

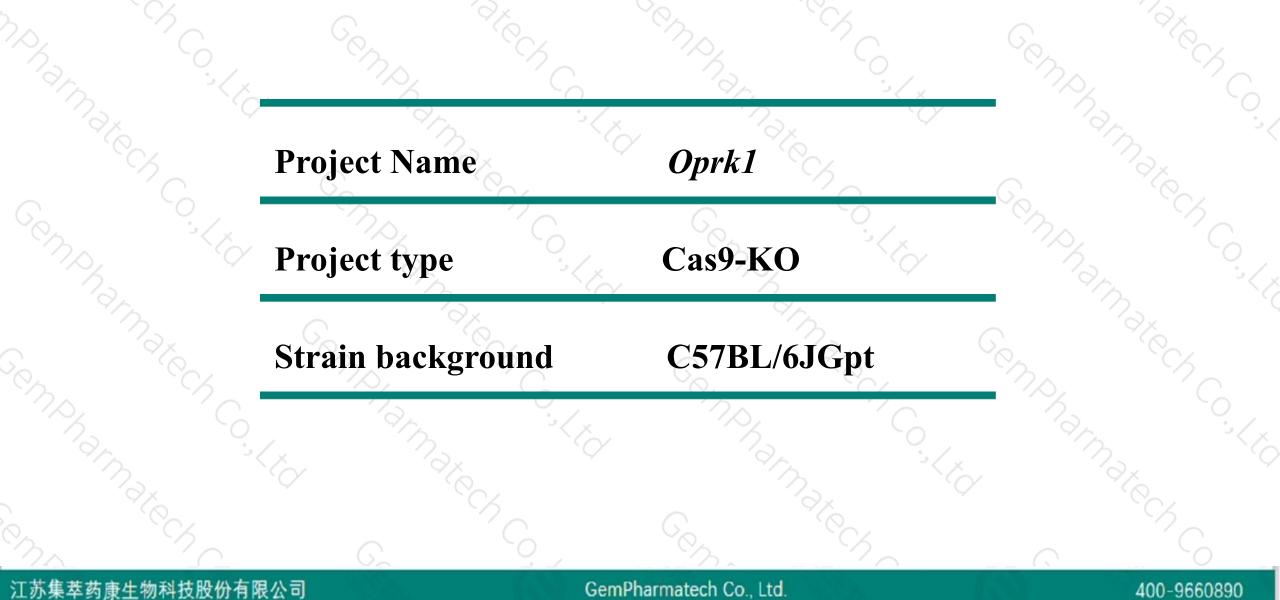
Oprk1 Cas9-KO Strategy

Designer: Reviewer Design Date: Ruirui Zhang Huimin Su

2019-8-19

Project Overview

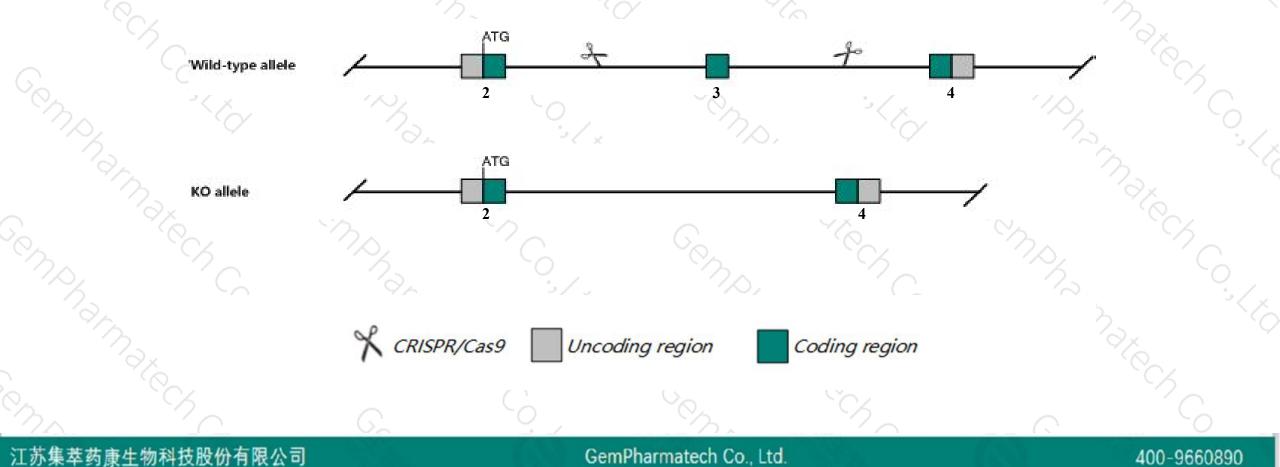




Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Oprk1 gene. The schematic diagram is as follows:





- The Oprk1 gene has 5 transcripts. According to the structure of Oprk1 gene, exon3 of Oprk1-204 (ENSMUST00000160777.7) transcript is recommended as the knockout region. The region contains 353bp coding sequence. Knock out the region will result in disruption of protein function.
- > In this project we use CRISPR/Cas9 technology to modify Oprk1 gene. The brief process is as follows: CRISPR/Cas9 system

- According to the existing MGI data, mice homozygous for a knock-out allele exhibit impaired response to morphine and an opioid agonist, abnormal pain threshold, and increased litter size.
- > The Oprk1 gene is located on the Chr1. 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)



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Oprk1 opioid receptor, kappa 1 [Mus musculus (house mouse)]

Gene ID: 18387, updated on 12-Aug-2019

Summary

Official Symbol Oprk1 provided by MGI Official Full Name opioid receptor, kappa 1 provided by MGI Primary source MGI:MGI:97439 Ensembl:ENSMUSG0000025905 See related Gene type protein coding **RefSeg status** REVIEWED Organism Mus musculus Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Lineage Murinae; Mus; Mus KOR; R21; KOR-1; MSL-1; Oprk2; K-OR-1 Also known as This gene encodes an opioid receptor, which is a member of the 7 transmembrane-spanning G protein-coupled receptor family. It functions as a receptor for Summary