

# ***Crhr1* Cas9-CKO Strategy**

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# Project Overview

**Project Name**

***Crhr1***

**Project type**

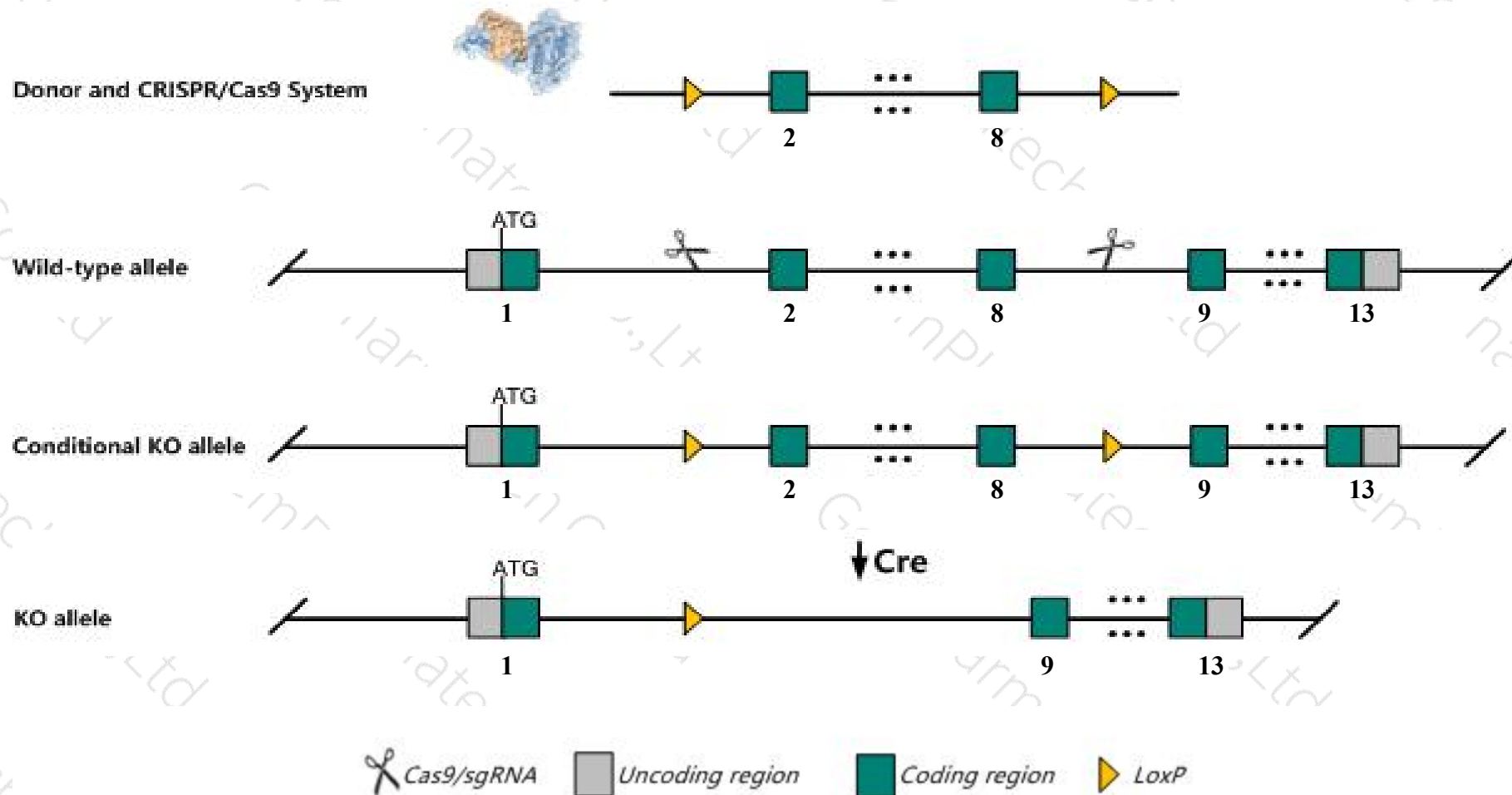
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

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



- 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, donor vector 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.

