# *Shroom2* Cas9-CKO Strategy

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# Project Overview

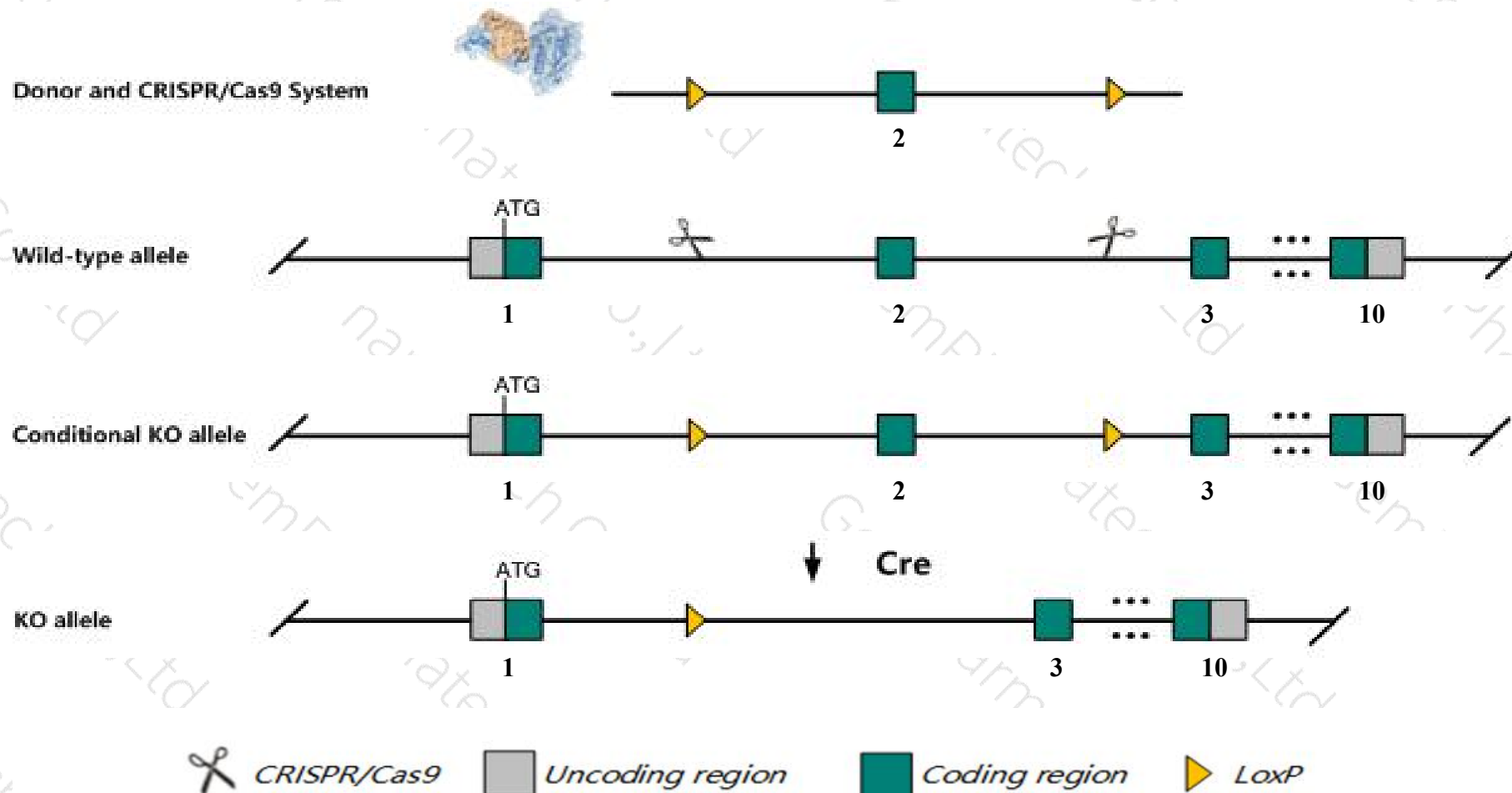| | |
|---|---|
| **Project Name** | *Shroom2* |
| **Project type** | **Cas9-CKO** |
| **Strain background** | **C57BL/6JGpt** |

# Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Shroom2* gene. The schematic diagram is as follows:



CRISPR/Cas9    Uncoding region    Coding region    LoxP

# Technical routes

➤ The *Shroom2* gene has 6 transcripts. According to the structure of *Shroom2* gene, exon2 of *Shroom2-202* (ENSMUST00000101141.8) transcript is recommended as the knockout region. The region contains Ens 152bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Shroom2* 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

➢ The *Shroom2* gene is located on the ChrX. 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）

## Shroom2 shroom family member 2 [Mus musculus (house mouse)]

Gene ID: 110380, updated on 20-Mar-2020

### ▲ Summary

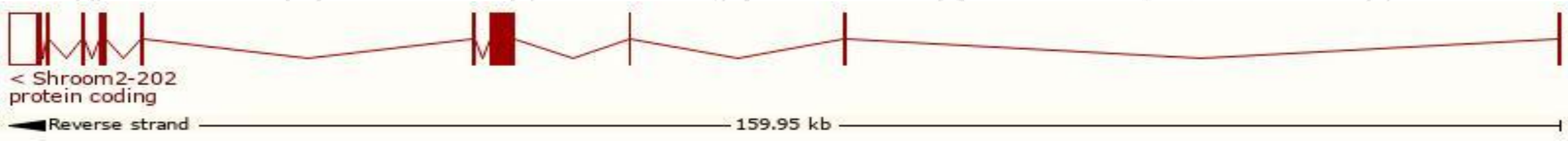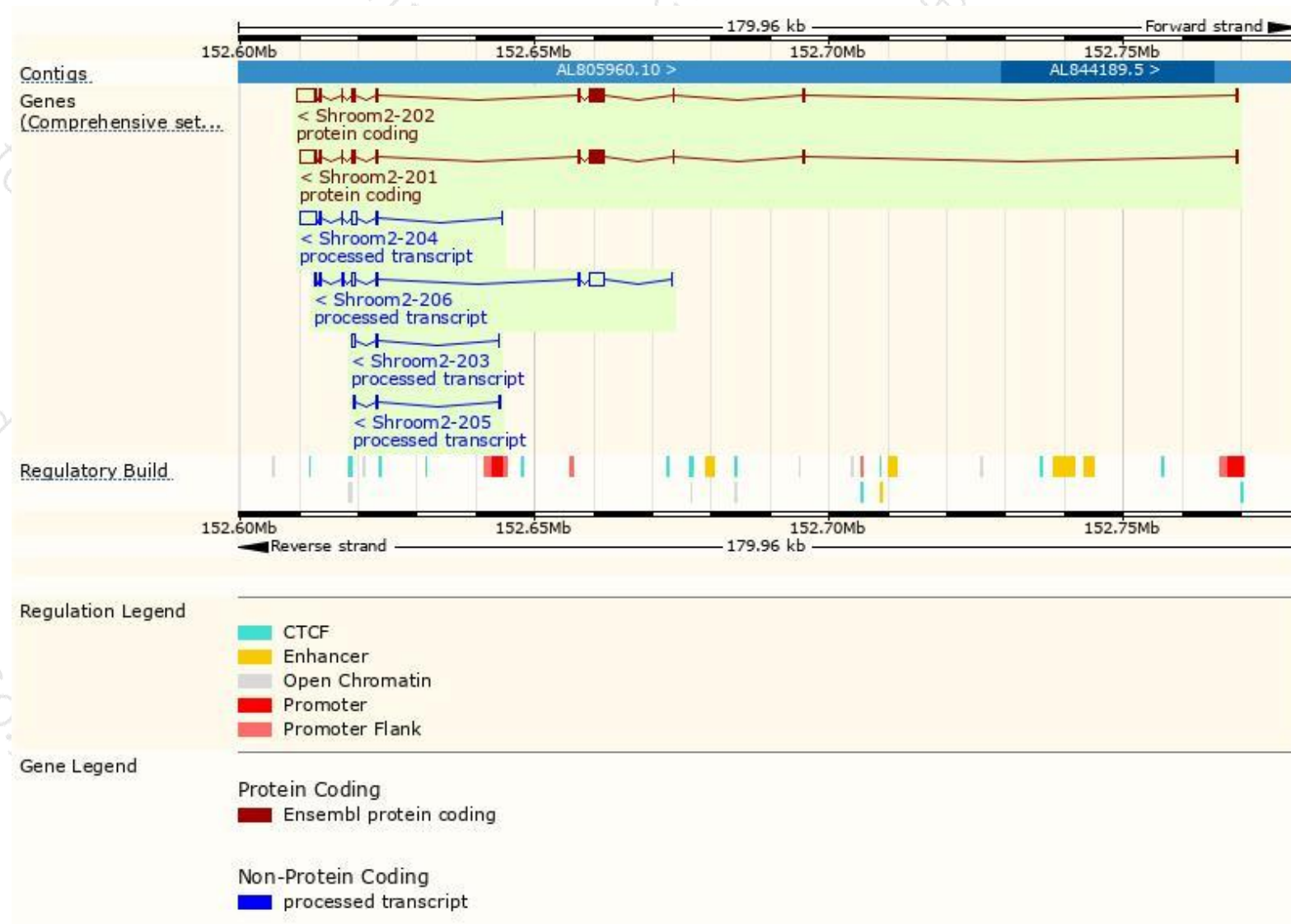| | |
|---|---|
| **Official Symbol** | Shroom2 provided by MGI |
| **Official Full Name** | shroom family member 2 provided by MGI |
| **Primary source** | MGI:MGI:107194 |
| **See related** | Ensembl:ENSMUSG00000045180 |
| **Gene type** | protein coding |
| **RefSeq status** | VALIDATED |
| **Organism** | Mus musculus |
| **Lineage** | Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus |
| **Also known as** | 4832440C16, Apxl, C630003H05Rik, Shrm2 |
| **Expression** | Broad expression in CNS E18 (RPKM 10.6), frontal lobe adult (RPKM 7.9) and 22 other tissuesSee more |
| **Orthologs** | human all |

