

Aanat Cas9-CKO Strategy

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Project Overview

Project Name

Aanat

Project type

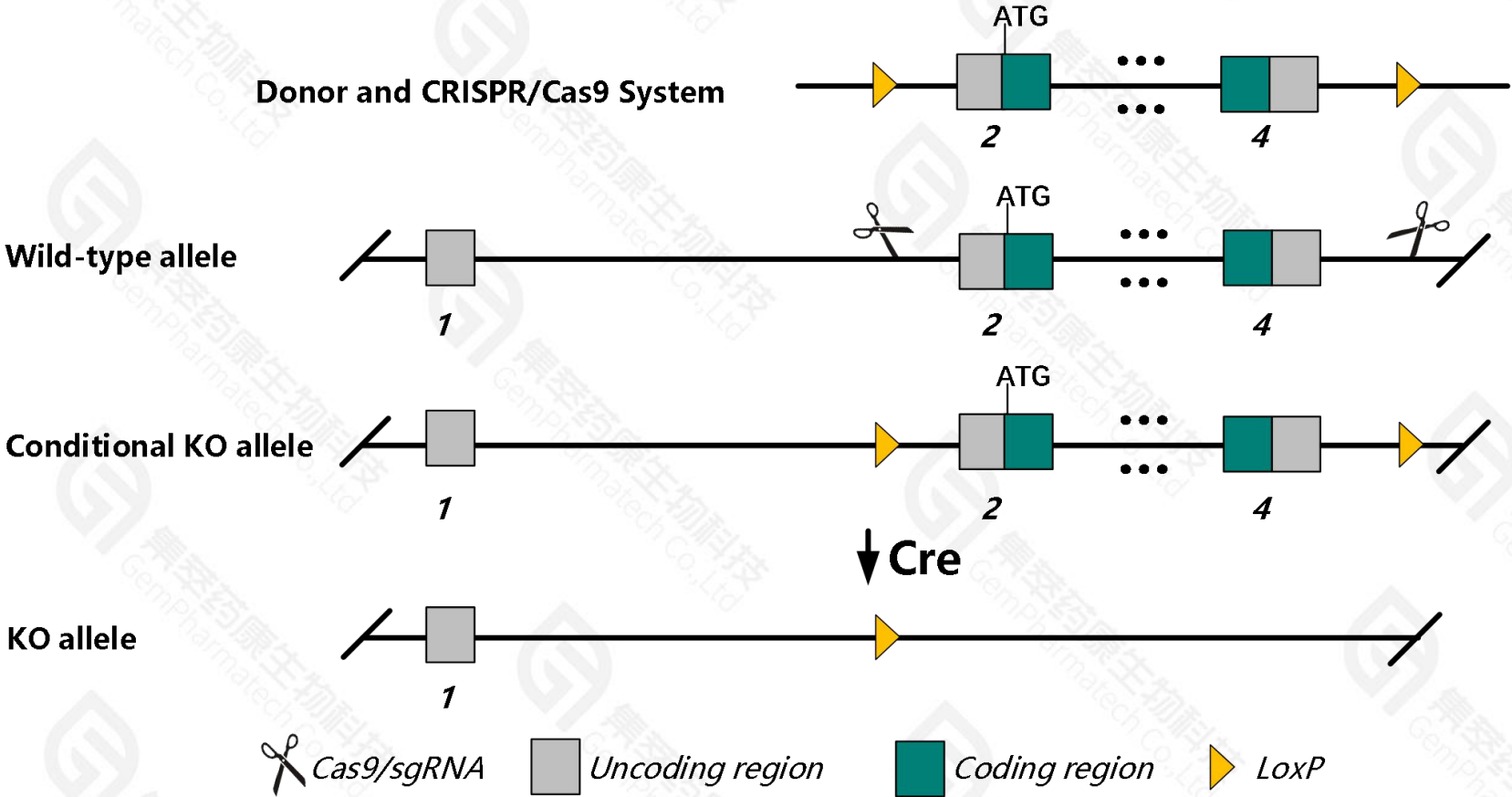
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



- The *Aanat* gene has 5 transcripts. According to the structure of *Aanat* gene, exon2-exon4 of *Aanat*-205(ENSMUST00000153476.7) 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 *Aanat* 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.

