

# Stat5b Cas9-CKO Strategy

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Reviewer: Lingyan Wu

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# **Project Overview**



**Project Name** 

Stat5b

**Project type** 

Cas9-CKO

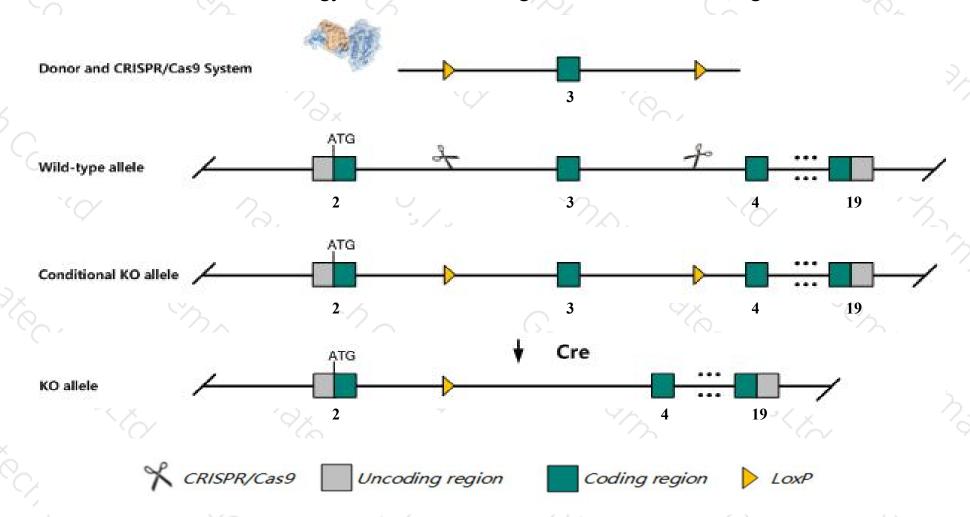
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Stat5b* gene. The schematic diagram is as follows:



### **Technical routes**



- ➤ The *Stat5b* gene has 3 transcripts. According to the structure of *Stat5b* gene, exon3 of *Stat5b*
  201(ENSMUST00000004143.2) transcript is recommended as the knockout region. The region contains 157bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Stat5b* 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.

### **Notice**



- > According to the existing MGI data, mice homozygous for disruptions in this are reduced in size and mammary glands secrete reduced levels of some milk proteins during lactation.
- The *Stat5b* 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)



#### Stat5b signal transducer and activator of transcription 5B [Mus musculus (house mouse)]

Gene ID: 20851, updated on 13-Mar-2020

#### Summary

☆ ?

Official Symbol Stat5b provided by MGI

Official Full Name signal transducer and activator of transcription 5B provided by MGI

Primary source MGI:MGI:103035

See related Ensembl: ENSMUSG00000020919

RefSeq status VALIDATED
Organism Mus musculus

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

Muroidea; Muridae; Murinae; Mus; Mus

Expression Ubiquitous expression in thymus adult (RPKM 41.1), ovary adult (RPKM 18.1) and 27 other tissues See more

Orthologs <u>human</u> all

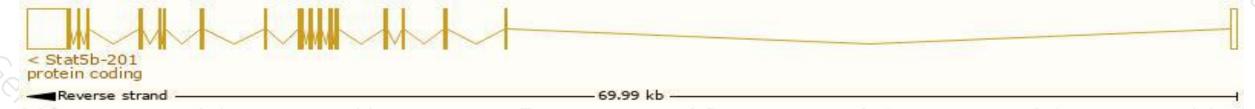
# Transcript information (Ensembl)



The gene has 3 transcripts, all transcripts are shown below:

