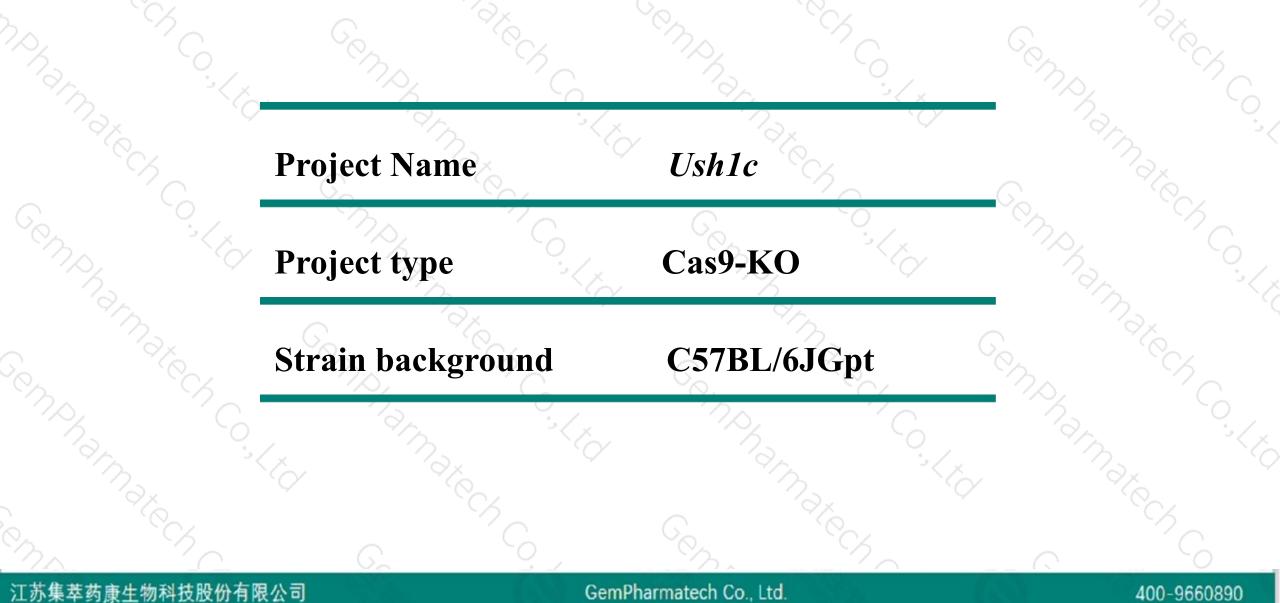


# Ush1c Cas9-KO Strategy

Designer: Reviewer: Design Date: Ruirui Zhang Huimin Su 2019-11-13

## **Project Overview**

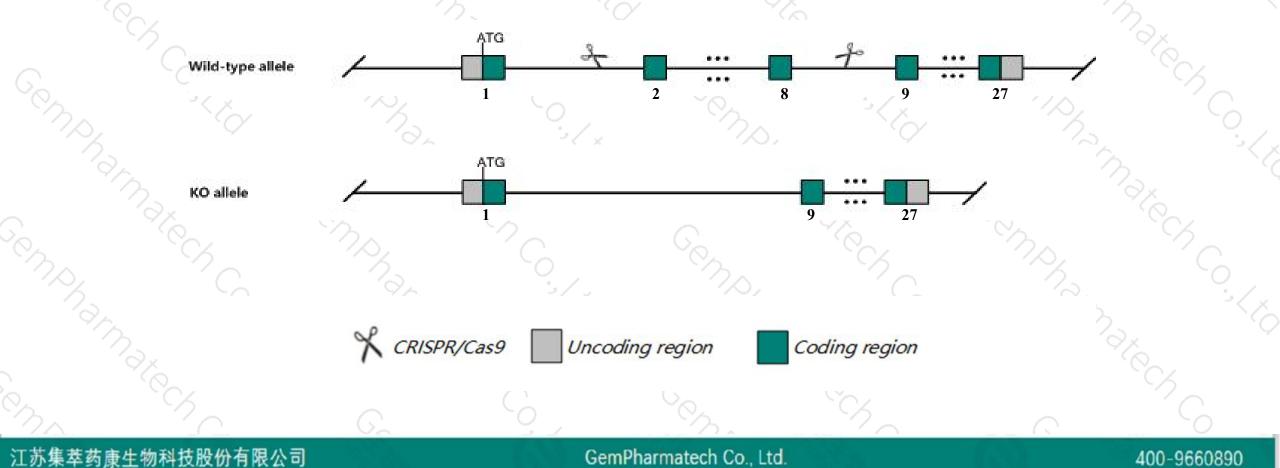




# **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the Ush1c gene. The schematic diagram is as follows:





- The Ush1c gene has 9 transcripts. According to the structure of Ush1c gene, exon2-exon8 of Ush1c-201 (ENSMUST0000009667.11) transcript is recommended as the knockout region. The region contains 638bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify Ush1c gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA 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.