- According to the existing MGI data, mutations in this gene result in abnormal melatonin production.
- The floxed region is near to the N-terminal of C-terminal of *Rhbdf2* gene, this strategy extremely likely influence the regulatory function of the C-terminal of *Rhbdf2* gene.
- The *Aanat* 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)

Aanat arylalkylamine N-acetyltransferase [Mus musculus (house mouse)]

Gene ID: 11298, updated on 13-Mar-2020

Summary



Official Symbol Aanat provided by [MGI](#)

Official Full Name arylalkylamine N-acetyltransferase provided by [MGI](#)

Primary source [MGI:MGI:1328365](#)

See related [Ensembl:ENSMUSG00000020804](#)

Gene type protein coding

RefSeq status REVIEWED

Organism [Mus musculus](#)

Also known as AA-NAT, Nat-2, Nat4, Snat

Summary The protein encoded by this gene belongs to the acetyltransferase superfamily. It is the penultimate enzyme in melatonin synthesis and controls the night/day rhythm in melatonin production in the vertebrate pineal gland. Melatonin is essential for the function of the circadian clock that influences activity and sleep. This enzyme is regulated by cAMP-dependent phosphorylation that promotes its interaction with 14-3-3 proteins and thus protects the enzyme against proteasomal degradation. This gene may contribute to numerous genetic diseases such as delayed sleep phase syndrome. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Jan 2010]

Expression Low expression observed in reference dataset [See more](#)

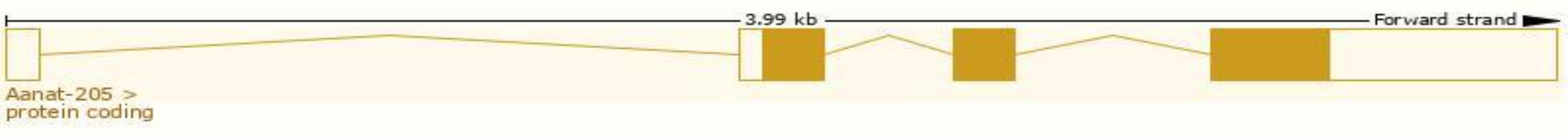
Orthologs [human](#) [all](#)

Transcript information (Ensembl)