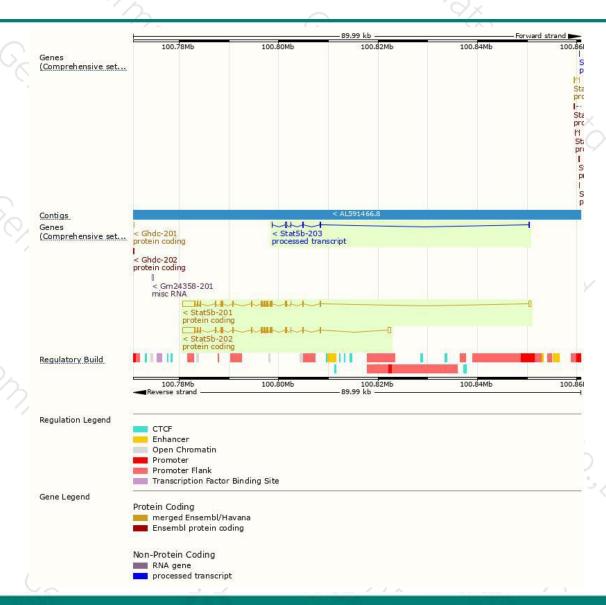
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Stat5b-202	ENSMUST00000107358.8	5213	786aa	Protein coding	CCDS25438	P42232	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Stat5b-201	ENSMUST00000004143.2	5062	786aa	Protein coding	CCDS25438	P42232	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Stat5b-203	ENSMUST00000126266.1	727	No protein	Processed transcript	21	(2)	TSL:3

The strategy is based on the design of *Stat5b-201* transcript, the transcription is shown below:



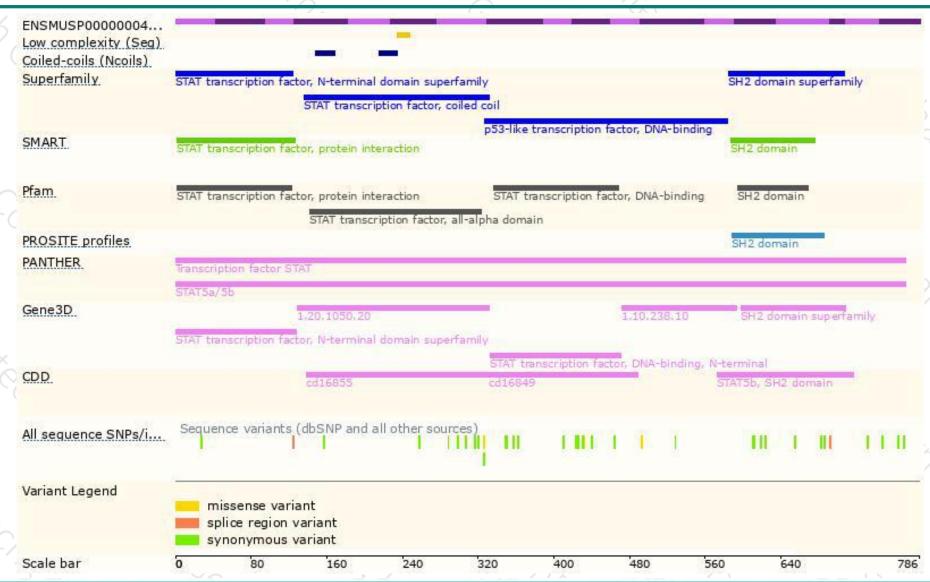
### Genomic location distribution





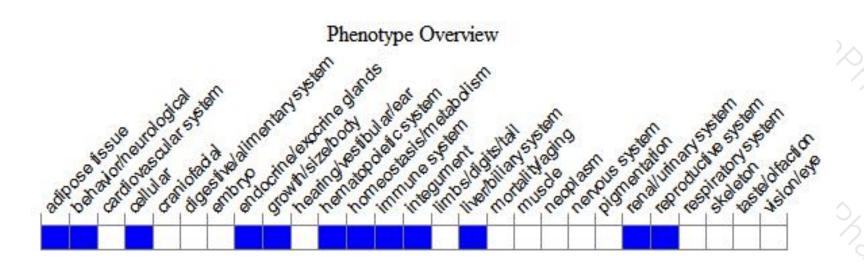
### Protein domain





# 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,mice homozygous for disruptions in this are reduced in size and mammary glands secrete reduced levels of some milk proteins during lactation.



If you have any questions, you are welcome to inquire. Tel: 400-9660890





