Rassf6 Cas9-KO Strategy

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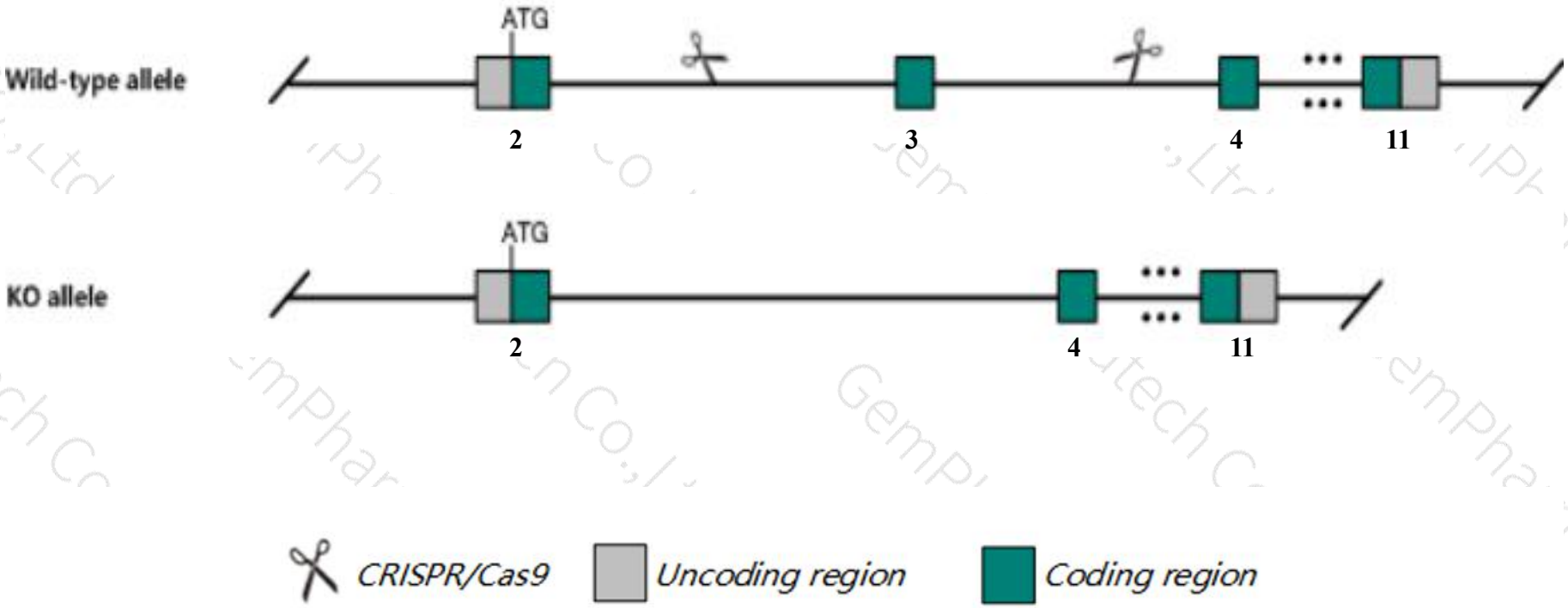
Project Overview

Project Name	***Rassf6***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rassf6* gene. The schematic diagram is as follows:



Technical routes

➢ The *Rassf6* gene has 7 transcripts. According to the structure of *Rassf6* gene, exon3 of *Rassf6-201*(ENSMUST00000031317.9) transcript is recommended as the knockout region.The region contains 79bp coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Rassf6* gene. The brief process is as follows: CRISPR/Cas9 system 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

➢ The *Rassf6* gene is located on the Chr5. 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.

➢ Transcript *Rassf6*-207 may not be affected.

➢ The effect on transcript *Rassf6*-203 is unknown.

➢ 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）

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Rassf6 Ras association (RalGDS/AF-6) domain family member 6 [Mus musculus (house mouse)]

Gene ID: 73246, updated on 13-Mar-2020

Summary

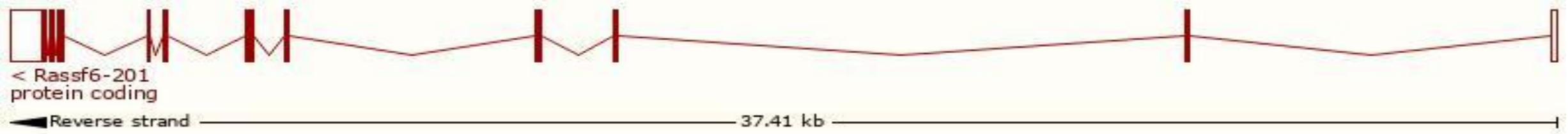
Official Symbol	Rassf6 provided by MGI
Official Full Name	Ras association (RalGDS/AF-6) domain family member 6 provided byMGI
Primary source	MGI:MGI:1920496
See related	Ensembl:ENSMUSG00000029370
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1600016B17Rik
Expression	Broad expression in placenta adult (RPKM 6.2), colon adult (RPKM 3.6) and 15 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

