

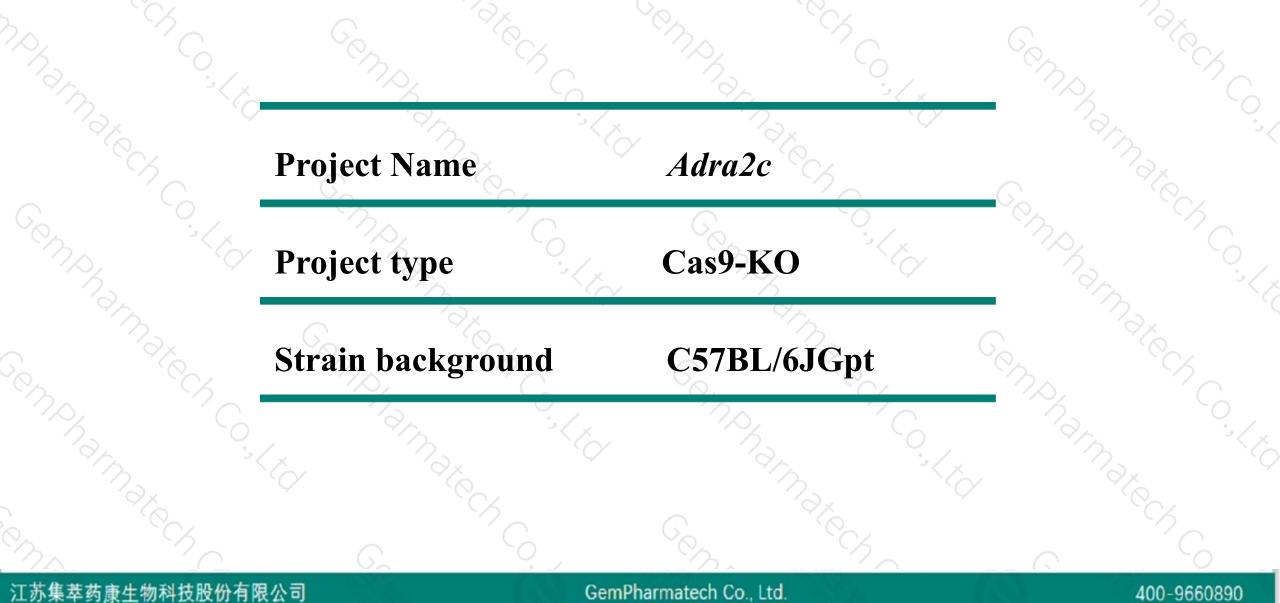
# Adra2c Cas9-KO Strategy

0

Designer: Xiaojing Li Design Date: 2019-9-19 Reviewer: JiaYu

### **Project Overview**





## **Knockout** strategy



This model will use CRISPR/Cas9 technology to edit the Adra2c gene. The schematic diagram is as follows:



KO allele

CRISPR/Cas9 Uncoding region

Coding region

ATG

#### 江苏集萃药康生物科技股份有限公司

#### GemPharmatech Co., Ltd.

#### 400-9660890



The Adra2c gene has 1 transcript. According to the structure of Adra2c gene, exon1 of Adra2c-201 (ENSMUST00000049545.6) 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 *Adra2c* 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.



- > Mice homozygous for targeted mutations that inactivate the gene are viable and fertile and appear grossly normal.
- The Adra2c gene is located on the Chr5. 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.

# **Gene information (NCBI)**



#### Adra2c adrenergic receptor, alpha 2c [ Mus musculus (house mouse) ]

Gene ID: 11553, updated on 12-Aug-2019

#### Summary

Official Symbol Adra2c provided by MGI Official Full Name adrenergic receptor, alpha 2c provided by MGI MGI:MGI:87936 Primary source Ensembl:ENSMUSG0000045318 See related Gene type protein coding VALIDATED **RefSeq status** Organism Mus musculus Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Lineage Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus Also known as [a]2C; Adra-2c; alpha2C; alpha2-C4 Orthologs human all

GemPharmatech Co., Ltd.

# **Transcript information (Ensembl)**



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

Name 🔺	Transcript ID	bp 🍦	Protein 🖕	Biotype 🍦	CCDS 🍦	UniProt 🖕	Flags			
Adra2c-201	ENSMUST0000049545.6	3445	<u>458aa</u>	Protein coding	<u>CCDS19226</u> &	<u>Q01337</u> &	TSL:NA	GENCODE basic	APPRIS P1	

The strategy is based on the design of Adra2c-201 transcript, The transcription is shown below

