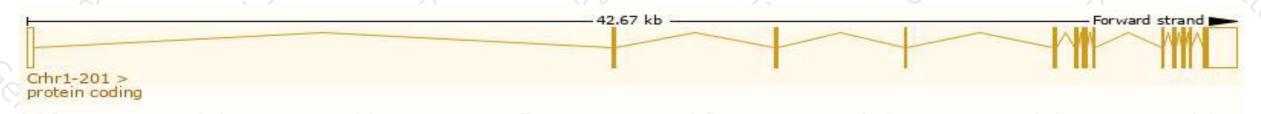
# Transcript information (Ensembl)



The gene has 1 transcript, and the transcript is shown below:

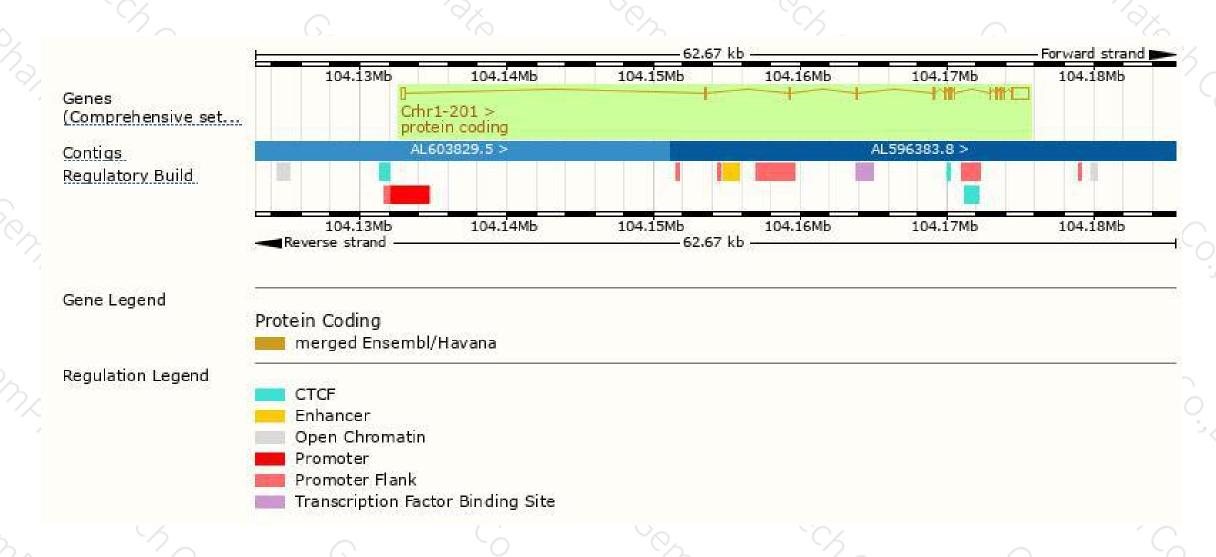
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags	
Crhr1-201	ENSMUST00000093925.4	2460	<u>415aa</u>	Protein coding	CCDS25526	P35347 Q3ZAT0	TSL:1 GENCODE basic APPRIS P1	L

The strategy is based on the design of Crhr1-201 transcript, The transcription is shown below



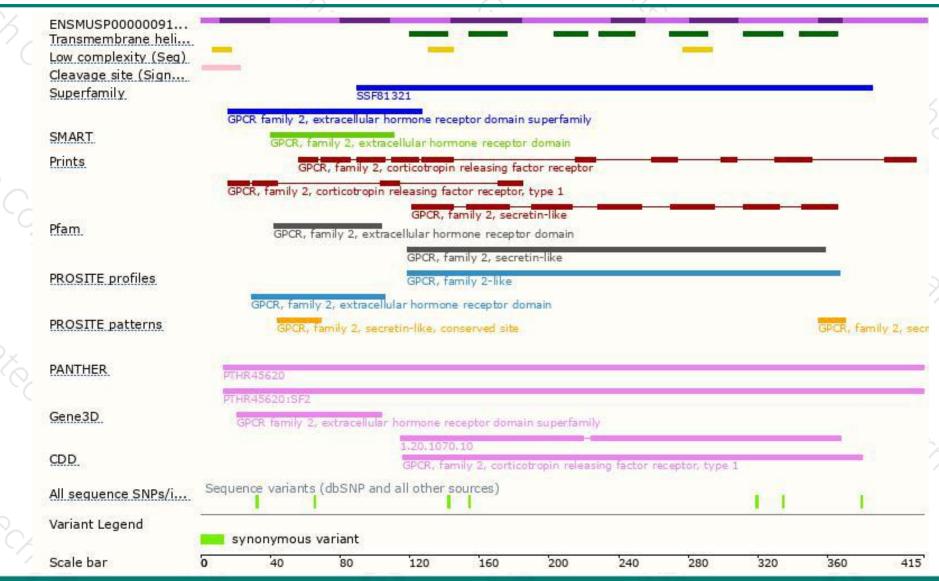
### Genomic location distribution





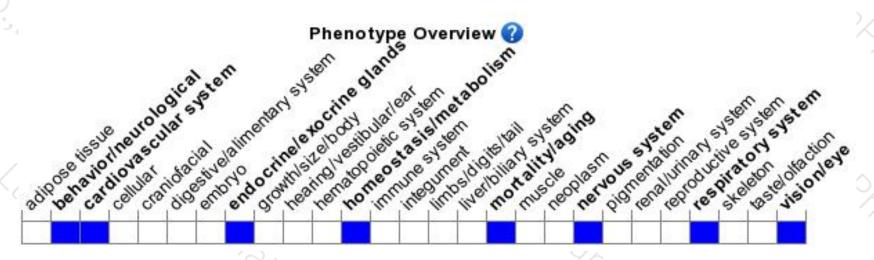
### Protein domain





## 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, Homozygotes for targeted null mutations exhibit agenesis of the adrenal zona fasciculata, low adrenocorticotropic hormone production, and reduced anxiety-related behaviors. Progeny of homozygote matings die from lung dysplasia within 48 hours of birth.



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

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