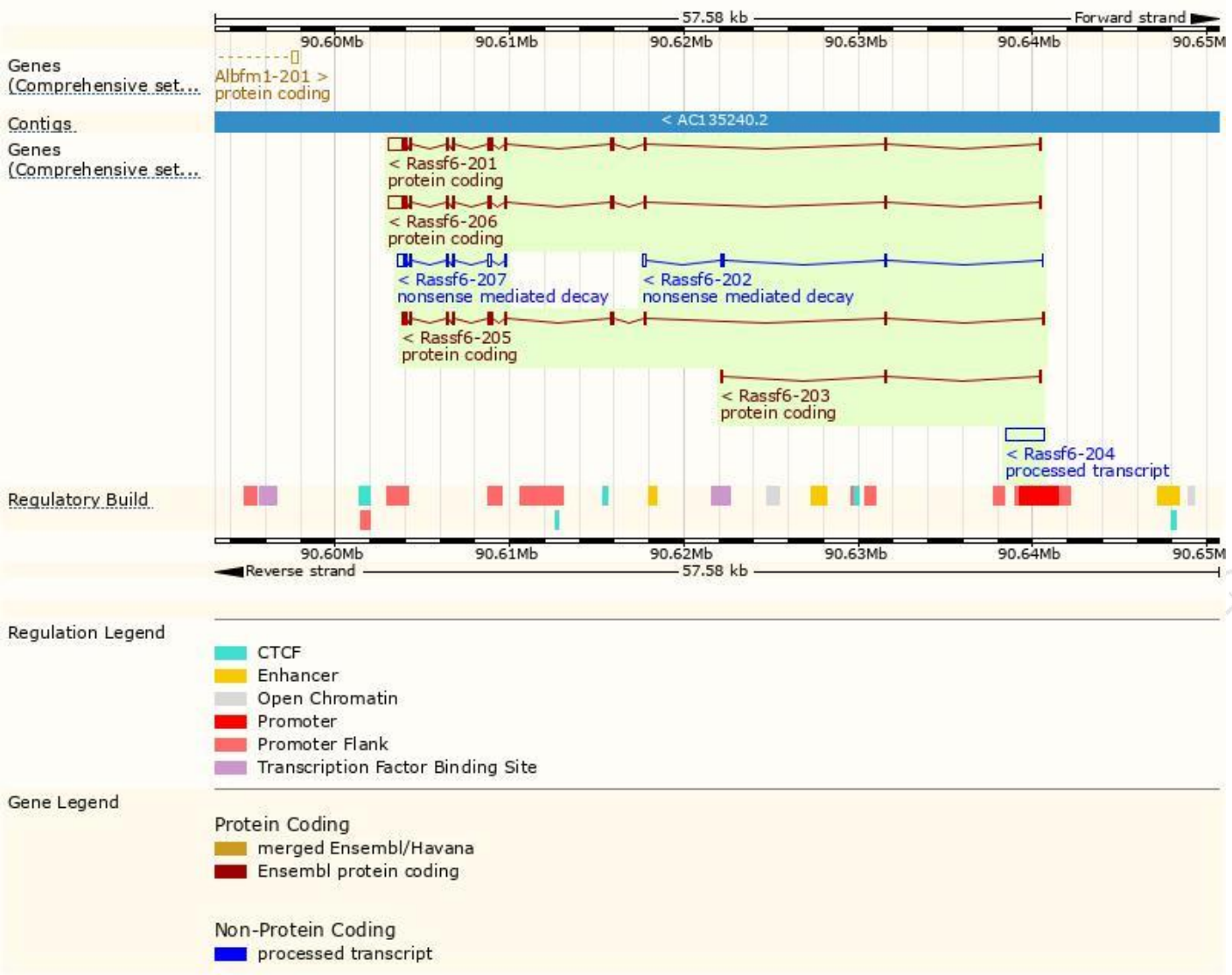
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rassf6-201	ENSMUST00000031317.9	1958	341aa	Protein coding	CCDS39142	G3X8Z9	TSL:5 GENCODE basic APPRIS P3
Rassf6-206	ENSMUST00000202784.3	1919	328aa	Protein coding	CCDS80327	Q80UQ2	TSL:1 GENCODE basic APPRIS ALT2
Rassf6-205	ENSMUST00000202704.3	1178	341aa	Protein coding	CCDS39142	G3X8Z9	TSL:5 GENCODE basic APPRIS P3
Rassf6-203	ENSMUST00000201370.1	326	50aa	Protein coding	-	A0A0J9YUI5	CDS 3' incomplete TSL:3
Rassf6-207	ENSMUST00000202807.1	959	31aa	Nonsense mediated decay	-	A0A0J9YU65	CDS 5' incomplete TSL:3
Rassf6-202	ENSMUST00000201121.3	442	41aa	Nonsense mediated decay	-	A0A0J9YVB6	TSL:3
Rassf6-204	ENSMUST00000201436.1	2184	No protein	Processed transcript	-	-	TSL:NA