3.44 kb

Adra2c-201 > protein coding

#### 江苏集萃药康生物科技股份有限公司

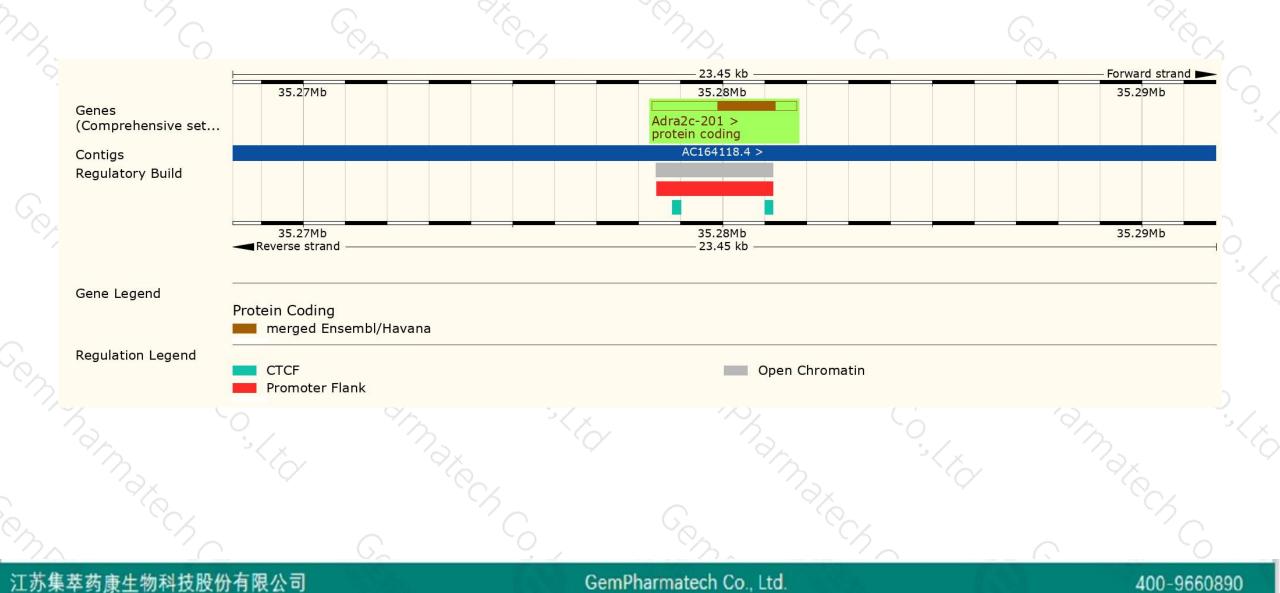
#### GemPharmatech Co., Ltd.

#### 400-9660890

Forward strand

# **Genomic location distribution**



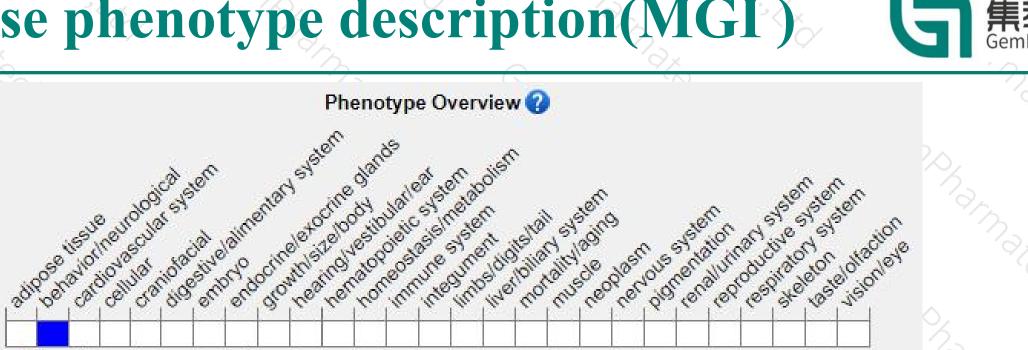


# **Protein domain**



江苏集	萃药康生物科技股	份有限公司			Gen	nPharmatec	h Co., Ltd.				4	100-966089	0
9 75	Č.	2	G.	í G		60/		Č,	$\sim$			"Co	
\ \	Max.	NG/									9×		
	Scale bar	0 4	0 80	120	160	200	240	280	320	360	400	458	
	Variant Legend	missense v	variant		synonymous variant								
$(\mathcal{D})$			1 11	1		l			1 1 1		1	<u> </u>	
	All sequence SNPs/i	Sequence varia	nts (dbSNP and all o	ther sources)		2			201 Mar 14			1.11	
	CDD		cd15323										
	Gene3D		1.20.1070.10				1.20.120.1	0					
		PTHR24248 Alpha 2C a	drenoceptor										2
	PANTHER			G p	protein-coupled r	eceptor, rhodopsin	-like					(	)
	PROSITE patterns		GPCR, rhod	opsin-like, 7TM			10.0						
G	PROSITE profiles			oupled receptor, rhod	opsin-like								
	Pfam	Alpha	2C adrenoceptor	upled recenter when the	anain lika								
		Adrenoceptor family											
	Prints		G protein-coupled	receptor, rhodopsin-l	like								
	SMART		G protein-cou	oled receptor, rhodop	sin-like								
×.	Superfamily		SSF81321										
	MobiDB lite Low complexity (Seg)							_	-				
$\langle \mathcal{A}_{\mathcal{L}} \rangle$	Transmembrane heli				-		-	_		-	-		
	ENSMUSP00000059			(Ox		YO.		YO/				3.	

# **Mouse phenotype description(MGI)**



Click cells to view annotations.

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

Mice homozygous for targeted mutations that inactivate the gene are viable and fertile and appear grossly normal.



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



