B2m Cas9-KO *Strategy*

Designer: **Lixin Lv**

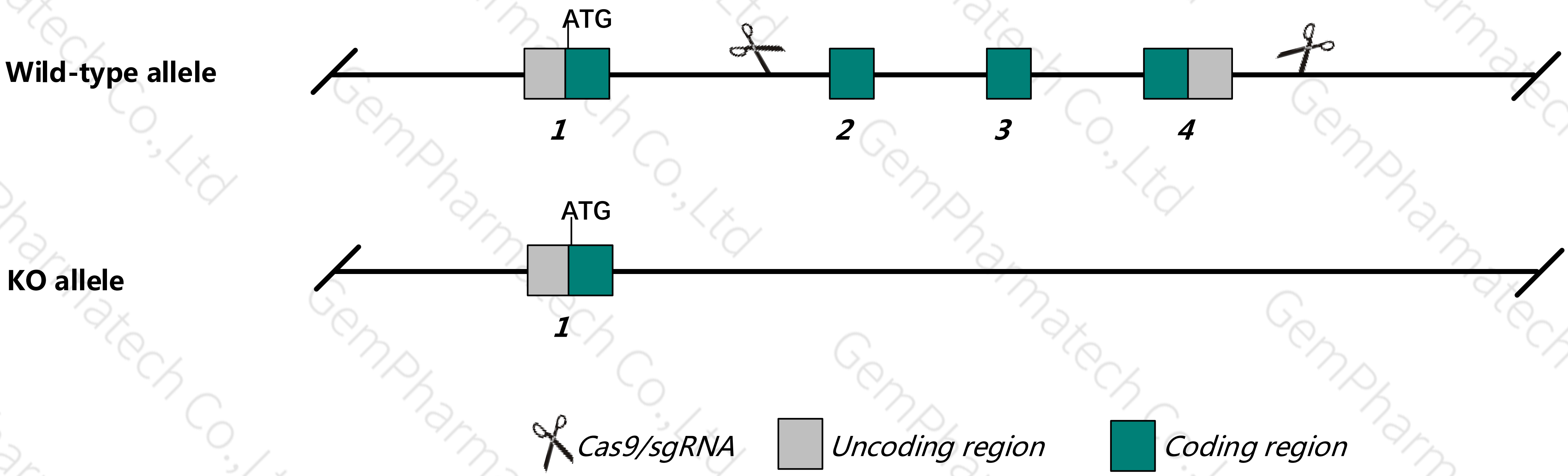
Design Date: **2018/11/13**

Project Overview

Project Name	***B2m***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *B2m* gene. The schematic diagram is as follows:



Technical routes

- The *B2m* gene has 1 transcripts. According to the structure of *B2m* gene, exon2~exon4 of *B2m*-201 (ENSMUSG00000060802) transcript is recommended as the knockout region.The region contains most 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 *B2m* 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 lacking B2m appear normal, but have no detectable MHC class I antigen on their cells and are deficient in CD4- CD8+ T cells which mediate cytotoxic T cell function. Mutant mice are also subject to systemic iron loading.
- The *B2m* gene is located on the Chr2. 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）

集萃药康
GemPharmatech

B2m beta-2 microglobulin [*Mus musculus* (house mouse)]

Gene ID: 12010, updated on 4-Jul-2019

Summary

Official Symbol	B2m provided by MGI
Official Full Name	beta-2 microglobulin provided by MGI
Primary source	MGI:MGI:88127
See related	Ensembl:ENSMUSG00000060802
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	Ly-m11; beta2m; beta2-m
Expression	Broad expression in liver adult (RPKM 838.6), mammary gland adult (RPKM 675.3) and 20 other tissues See more
Orthologs	human all

Genomic context

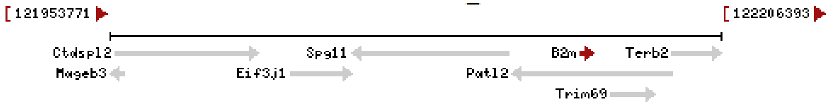
Location: 2 E5; 2 60.55 cM

See B2m in Genome Data Viewer

Exon count: 4

Annotation release	Status	Assembly	Chr	Location
106	current	GRCm38.p4 (GCF_000001635.24)	2	NC_000068.7 (122147687..122153082)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	2	NC_000068.6 (121973423..121978818)