- According to the existing MGI data, mutations at this locus affect hearing and result in movement anomalies generally associated with vestibular mutants, such as head tossing and circling.
  - The Ush1c gene is located on the Chr7. 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.

Notice

# Gene information (NCBI)

#### Ush1c USH1 protein network component harmonin [ Mus musculus (house mouse) ]

| Gene ID: 72088 | , updated on | 24-Sep-2019 |
|----------------|--------------|-------------|
|----------------|--------------|-------------|

#### Summary

| Official Symbol           | Ush1c provided by MGI   |
|---------------------------|---|
| <b>Official Full Name</b> | USH1 protein network component harmonin provided by MGI   |
| Primary source            | MGI:MGI:1919338   |
| See related               | Ensembl:ENSMUSG0000030838   |
| Gene type                 | protein coding  |
| <b>RefSeq status</b>      | 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             | harmonin; 2010016F01Rik   |
| Expression                | Biased expression in large intestine adult (RPKM 31.9), colon adult (RPKM 22.1) and 4 other tissues See more              |
| Orthologs                 | human all   |
| 760                       | S S S S S S S S S S S S S S S S S S S   |

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# **Transcript information (Ensembl)**



| Name 🍦    | Transcript ID 💧      | bp 🖕              | Protein 🖕    | Biotype 🝦               | CCDS 🖕             | UniProt 🖕       | Flags                           |
|-----------|----------------------|-------------------|--------------|-------------------------|--------------------|-----------------|---------------------------------|
| Jsh1c-201 | ENSMUST0000009667.11 | 3072              | <u>910aa</u> | Protein coding          | CCDS21276          | <u>E9QMN1</u> & | TSL:1 GENCODE basic APPRIS P3   |
| Jsh1c-206 | ENSMUST00000154292.8 | 2767              | <u>891aa</u> | Protein coding          | <u>CCDS52250</u>   | <u>E9PYX1</u> & | TSL:1 GENCODE basic             |
| Ush1c-209 | ENSMUST00000222454.1 | 2580              | <u>859aa</u> | Protein coding          | <u>CCDS85309</u> & | D6RIM8 &        | TSL:5 GENCODE basic             |
| Ush1c-202 | ENSMUST0000078680.12 | 2047              | <u>548aa</u> | Protein coding          | <u>CCDS39963</u> & | A0A0R4J0Z8      | TSL:1 GENCODE basic APPRIS ALT2 |
| Ush1c-207 | ENSMUST00000176371.1 | 1865              | <u>517aa</u> | Protein coding          | -                  | <u>H3BIZ2</u>   | TSL:5 GENCODE basic             |
| Jsh1c-208 | ENSMUST00000177212.7 | <mark>1696</mark> | <u>529aa</u> | Protein coding          |                    | H3BLC4          | TSL:5 GENCODE basic             |
| Ush1c-210 | ENSMUST00000238793.1 | 711               | <u>223aa</u> | Protein coding          | -                  | 1               | CDS 3' incomplete               |
| Ush1c-204 | ENSMUST00000143155.7 | 3036              | <u>859aa</u> | Nonsense mediated decay | 141                | <u>D6RIM8</u>   | TSL:1                           |
| Ush1c-203 | ENSMUST00000129266.2 | 708               | No protein   | Retained intron         | -                  | -               | TSL:5                           |
| Ush1c-205 | ENSMUST00000148527.1 | 395               | No protein   | IncRNA                  | -                  | -               | TSL:3                           |

The strategy is based on the design of *Ush1c-201* transcript, the transcription is shown below:

Reverse strand

< Ush1c-201 protein coding

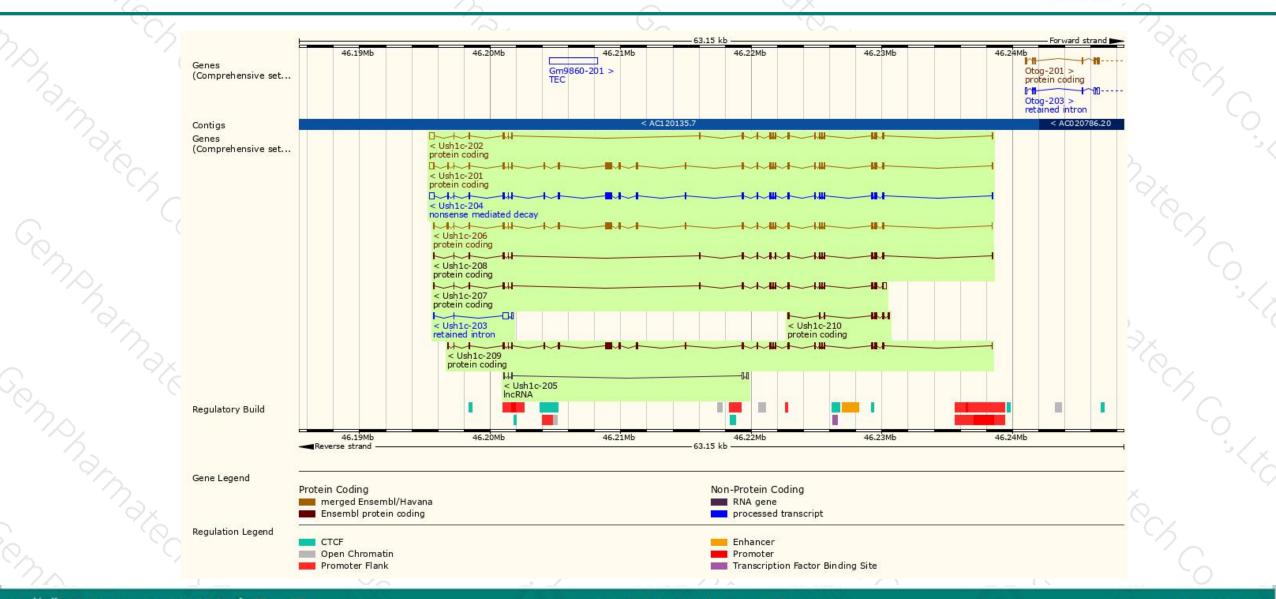
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### **Genomic location distribution**



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集举约康 GemPharmatech

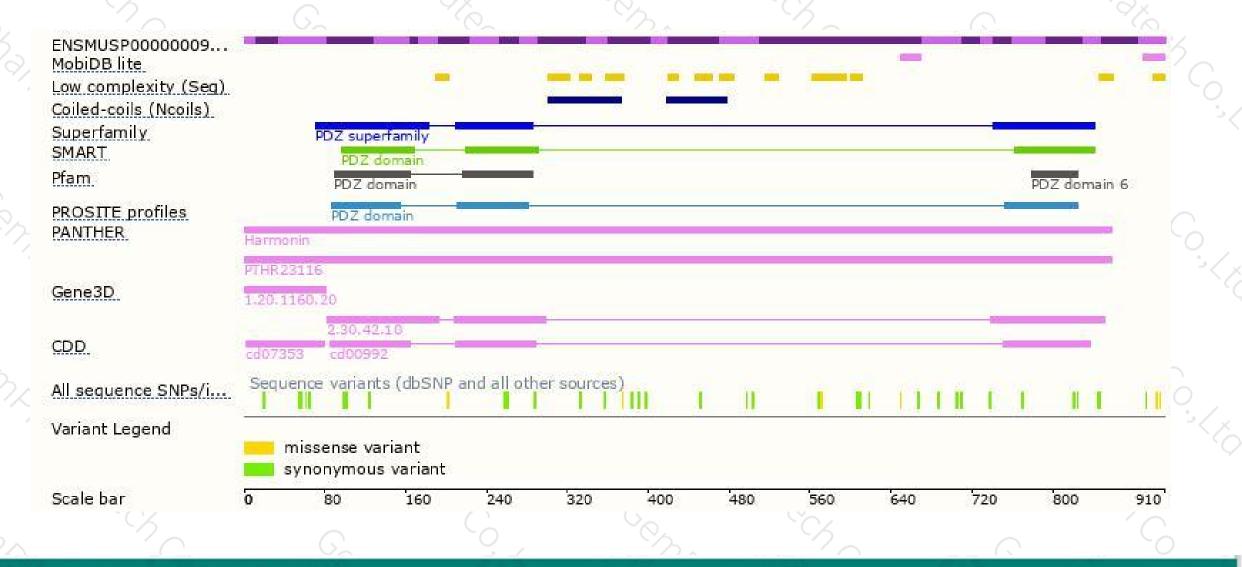
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### **Protein domain**



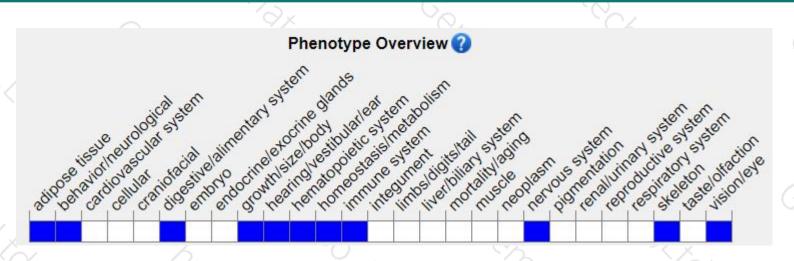


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### Mouse phenotype description(MGI)



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, mutations at this locus affect hearing and result in movement anomalies generally associated with vestibular mutants, such as head tossing and circling.

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