- According to the existing MGI data, Homozygotes for targeted null mutations exhibit agenesis of the adrenal zona fasciculata, low adrenocorticotrophic 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 loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological 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](#)

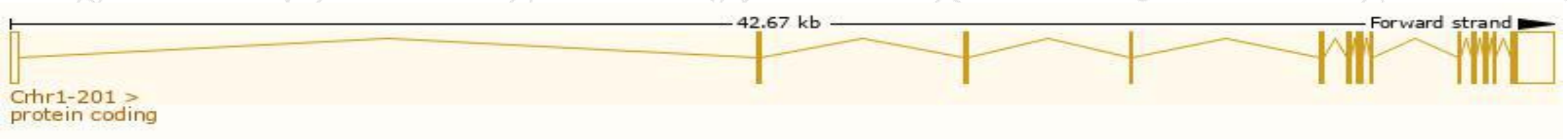
**Orthologs** [human](#) [all](#)

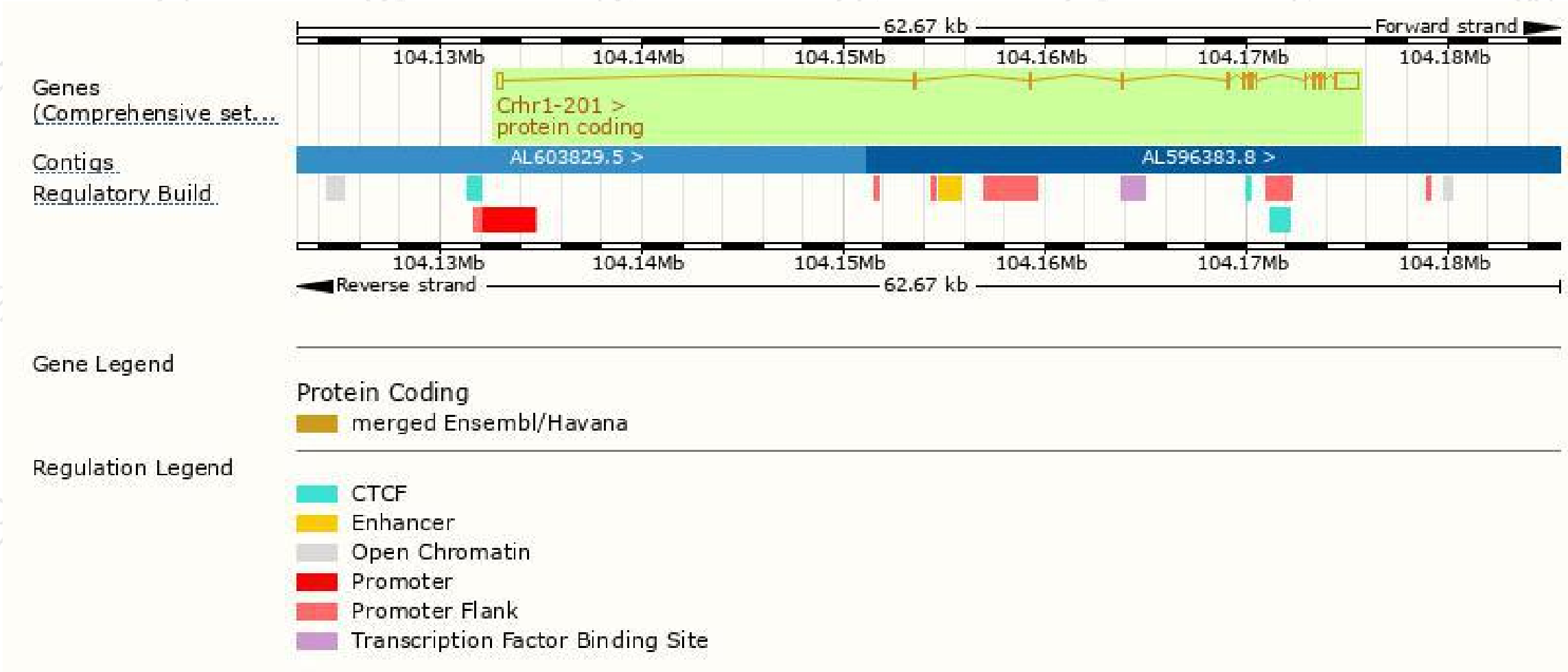
# Transcript information (Ensembl)