Chromosome 2 - NC_000068.7



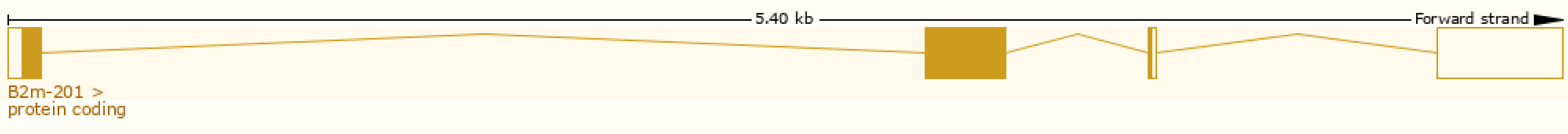
Transcript information（Ensembl）

集萃药康 GemPharmatech

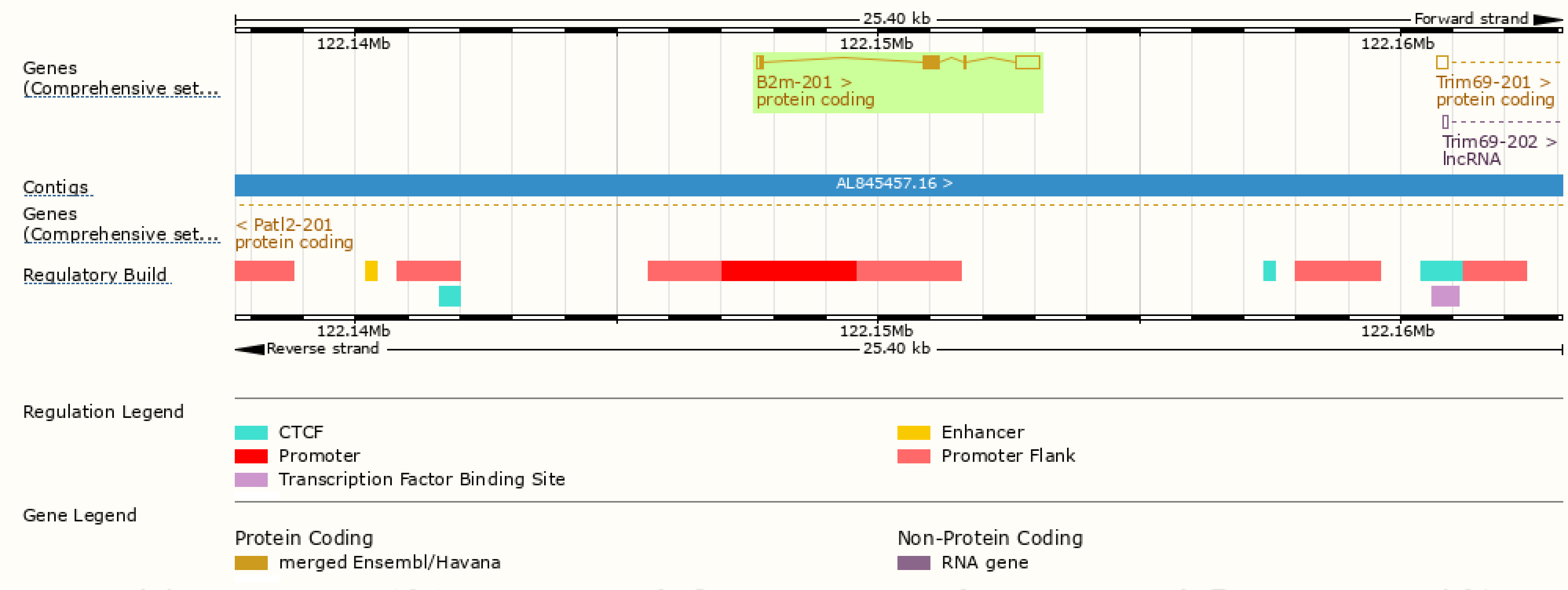
The gene has 1 transcript, and the transcript is shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
B2m-201	ENSMUST00000102476.4	860	119aa	Protein coding	CCDS16654	P01887	TSL:1 GENCODE basic APPRIS P1

The strategy is based on the design of *B2m*-201 transcript,The transcription is shown below

