

# John Co. (x) Fzd2 Cas9-CKO Strategy Rond College Comphannakoch

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# **Project Overview**



Project Name Fzd2

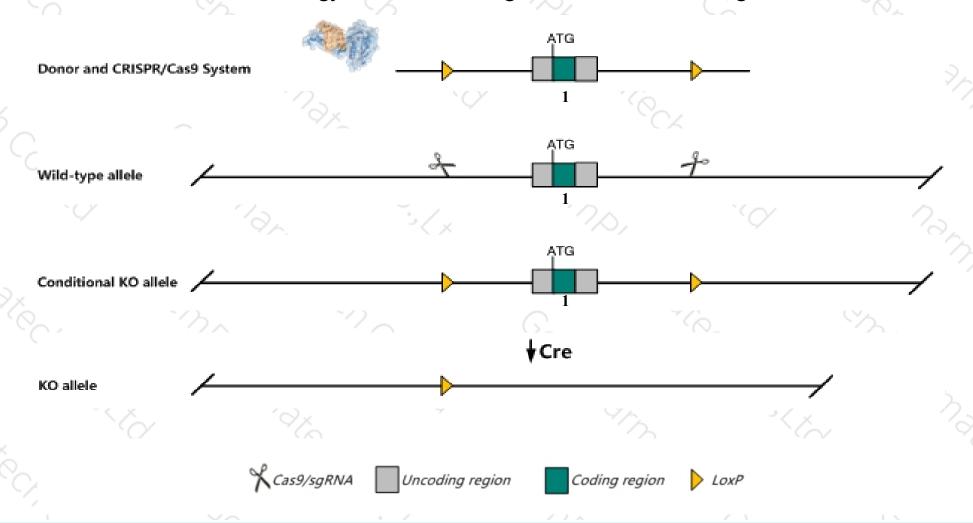
Project type Cas9-CKO

Strain background C57BL/6J

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Fzd2 gene. The schematic diagram is as follows:



### **Technical routes**



- The *Fzd2* gene has 1 transcript. According to the structure of *Fzd2* gene, exon1 of *Fzd2-201* (ENSMUST0000057893.6) transcript is recommended as the knockout region. The region contains all 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 *Fzd2* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA and Donor were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.
- > The flox mice was 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**



- > According to the existing MGI data, About 50% of mice homozygous for a reporter allele display a cleft palate and die as neonates; the remaining 50% survive exhibiting a variable degree of postnatal runting and reduced olfactory sensitivity to various odorants.
- > The *Fzd2* gene is located on the Chr11. 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)



#### Fzd2 frizzled class receptor 2 [ Mus musculus (house mouse) ]

Gene ID: 57265, updated on 31-Jan-2019

Summary

☆ ?

Official Symbol Fzd2 provided by MGI

Official Full Name frizzled class receptor 2 provided by MGI

Primary source MGI:MGI:1888513

See related Ensembl: ENSMUSG00000050288

Gene type protein coding
RefSeq status VALIDATED
Organism <u>Mus musculus</u>

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;

Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus

Also known as Fz10; Fzd10; Mfz10; Mfz10a; AL033370; AW456835

Orthologs human all

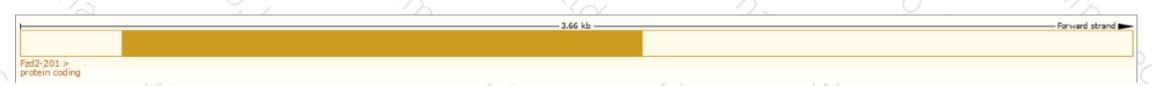
# Transcript information (Ensembl)



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

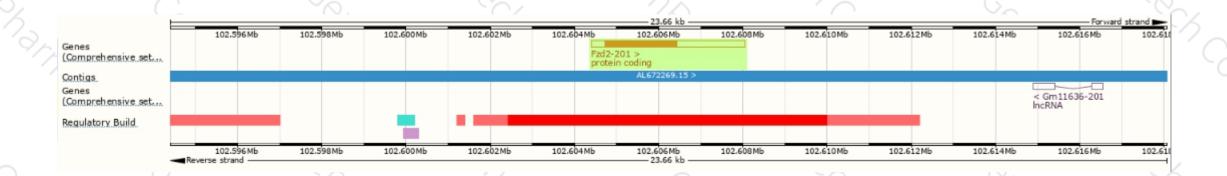
Name	Transcript ID	bp 🍦	Protein	Biotype	CCDS	UniProt	Flags		
Fzd2-201	ENSMUST00000057893.6	3663	<u>570aa</u>	Protein coding	CCDS25501@	Q9JIP6₽	TSL:NA	GENCODE basic	APPRIS P1

The strategy is based on the design of Fzd2-201 transcript, The transcription is shown below



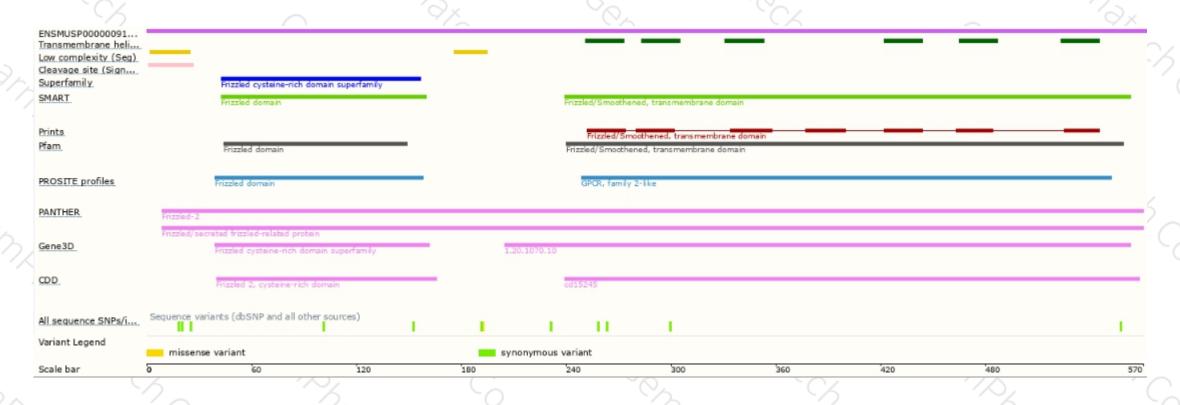
### Genomic location distribution





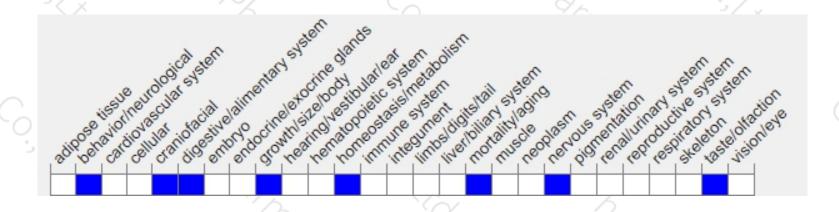
### Protein domain





# 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, About 50% of mice homozygous for a reporter allele display a cleft palate and die as neonates; the remaining 50% survive exhibiting a variable degree of postnatal runting and reduced olfactory sensitivity to various odorants.



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

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