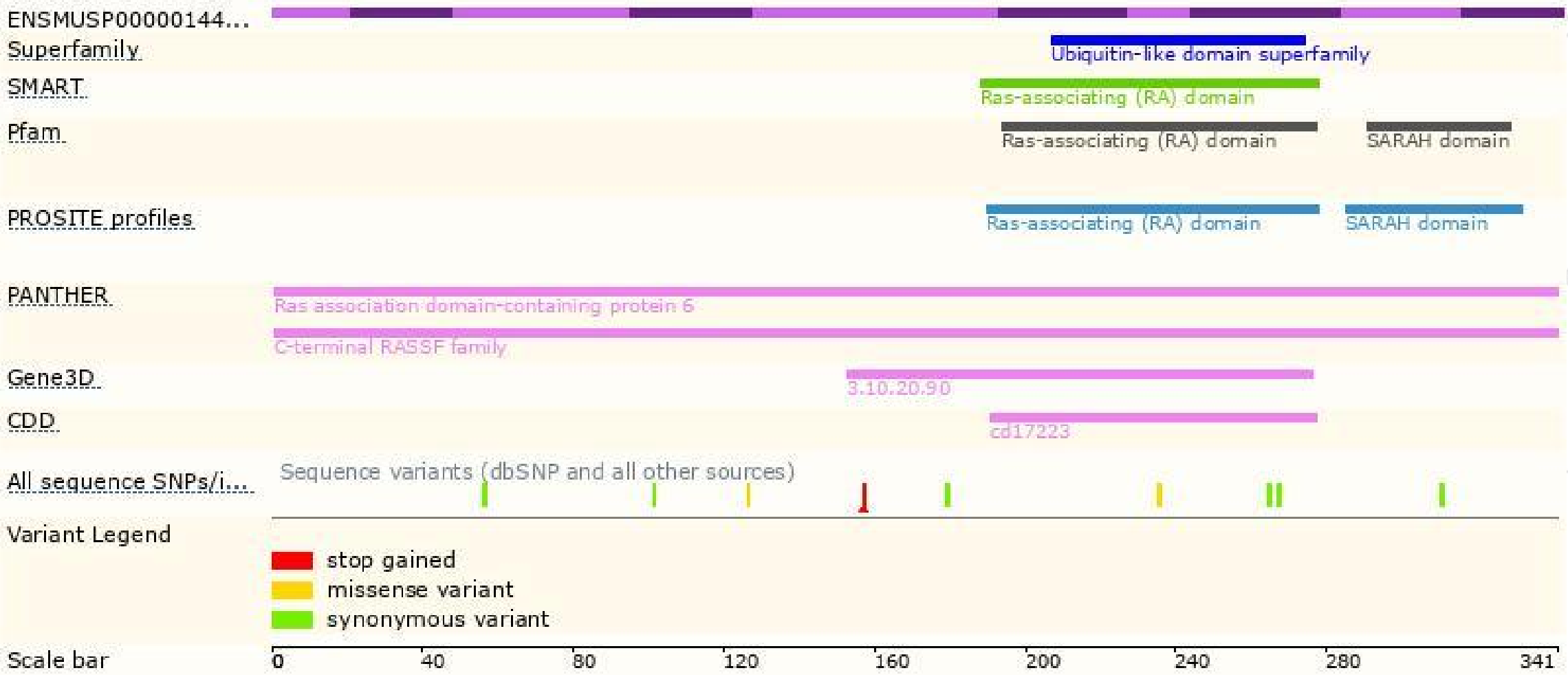
The strategy is based on the design of *Rassf6-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

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