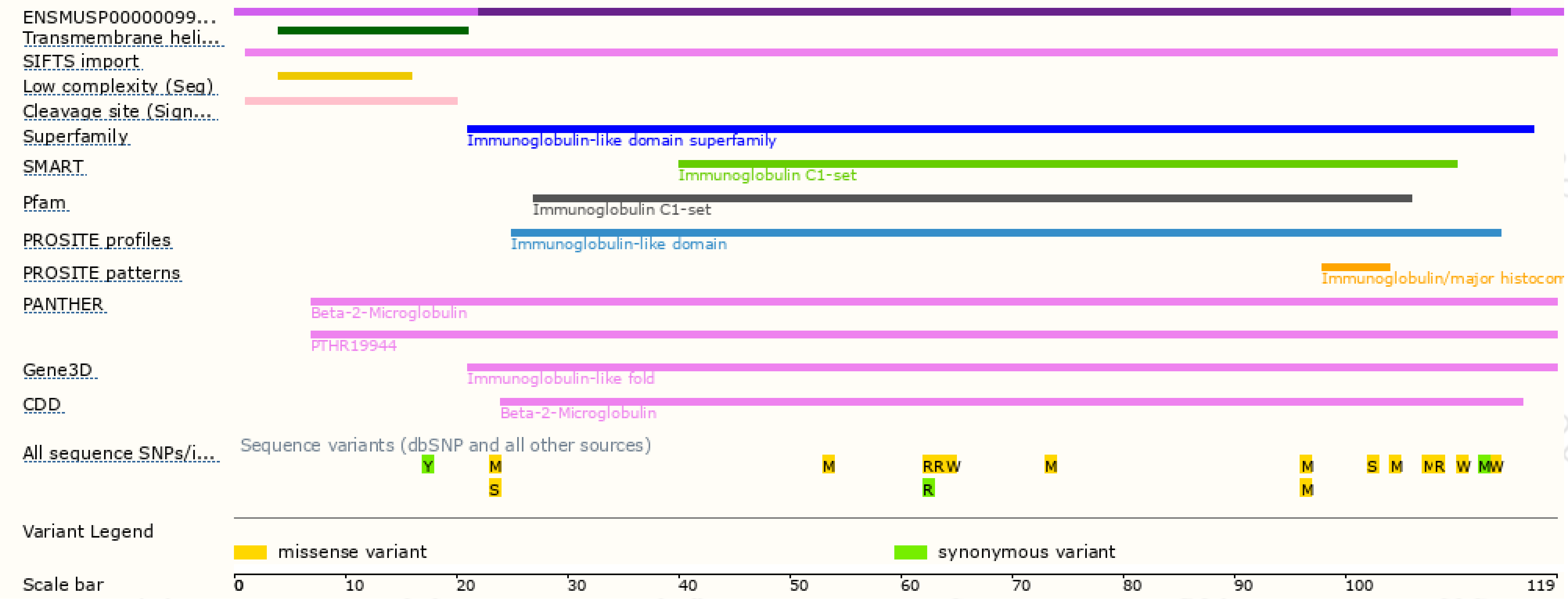


Genomic location distribution



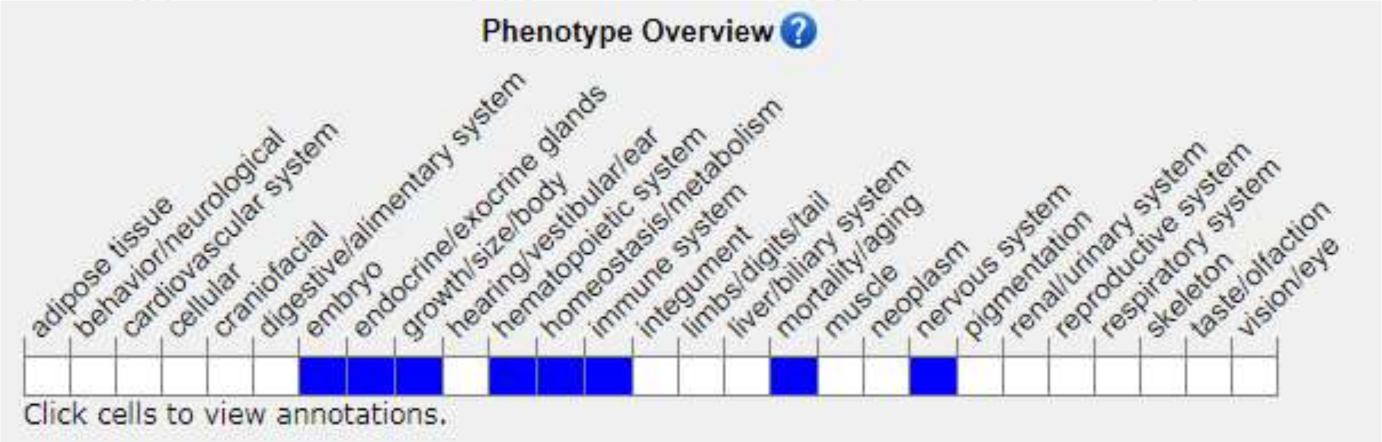
Protein domain

集萃药康 GemPharmatech



Mouse phenotype description(MGI)

集萃药康 GemPharmatech



Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/) .

Homozygotes lacking B2m appear normal, but have no detectable MHC class I antigen on their cells and are deficient in CD4- CD8+ T cells which mediate cytotoxic T cell function. Mutant mice are also subject to systemic iron loading.

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

Tel: 025-5864 1534

集萃药康生物科技
GemPharmatech Co.,Ltd