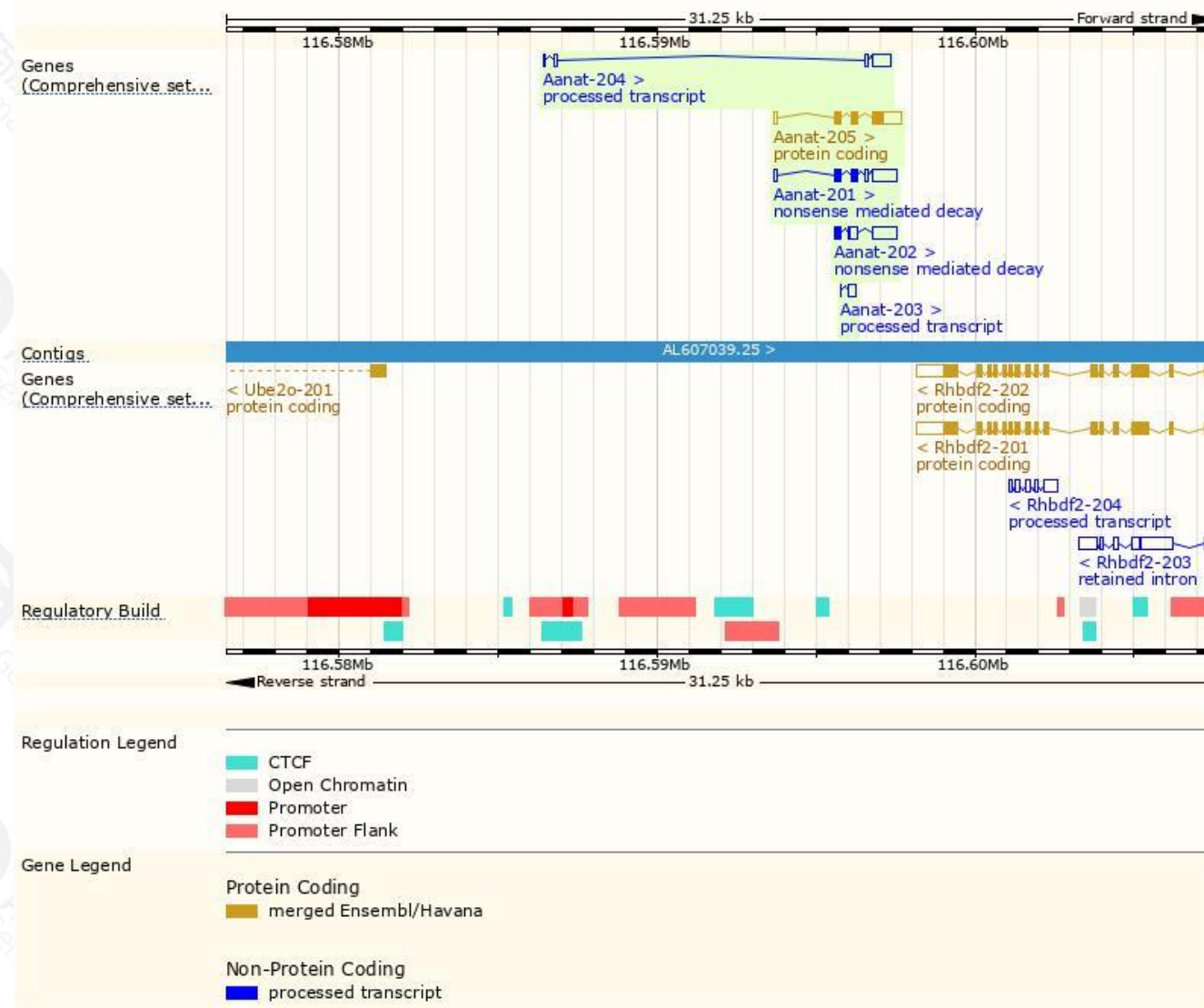
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Aanat-205	ENSMUST00000153476.7	1353	205aa	Protein coding	CCDS25671	Q88816 Q14A64	TSL:1 GENCODE basic APPRIS P1
Aanat-201	ENSMUST00000021160.9	1320	105aa	Nonsense mediated decay	-	Q9QUP1	TSL:1
Aanat-202	ENSMUST00000123507.1	1219	59aa	Nonsense mediated decay	-	F2Z3V1	TSL:1
Aanat-204	ENSMUST00000142978.1	766	No protein	Processed transcript	-	-	TSL:3
Aanat-203	ENSMUST00000132601.1	252	No protein	Processed transcript	-	-	TSL:1

