GemPharmatech™

Adgrl2 Cas9-CKO Strategy

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Design Date: 2023.11.28

Overview

Target Gene Name

- Adgrl2

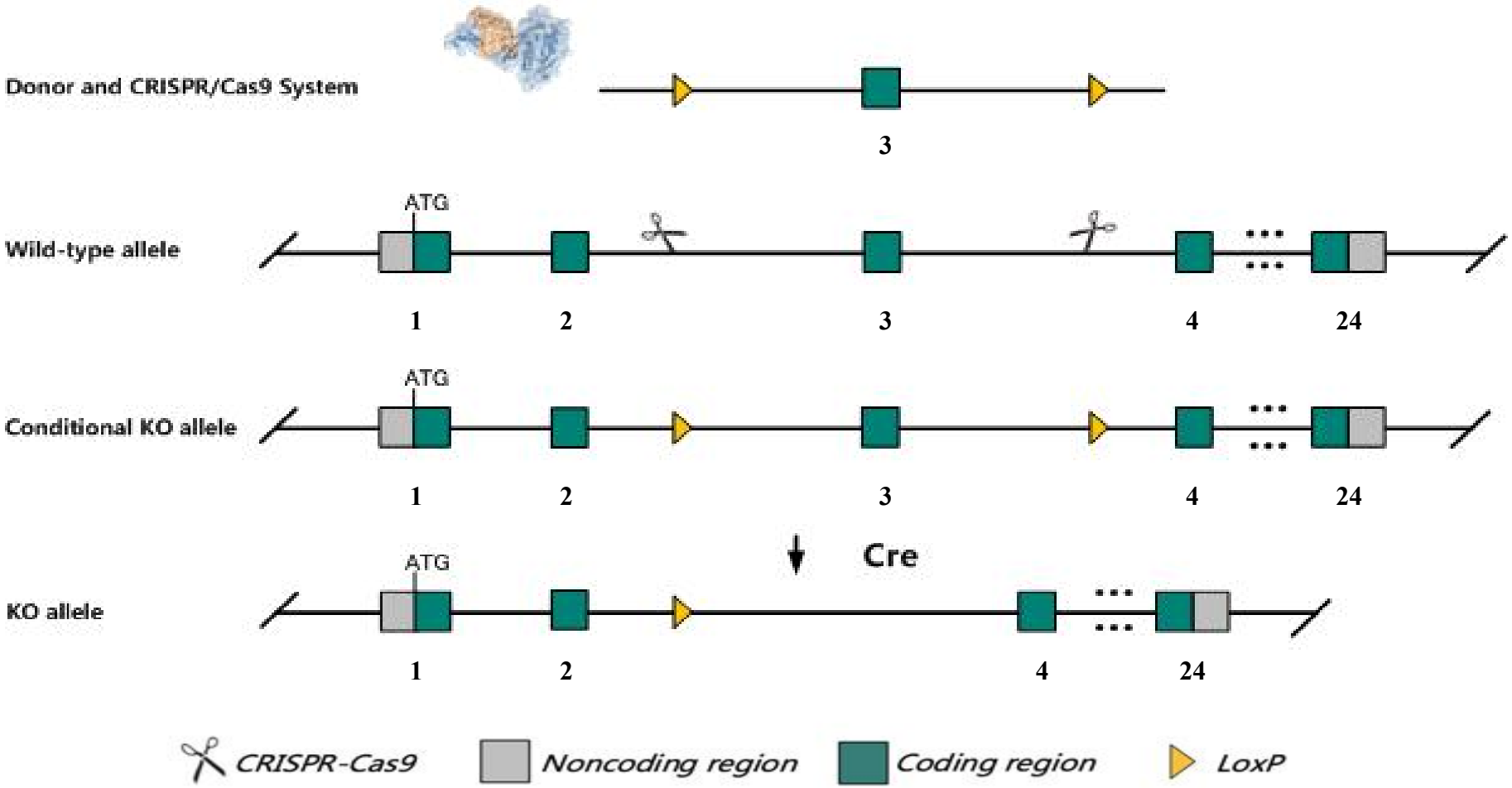
Project Type

- Cas9-CKO

Genetic Background

- C57BL/6JGpt

Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the *Adgrl2* gene.

Technical Information

- The *Adgrl2* gene has 20 transcripts. According to the structure of *Adgrl2* gene, exon3 of *Adgrl2*-208 (ENSMUST00000197567.5) transcript is recommended as the knockout region. The region contains 110bp coding sequence. Knocking out the region will result in disruption of protein function.
- In this project we use CRISPR-Cas9 technology to modify *Adgrl2* 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.
- 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.

