

# Htr6 Cas9-CKO Strategy

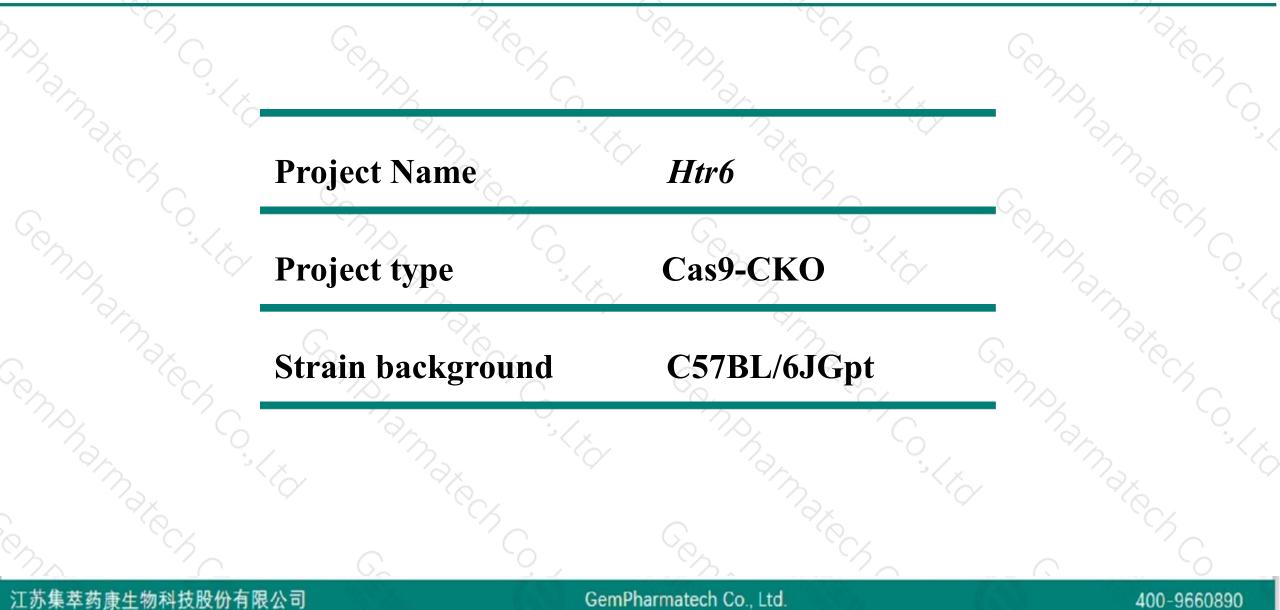
Designer: Design Date:

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Jinling Wang 2019-7-17

## **Project Overview**

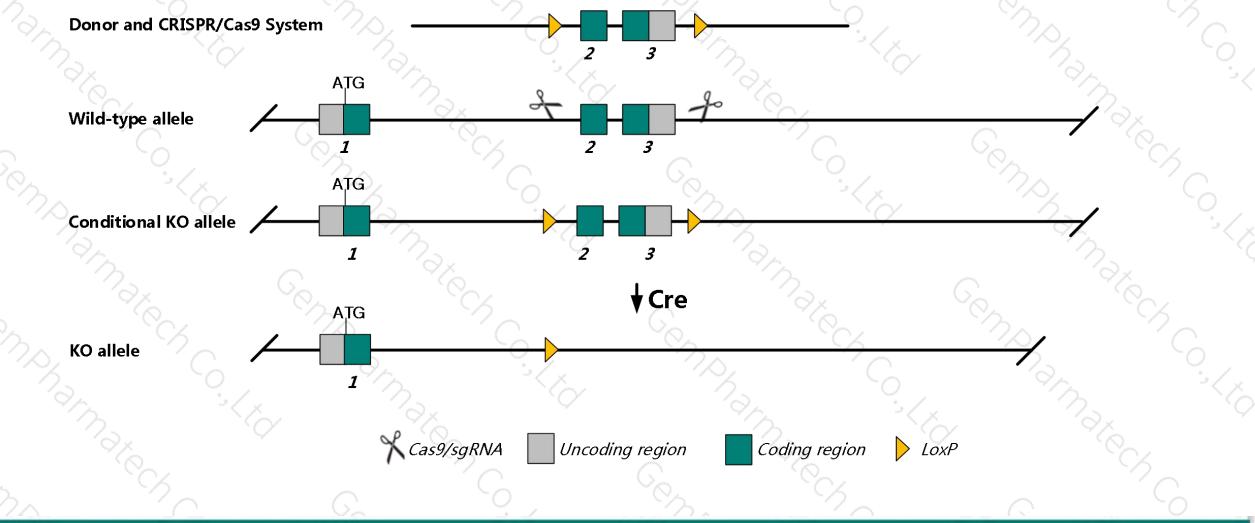




## **Conditional Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Htr6* gene. The schematic diagram is as follows:



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The *Htr6* gene has 2 transcripts. According to the structure of *Htr6* gene, exon2-exon3 of *Htr6-201* (ENSMUST00000068036.1) transcript is recommended as the knockout region. The region contains 603bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Htr6* 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.