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

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

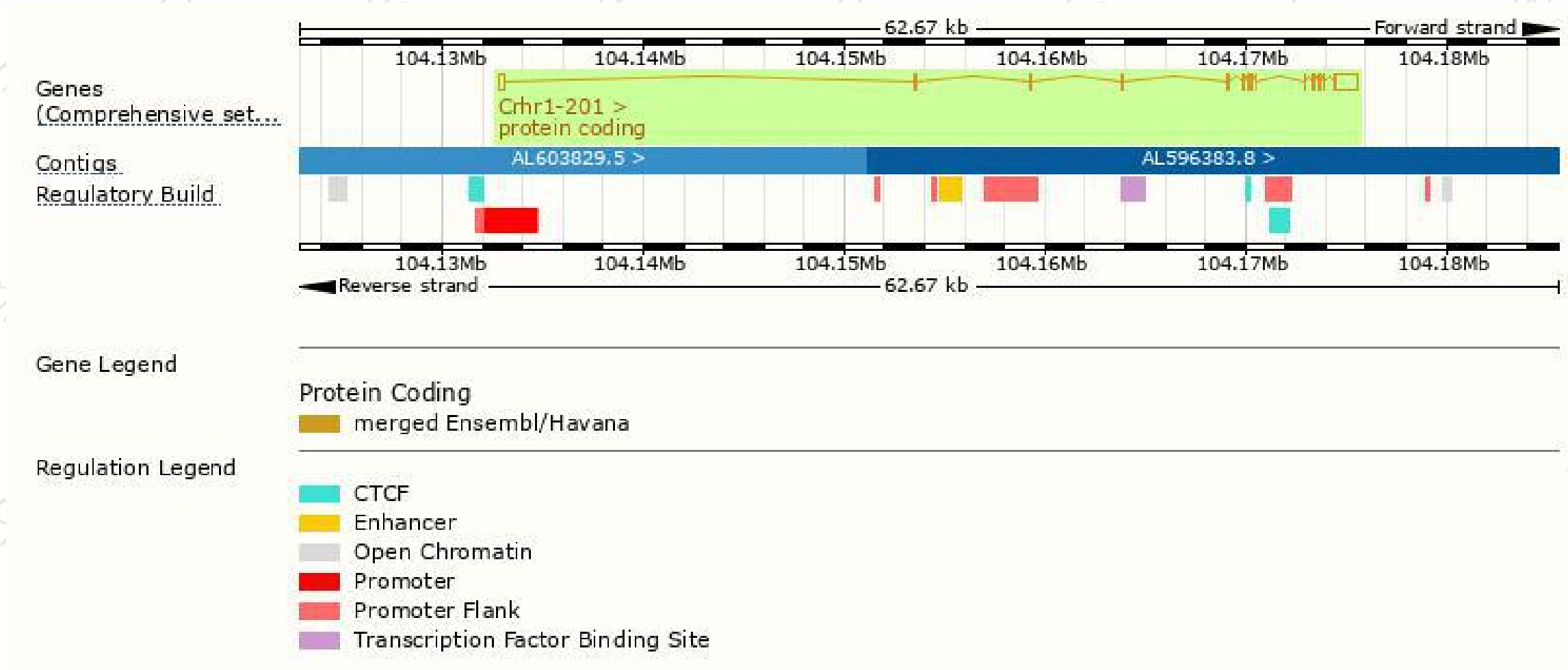
The strategy is based on the design of *Crhr1-201* transcript, The transcription is shown below