Gene Information

Adgrl2 adhesion G protein-coupled receptor L2 [Mus musculus (house mouse)]

Gene ID: 99633, updated on 18-May-2023

Summary

Official Symbol	Adgrl2 provided by MGI
Official Full Name	adhesion G protein-coupled receptor L2 provided by MGI
Primary source	MGI:MGI:2139714
See related	Ensembl:ENSMUSG00000028184
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	CIRL-2, Gm619, Lec1, Lphh1, Lphn2, mKIAA0786
Summary	Predicted to enable G protein-coupled receptor activity and PDZ domain binding activity. Involved in synapse organization. Acts upstream of or within positive regulation of synapse assembly and response to bacterium. Is active in glutamatergic synapse. Is integral component of postsynaptic membrane. Is expressed in heart. Orthologous to human ADGRL2 (adhesion G protein-coupled receptor L2). [provided by Alliance of Genome Resources, Apr 2022]
Expression	Broad expression in CNS E18 (RPKM 41.1), CNS E14 (RPKM 39.9) and 20 other tissues See more
Orthologs	human all

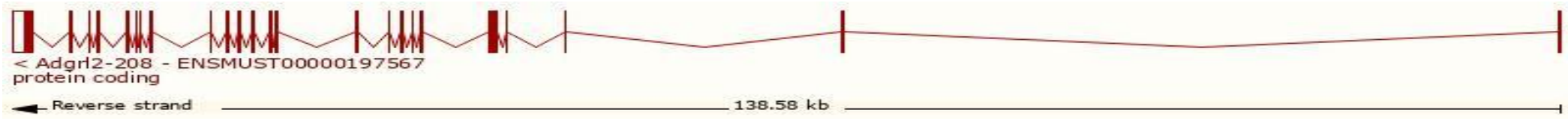
Source: https://www.ncbi.nlm.nih.gov/

Transcript Information

The gene has 20 transcripts,all transcripts are shown below:

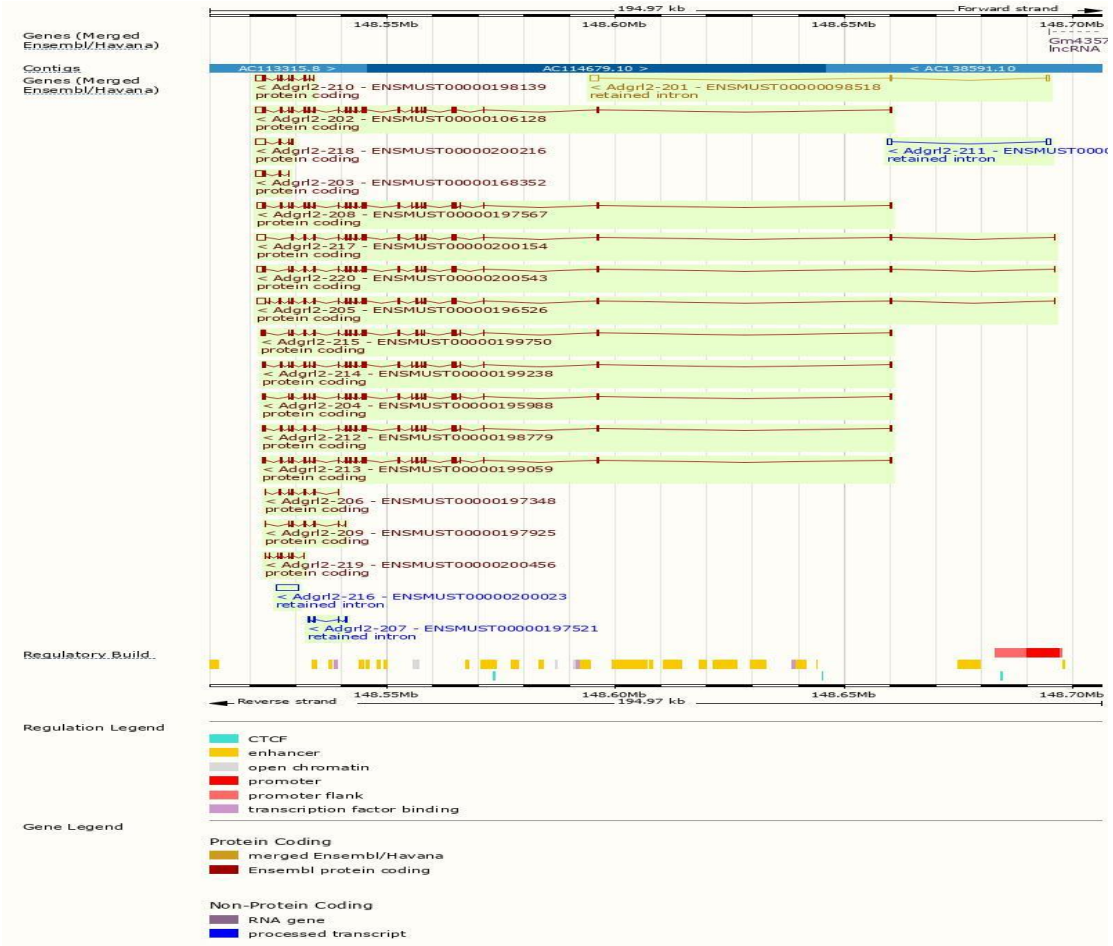
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Adgrl2-202	ENSMUST00000106128.7	6052	1487aa	Protein coding			A single transcript chosen for a gene which is the most conserved, most highly expressed, has the longest coding sequence and is represented in other key resources, such as NCBI and UniProt. This is defined in detail on http://www.ensembl.org/info/genome/genebuild/canonical.html Ensembl Canonical , The GENCODE set is the gene set for human and mouse. GENCODE basic , APPRIS ALT1 , TSL:5 ,
Adgrl2-205	ENSMUST00000196526.5	5615	1177aa	Protein coding			The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:5 ,
Adgrl2-208	ENSMUST00000197567.5	5585	1487aa	Protein coding	CCDS80059		The GENCODE set is the gene set for human and mouse. GENCODE basic , APPRIS P4 , TSL:2 ,
Adgrl2-220	ENSMUST00000200543.5	5509	1403aa	Protein coding			The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:5 ,
Adgrl2-217	ENSMUST00000200154.5	5412	1123aa	Protein coding			The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:5 ,
Adgrl2-214	ENSMUST00000199238.5	4670	1478aa	Protein coding			The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:5 ,
Adgrl2-213	ENSMUST00000199059.5	4637	1467aa	Protein coding			The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:5 ,
Adgrl2-212	ENSMUST00000198779.5	4592	1452aa	Protein coding			The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:5 ,
Adgrl2-204	ENSMUST00000195988.5	4541	1435aa	Protein coding			The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:5 ,
Adgrl2-215	ENSMUST00000199750.5	4399	1341aa	Protein coding			The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:5 ,
Adgrl2-210	ENSMUST00000198139.5	3005	471aa	Protein coding			TSL:1 , CDS 5' incomplete ,
Adgrl2-218	ENSMUST00000200216.5	2300	79aa	Protein coding			TSL:3 , CDS 5' incomplete ,
Adgrl2-203	ENSMUST00000168352.2	2151	239aa	Protein coding			The GENCODE set is the gene set for human and mouse. GENCODE basic , TSL:1 ,
Adgrl2-206	ENSMUST00000197348.5	843	196aa	Protein coding			TSL:3 , CDS 5' incomplete ,
Adgrl2-209	ENSMUST00000197925.5	793	264aa	Protein coding			TSL:3 , CDS 5' and 3' incomplete ,
Adgrl2-219	ENSMUST00000200456.2	610	138aa	Protein coding			TSL:3 , CDS 5' incomplete ,
Adgrl2-216	ENSMUST00000200023.2	4855	No protein	Retained intron			TSL:NA ,
Adgrl2-201	ENSMUST00000098518.3	2585	No protein	Retained intron			TSL:1 ,
Adgrl2-211	ENSMUST00000198563.2	1669	No protein	Retained intron			TSL:1 ,
Adgrl2-207	ENSMUST00000197521.2	795	No protein	Retained intron			TSL:3 ,

The strategy is based on the design of *Adgrl2*-208 transcript, the transcription is shown below:



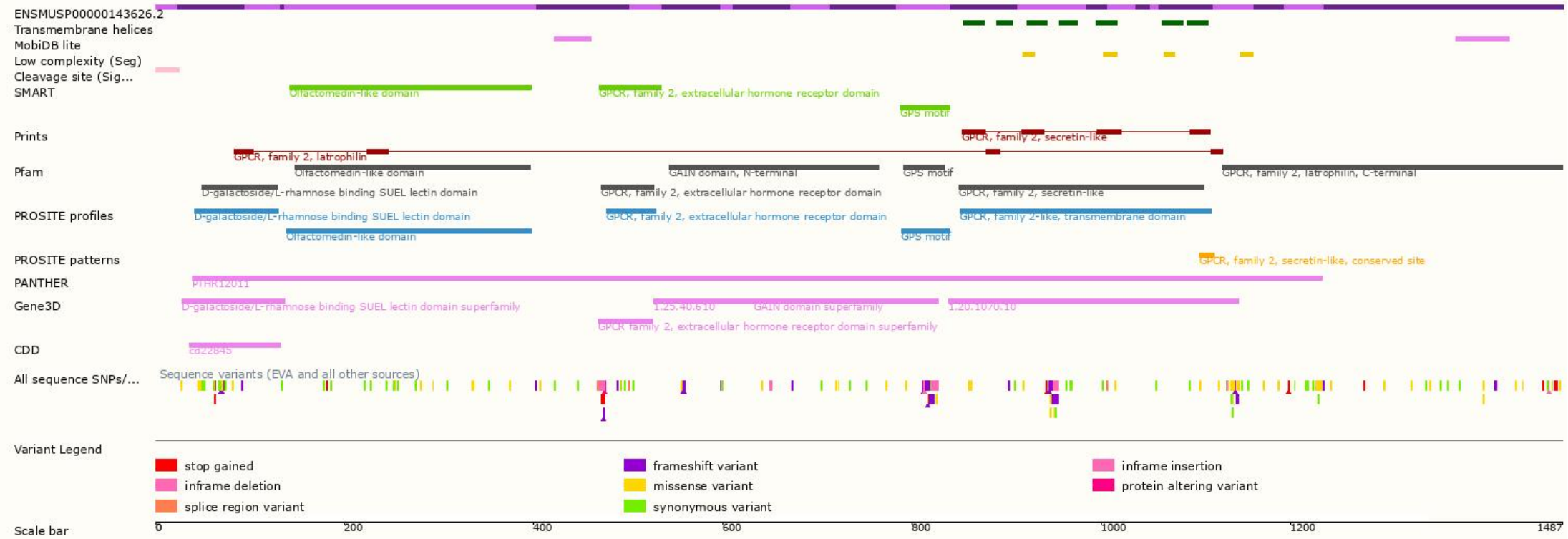
Source: https://www.ensembl.org

Genomic Information



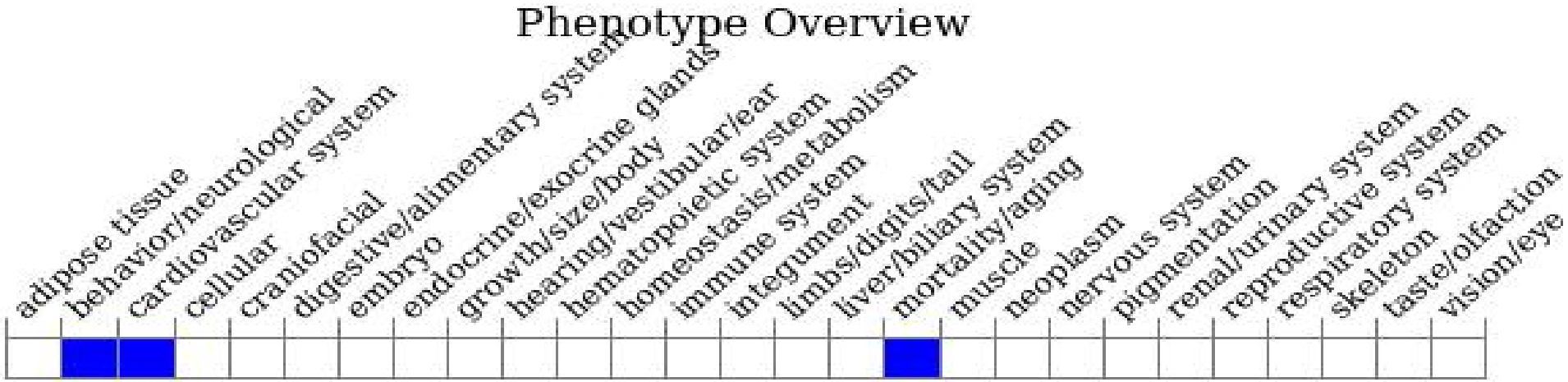
Source: https://www.ensembl.org

Protein Information



Source: : https://www.ensembl.org

Mouse Phenotype Information (MGI)



- Homozygous null mice die prenatally at fetal stages with cardiac defects. Heterozygous mice exhibit decreased locomotor activity in an open field test.

Source: https://www.informatics.jax.org

Important Information

- According to MGI, homozygous null mice die prenatally at fetal stages with cardiac defects. Heterozygous mice exhibit decreased locomotor activity in an open field test.
- The impact of this strategy on transcript *Adgrl2*-206, *Adgrl2-209, Adgrl2-210, Adgrl2*-218 *and Adgrl2*-219 are unknown.
- This stratergy may not affect *Adgrl2*-201, *Adgrl2*-203, *Adgrl2*-207, *Adgrl2*-211 and *Adgrl2*-216 transcript.
- *Adgrl2* is located on Chr3. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is 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 the existing technology level.