

# Stat1 Cas9-KO Strategy

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Design Date: 2019-9-19

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# **Project Overview**



**Project Name** 

Stat1

**Project type** 

Cas9-KO

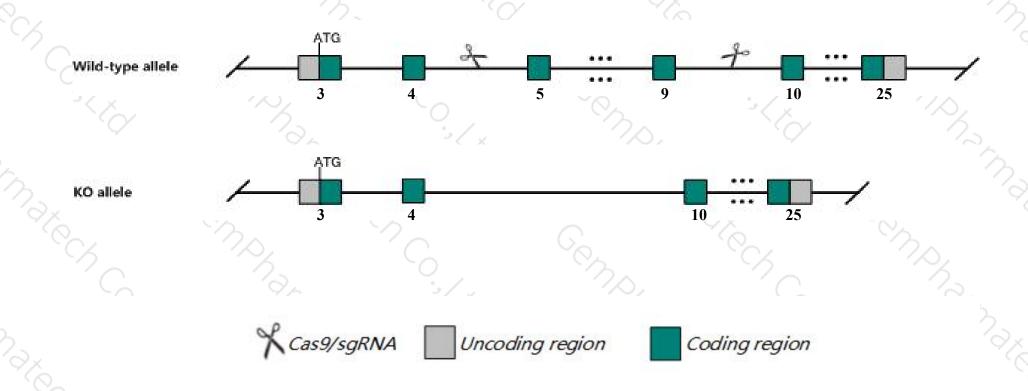
Strain background

**C57BL/6J** 

# **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Stat1* gene. The schematic diagram is as follows:



### **Technical routes**



- ➤ The *Stat1* gene has 12 transcripts. According to the structure of *Stat1* gene, exon5-exon9 of *Stat1-203*(ENSMUST00000186057.6) transcript is recommended as the knockout region. The region contains 512bp coding sequence.

  Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Stat1* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

### **Notice**



- > According to the existing MGI data, homozygotes for targeted null mutations are largely unresponsive to interferon, fail to thrive, are susceptible to viral diseases and cutaneous leishmaniasis, and show excess osteoclastogenesis leading to increased bone mass.
- ➤ Homozygous mice are viable and fertile when maintained under specific pathogen-free (SPF) conditions.

  In conventional health status vivaria (non-SPF), homozygotes tend to be runted and die within 48 hours of weaning due to hepatic necrosis due to viral infection.
- ➤ The *Stat1* gene is located on the Chr1. 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)



#### Stat1 signal transducer and activator of transcription 1 [Mus musculus (house mouse)]

Gene ID: 20846, updated on 9-Apr-2019

#### Summary

☆ ?

Official Symbol Stat1 provided by MGI

Official Full Name signal transducer and activator of transcription 1 provided by MGI

Primary source MGI:MGI:103063

See related Ensembl:ENSMUSG00000026104

Gene type protein coding
RefSeq status VALIDATED
Organism Mus musculus

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

Muroidea; Muridae; Murinae; Mus; Mus

Also known as 2010005J02Rik, AA408197

Expression Ubiquitous expression in thymus adult (RPKM 23.3), spleen adult (RPKM 19.5) and 28 other tissuesSee more

Orthologs <u>human</u> all

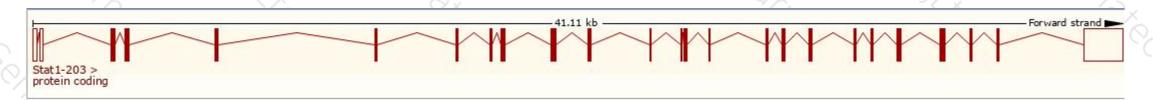
# Transcript information (Ensembl)



The gene has 12 transcripts, all transcripts are shown below:

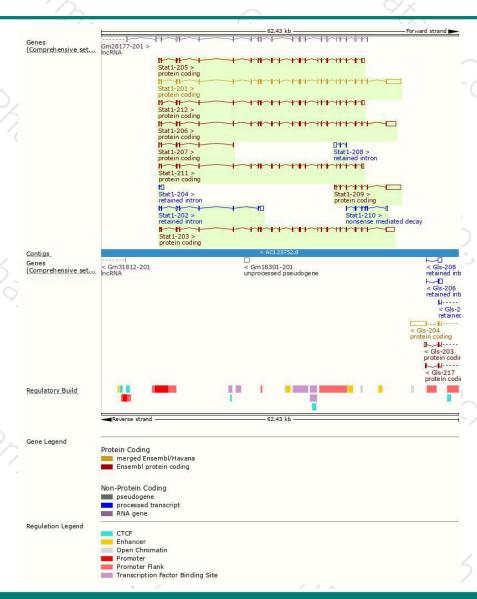
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Stat1-201	ENSMUST00000070968.13	5151	749aa	Protein coding	CCDS56628	Q8C3V4	TSL:1 GENCODE basic APPRIS P1
Stat1-206	ENSMUST00000186857.6	4241	749aa	Protein coding	CCDS56628	Q8C3V4	TSL:1 GENCODE basic APPRIS P1
Stat1-203	ENSMUST00000186057.6	3998	<u>755aa</u>	Protein coding	CCDS78581	A0A087WSP5	TSL:1 GENCODE basic
Stat1-209	ENSMUST00000189244.6	3298	243aa	Protein coding	-	Q8CFQ1	CDS 5' incomplete TSL:1
Stat1-205	ENSMUST00000186574.6	2834	712aa	Protein coding		Q99K94	TSL:1 GENCODE basic
Stat1-212	ENSMUST00000191435.6	2803	712aa	Protein coding	-	Q99K94	TSL:5 GENCODE basic
Stat1-211	ENSMUST00000189347.6	2588	712aa	Protein coding		Q99K94	TSL:5 GENCODE basic
Stat1-207	ENSMUST00000188681.6	715	<u>154aa</u>	Protein coding	2	A0A087WSQ5	CDS 3' incomplete TSL:2
Stat1-210	ENSMUST00000189336.1	886	<u>131aa</u>	Nonsense mediated decay	a	A0A087WRI1	CDS 5' incomplete TSL:3
Stat1-202	ENSMUST00000185743.1	1344	No protein	Retained intron	-	<del>.</del>	TSL:1
Stat1-208	ENSMUST00000189075.1	827	No protein	Retained intron	9	49	TSL:2
Stat1-204	ENSMUST00000186186.1	548	No protein	Retained intron	-	29	TSL:2

The strategy is based on the design of Stat1-203 transcript, The transcription is shown below



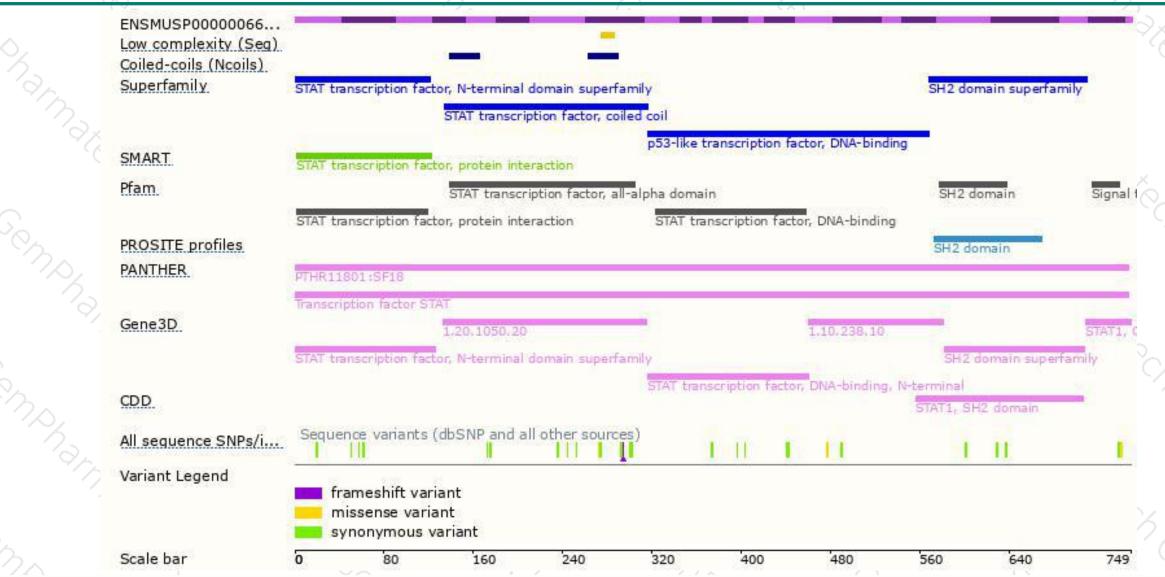
### Genomic location distribution





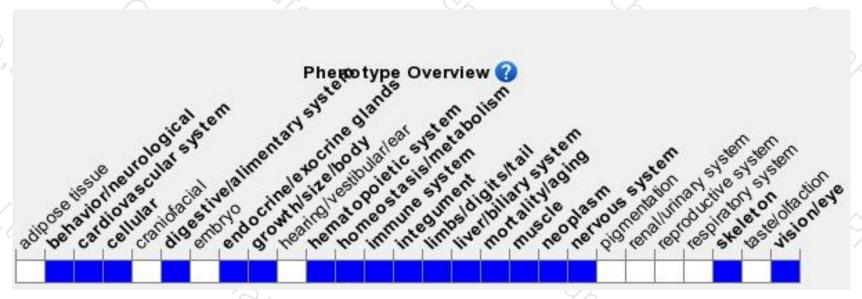
### Protein domain





### 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 are largely unresponsive to interferon, fail to thrive, are susceptible to viral diseases and cutaneous leishmaniasis, and show excess osteoclastogenesis leading to increased bone mass.



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

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