endogenous ligands, as well as a receptor for various synthetic opioids. Ligand binding results in inhibition of adenylate cyclase activity and neurotransmitter release. This opioid receptor plays a role in the perception of pain and mediating the hypolocomotor, analgesic and aversive actions of synthetic opioids. Variations in this gene have also been associated with alcohol dependence and opiate addiction. Alternatively spliced transcript variants have been found for this gene. A recent study provided evidence for translational readthrough in this gene, and expression of an additional C-terminally extended isoform via the use of an alternative in-frame translation termination codon. [provided by RefSeq, Dec 2017] Expression Biased expression in frontal lobe adult (RPKM 1.1), cortex adult (RPKM 0.9) and 8 other tissues See more Orthologs human all

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Transcript information (Ensembl)



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The gene has 5 transcripts, all transcripts are shown below:

Name 🖕	Transcript ID	bp 💧	Protein 🖕	Biotype 💧	CCDS	UniProt 💧	Flags 🖕	
Oprk1-201	ENSMUST0000027038.10	1376	<u>380aa</u>	Protein coding	<u>CCDS14809</u> &	P33534 @ Q14AL5 @	TSL:1 GENCODE basic APPRIS P1	
Oprk1-202	ENSMUST00000159083.1	3327	No protein	Retained intron	-	8-	TSL:1	
Oprk1-203	ENSMUST00000160339.1	2481	<u>380aa</u>	Protein coding	<u>CCDS14809</u> 교	P33534 & Q14AL5 &	TSL:5 GENCODE basic APPRIS P1	
Oprk1-204	ENSMUST00000160777.7	4675	<u>380aa</u>	Protein coding	<u>CCDS14809</u> &	P33534 @ Q14AL5 @	TSL:1 GENCODE basic APPRIS P1	
Oprk1-205	ENSMUST00000192685.1	2846	No protein	Retained intron	171	às t ai	TSL:NA	