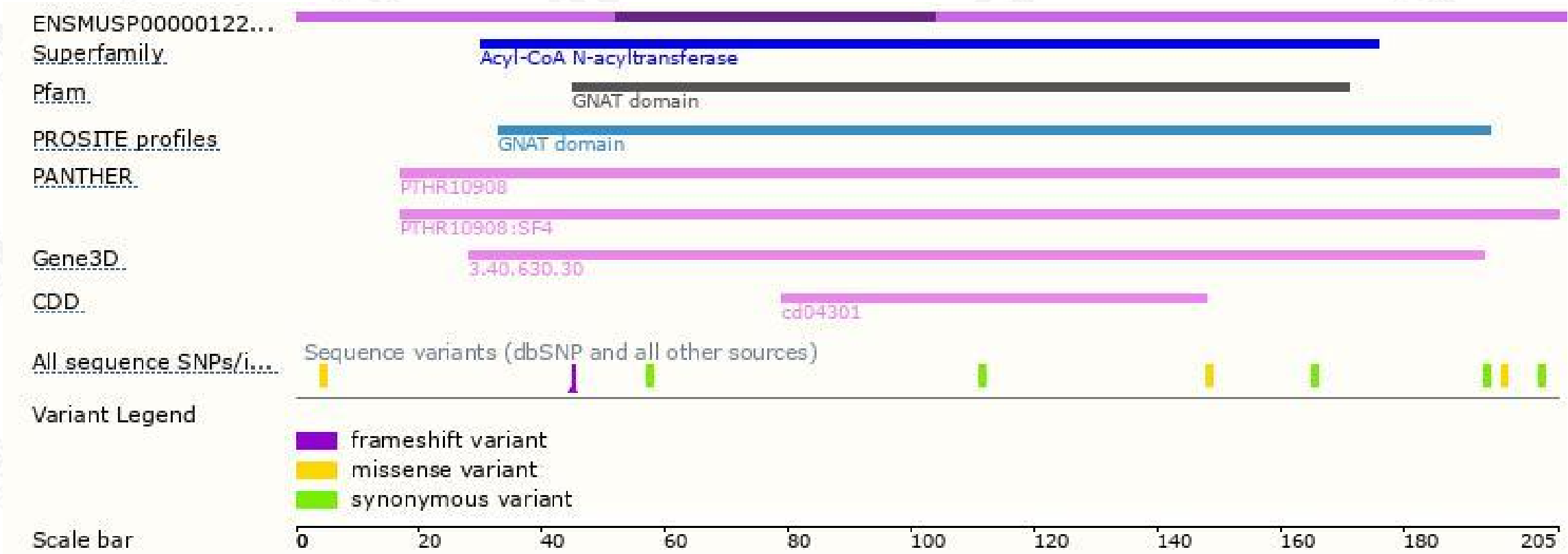
The strategy is based on the design of *Aanat-205* transcript,the transcription is shown below:



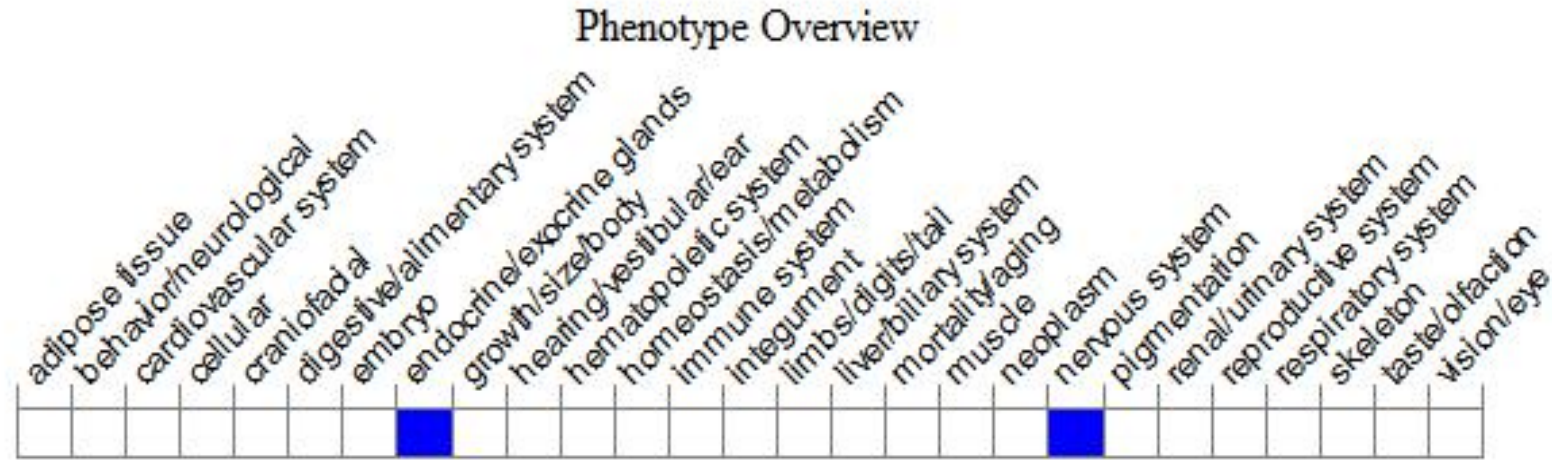
Genomic location distribution



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, mutations in this gene result in abnormal melatonin production.

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