- According to the existing MGI data, Male mice homozygous for some disruptions in this gene display decreased body size. Mice homozygous for a different null allele display decreased sensitivity to alcohol induced behavioral responses.
- The *Htr6* gene is located on the Chr4. 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)**



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#### Htr6 5-hydroxytryptamine (serotonin) receptor 6 [Mus musculus (house mouse)]

Gene ID: 15565, updated on 19-Mar-2019

#### Summary

Official Symbol	Htr6 provided by MGI
Official Full Name	5-hydroxytryptamine (serotonin) receptor 6 provided byMGI
Primary source	MGI:MGI:1196627
See related	Ensembl:ENSMUSG0000028747
Gene type	protein coding
<b>RefSeq status</b>	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;
	Muroidea; Muridae; Murinae; Mus; Mus
Also known as	5-HT6
Expression	Biased expression in cortex adult (RPKM 1.8), frontal lobe adult (RPKM 1.5) and 11 other tissues See more
Orthologs	human all



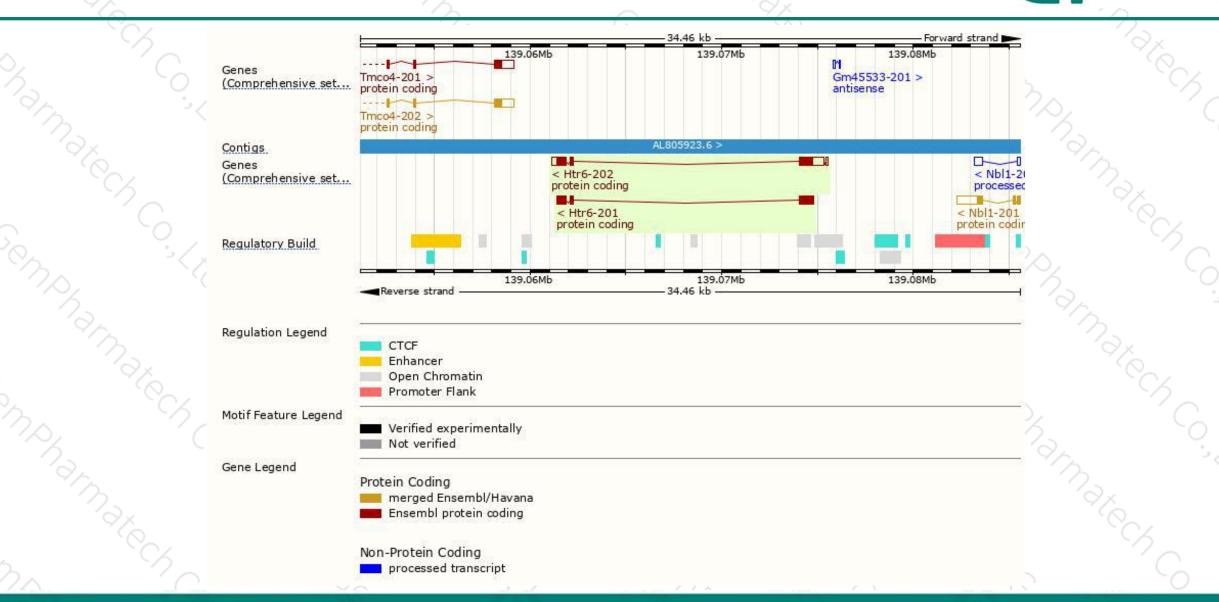
The gene has 2 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Htr6-202	ENSMUST00000105802.7	2341	<u>440aa</u>	Protein coding	CCDS18838	Q14AW8 Q9R1C8	TSL:5 GENCODE basic APPRIS P1
Htr6-201	ENSMUST0000068036.1	1362	<u>440aa</u>	Protein coding	CCDS18838	Q14AW8 Q9R1C8	TSL:1 GENCODE basic APPRIS P1

The strategy is based on the design of *Htr6-201* transcript, The transcription is shown below



### **Genomic location distribution**



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## **Protein domain**



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ENSMUSP00000068 Transmembrane heli Low complexity (Seg) Conserved Domains hmmpanther	РТНR24247				_	
Superfamily domains SMART domains	PTHR24247:SF114 SSF81321 G protein-coupled receptor, rhoo	Insin-like				
Prints domain	G protein-coupled receptor, rhodopsin	and a second				
Pfam domain	PR01102 G protein-coupled receptor,	rhodopsin-like			_	
PROSITE profiles PROSITE patterns Gene3D	GPCR, rhodopsin-like, 7TM	G protein-coupled receptor, rhodopsin-like				
All sequence SNPs/i		<u>1</u>		T TT 10	1.10	i i z
Variant Legend	missense variant	synonymous	variant			
Scale bar	0 40 80	120 160	200 240	280	320 360	400 440
ensharm.		Sharn She	r Complex	Nate Ch Co.	Cens,	
n N	NC YC			nate ch	×~	A Ch

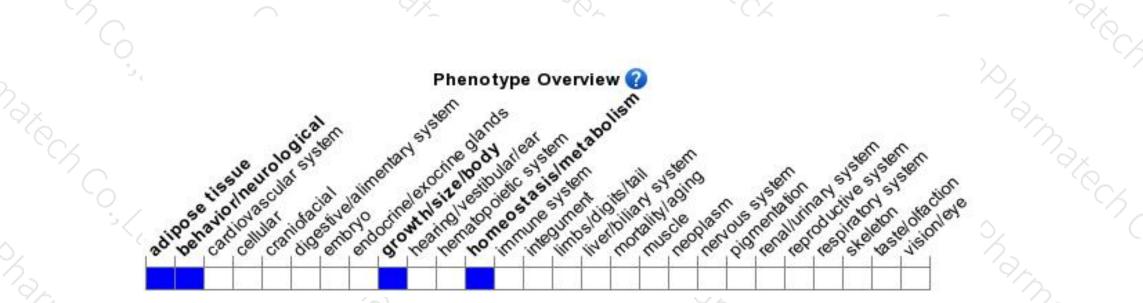
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## Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

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



