

# Ap3s1 Cas9-KO Strategy

Designer: Reviewer:

**Design Date:** 

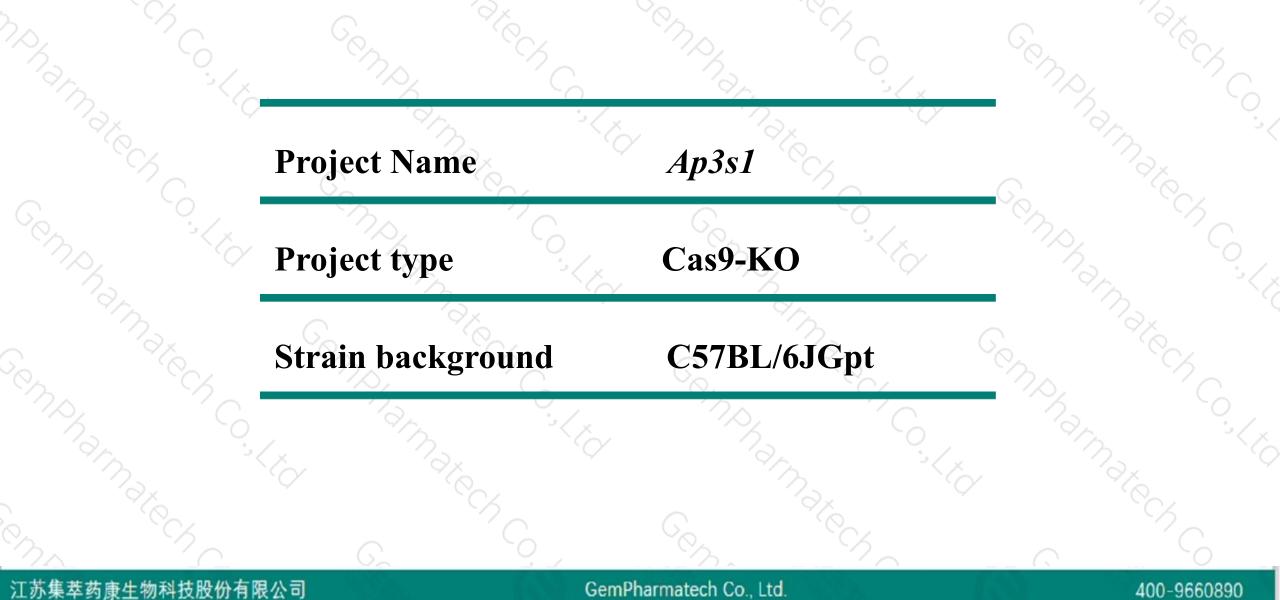
Ruirui Zhang

**Huimin Su** 

2020-4-9

### **Project Overview**

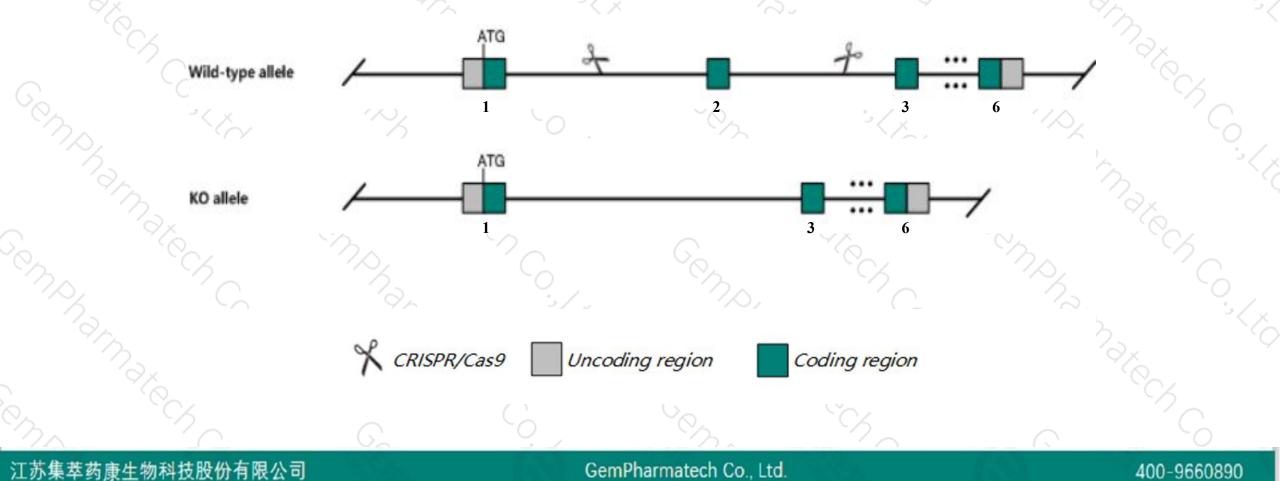




### **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Ap3s1* gene. The schematic diagram is as follows:





- The *Ap3s1* gene has 6 transcripts. According to the structure of *Ap3s1* gene, exon2 of *Ap3s1-201* (ENSMUST00000025357.8) transcript is recommended as the knockout region. The region contains 92bp coding sequence. Knock out the region will result in disruption of protein function.
- > In this project we use CRISPR/Cas9 technology to modify Ap3s1 gene. The brief process is as follows: CRISPR/Cas9 system

- The Ap3s1 gene is located on the Chr18. 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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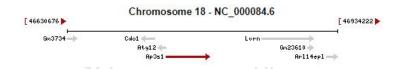
Ap3s1 adaptor-related protein complex 3, sigma 1 subunit [ Mus musculus (house mouse) ]

Gene ID: 11777, updated on 5-Apr-2020