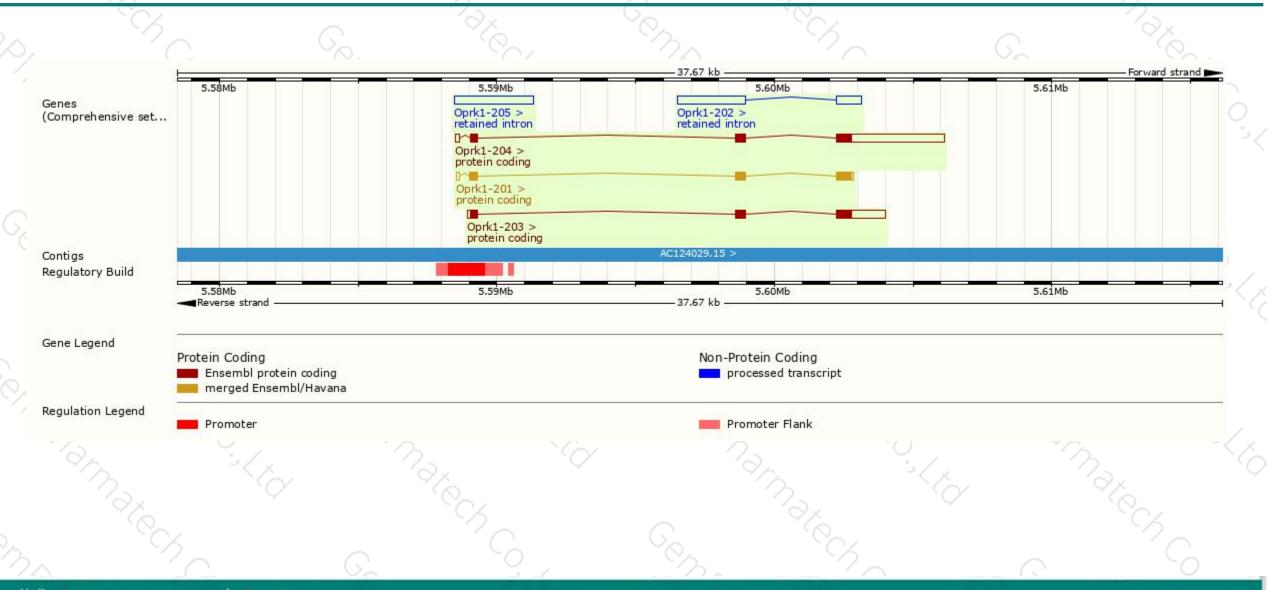
The strategy is based on the design of Oprk1-204 transcript, The transcription is shown below

Oprk	1-204 > ain coding		1	7.64 kb		F	Forward strand
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	° CH	6.	G.	Gen a	C'A O	C	30

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Genomic location distribution



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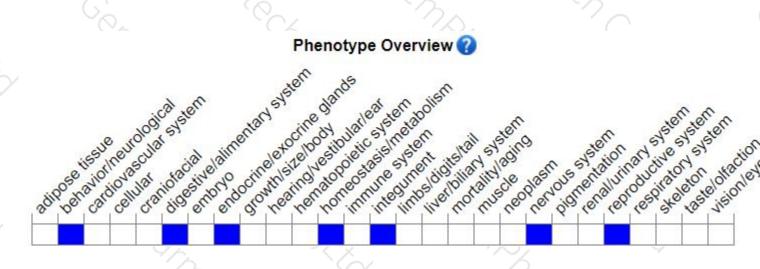
Protein domain



20				So.					
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			Op	ioid receptor					-
		Kappa opio	d receptor			_			
	Pfam		G protei	n-coupled receptor, rhodopsin-li	ke				
	PROSITE profiles		GPCR, r	nodopsin-like, 7TM					
	PROSITE patterns			G	protein-coupled receptor, rh	od opsin-like			
	PANTHER	PTHR24229:SF1							,/
		PTHR.24229							\
	Gene3D		1.20.1070.10						
	CDD		Kappa opioid rec	ptor					-
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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, Mice homozygous for a knock-out allele exhibit impaired response to morphine and an opioid agonist, abnormal pain threshold, and increased litter size.

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