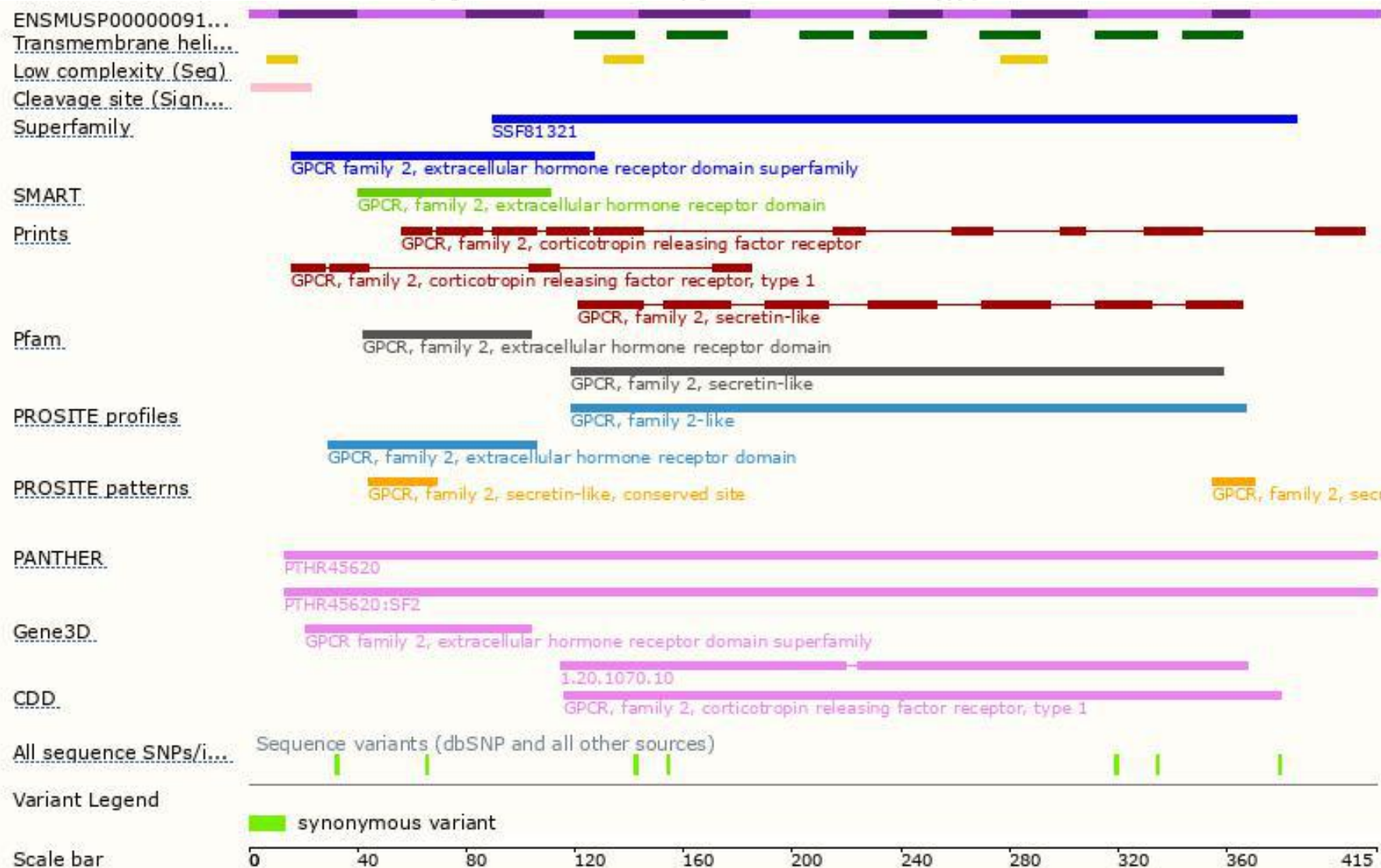




# Genomic location distribution



# 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 adrenocorticotrophic 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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