# Transcript information（Ensembl）

The gene has 6 transcripts,all transcripts are shown below:

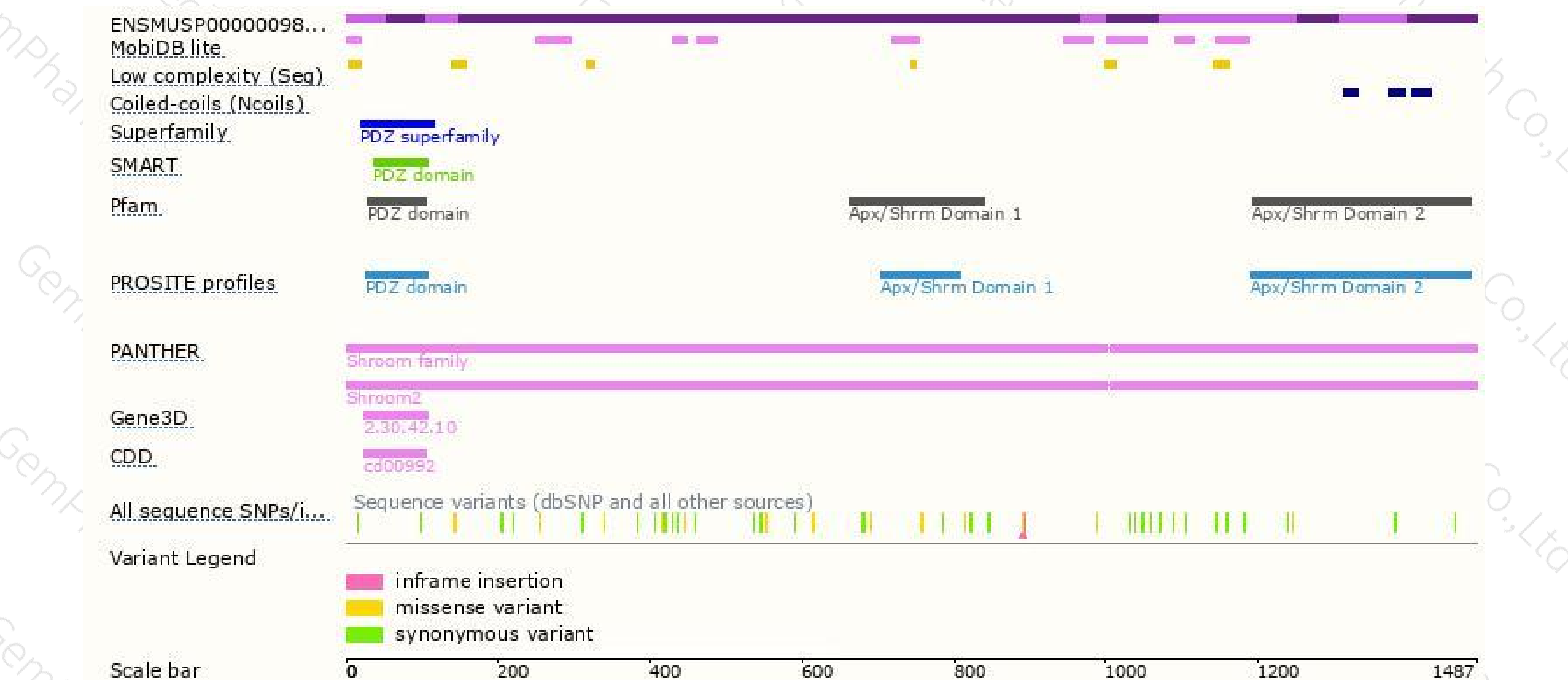| Name | Transcript ID | bp | Protein | Biotype | CCDS | UniProt | Flags |
|---|---|---|---|---|---|---|---|
| Shroom2-202 | ENSMUST00000101141.8 | 7494 | 1487aa | Protein coding | CCDS41179 | A7TU71 | TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3 |
| Shroom2-201 | ENSMUST00000062317.4 | 6997 | 1481aa | Protein coding | CCDS72455 | A2ALU4 | 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 ALT2 |
| Shroom2-204 | ENSMUST00000137217.7 | 4338 | No protein | Processed transcript | - | - | TSL:1 |
| Shroom2-206 | ENSMUST00000152746.1 | 4251 | No protein | Processed transcript | - | - | TSL:5 |
| Shroom2-203 | ENSMUST00000124141.1 | 837 | No protein | Processed transcript | - | - | TSL:3 |
| Shroom2-205 | ENSMUST00000141677.1 | 832 | No protein | Processed transcript | - | - | TSL:2 |

The strategy is based on the design of *Shroom2-202* transcript,the transcription is shown below:

# Genomic location distribution

# Protein domain



| | | | | |
|---|---|---|---|---|
| ENSMUSP00000098... | | | | |
| MobiDB lite | | | | |
| Low complexity (Seg) | | | | |
| Coiled-coils (Ncoils) | | | | |
| Superfamily | PDZ superfamily | | | |
| SMART | PDZ domain | | | |
| Pfam | PDZ domain | Apx/Shrm Domain 1 | | Apx/Shrm Domain 2 |
| PROSITE profiles | PDZ domain | Apx/Shrm Domain 1 | | Apx/Shrm Domain 2 |
| PANTHER | Shroom family | | | |
| | Shroom2 | | | |
| Gene3D | 2.30.42.10 | | | |
| CDD | cd00992 | | | |
| All sequence SNPs/i... | Sequence variants (dbSNP and all other sources) | | | |
| Variant Legend | | | | |

Variant Legend
- inframe insertion
- missense variant
- synonymous variant

Scale bar: 0   200   400   600   800   1000   1200   1487

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