#### Summary

Official SymbolAp3s1 provided by MGIOfficial Full Nameadaptor-related protein complex 3, sigma 1 subunit provided by MGIPrimary sourceMGI:MGI:1337062See relatedEnsembl:ENSMUSG0000024480Gene typeprotein codingRefSeq statusREVIEWEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae;<br/>Murinae; Mus; MusSummaryThis gene encodes the sigma subunit of the heterotetrameric adaptor protein complex AP-3 which is involved in the formation of specialized lysosome-related<br/>compartments such as melanosomes. Alternate splicing of this gene results in multiple transcript variants encoding different isoforms. Pseudogenes of this gene<br/>are found on chromosomes 1, 8, 16, 17 and X. [provided by RefSeq, Dec 2014]ExpressionUbiquitous expression in CNS E18 (RPKM 12.6), CNS E14 (RPKM 11.2) and 27 other tissues See more<br/>OrthologOrthologshuman all

Genomic context				8 ?
Location: 18; 18 C				See Ap3s1 in Genome Data Viewer
Exon count: 7 Annotation release	Status	Assembly	Chr	Location
<u>108</u> Build 37.2	current previous assembly	GRCm38.p6 ( <u>GCF_000001635.26</u> ) MGSCv37 ( <u>GCF_000001635.18</u> )	18 18	NC_000084.6 (4674169446790826) NC_000084.5 (4690157146950480)



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## **Transcript information (Ensembl)**



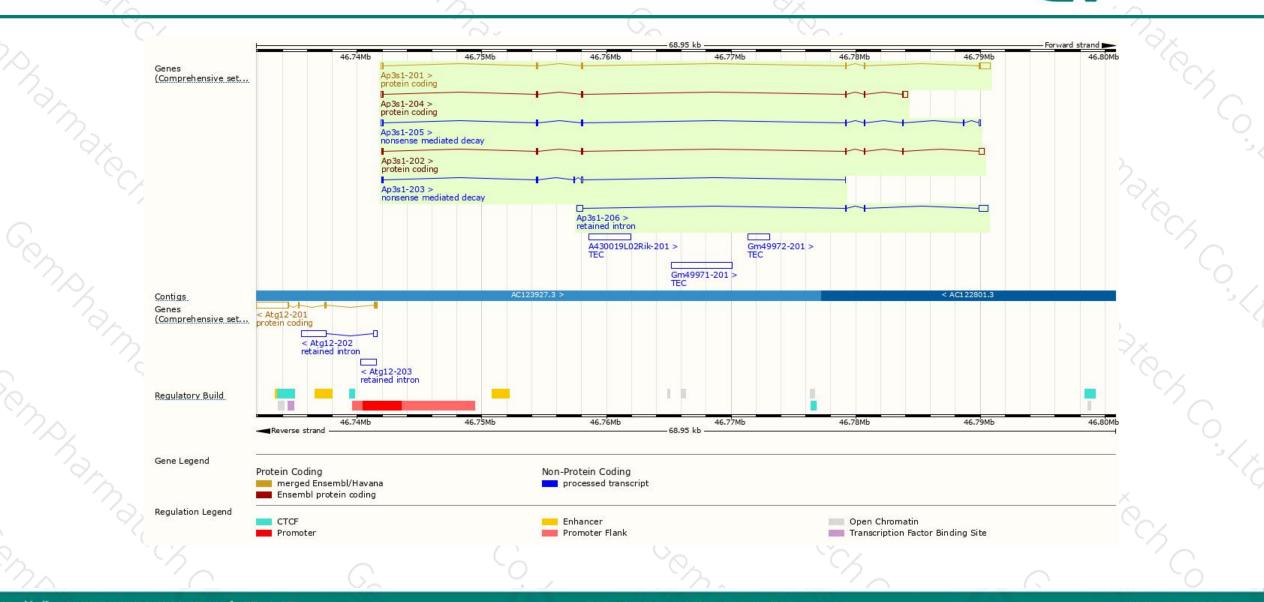
### The gene has 6 transcripts, all transcripts are shown below:

Name 🖕	Transcript ID 💧 bp		Protein 🖕	Biotype	CCDS 🖕	UniProt 🖕	Flags	
Ap3s1-201	ENSMUST0000025357.8	1520	<u>193aa</u>	Protein coding	<u>CCDS29236</u> &	<u>Q3U8S0</u> & <u>Q9DCR2</u> മ	TSL:1 GENCODE basic APPRIS P1	
Ap3s1-202	ENSMUST00000224622.1	978	<u>162aa</u>	Protein coding	=	<u>A0A286YDU3</u> 교	GENCODE basic	
Ap3s1-204	ENSMUST00000225520.1	957	<u>162aa</u>	Protein coding	-	A0A286YDU3@	GENCODE basic	
Ap3s1-205	ENSMUST00000226108.1	743	<u>162aa</u>	Nonsense mediated decay	=	A0A286YDU3		
Ap3s1-203	ENSMUST00000225415.1	417	<u>54aa</u>	Nonsense mediated decay	=	A0A286YD58	5.	
Ap3s1-206	ENSMUST0000234998.1	1407	No protein	Retained intron	×	2 <b>7</b> 3		

### The strategy is based on the design of Ap3s1-201 transcript, the transcription is shown below

Ap31-201 > protein coding				48.95 kb		Forward strand
	Ap3s1-201 > protein coding	6	- 9 pr	is/x	12 BL	73.
	arma.		Marco Co		narm.	Mar.
	7 C C	5		200		

### **Genomic location distribution**



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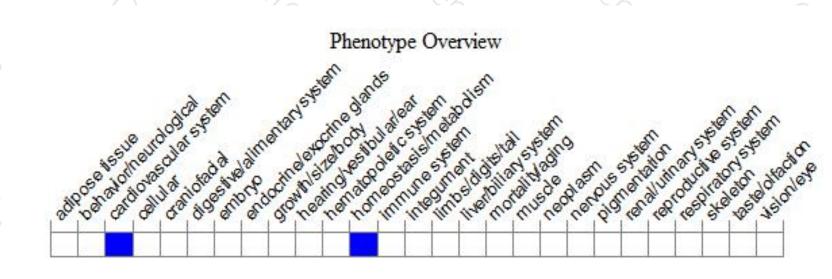
### **Protein domain**



3.	-3 ( ) -3 ( )	$   \left( \begin{array}{cccccccccccccccccccccccccccccccccccc$	No.
<ul> <li>S</li> </ul>	ENSMUSP00000025 Superfamily	Longin-like domain superfamily	
F	Pfam PROSITE patterns PANTHER	AP complex, mu/sigma subunit Clathrin adaptor complex, small chain	
	Gene3D	Adaptor protein complex, sigma subunit PTHR11753 :SF12 3.30.450.60	
	CDD	cd14834	0
5. <b>*</b>	All sequence SNPs/i	Sequence variants (dbSNP and all other sources)	24
~/ ``	/ariant Legend	missense variant synonymous variant	
5	Scale bar	0 20 40 60 80 100 120 140 160 193	
	°° A Q		6.
3	No <sup>25</sup>	in the second of	

## Mouse phenotype description(MGI)





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



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



