

Asap1 Cas9-CKO Strategy

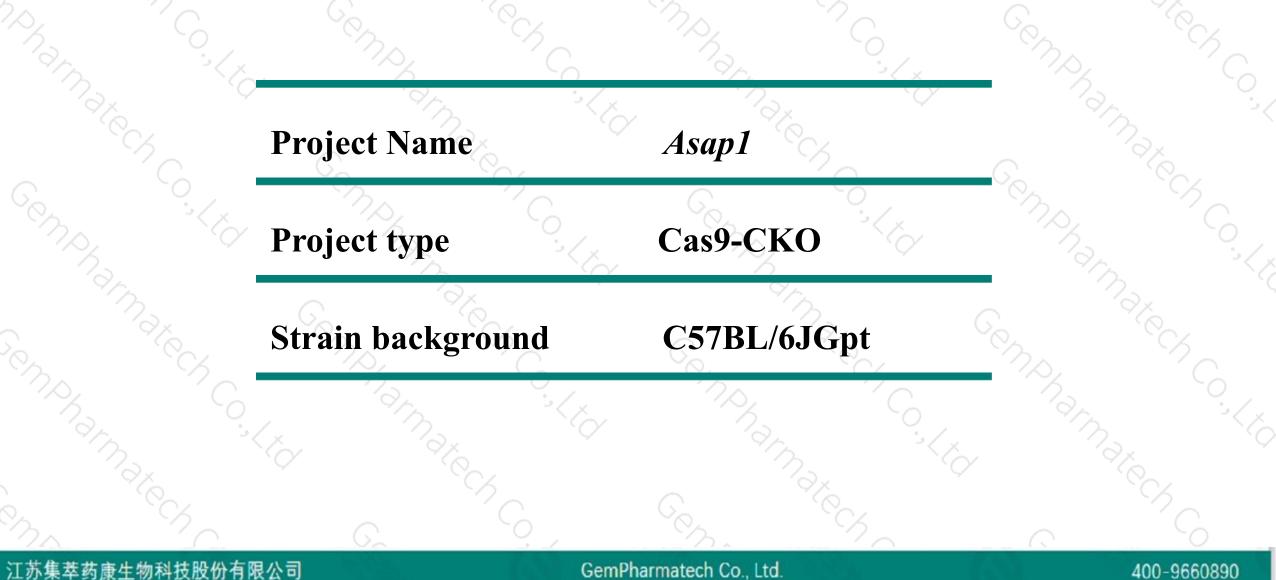
Designer: Reviewer:

Design Date:

Huan Wang Huan Fan 2020-3-6

Project Overview





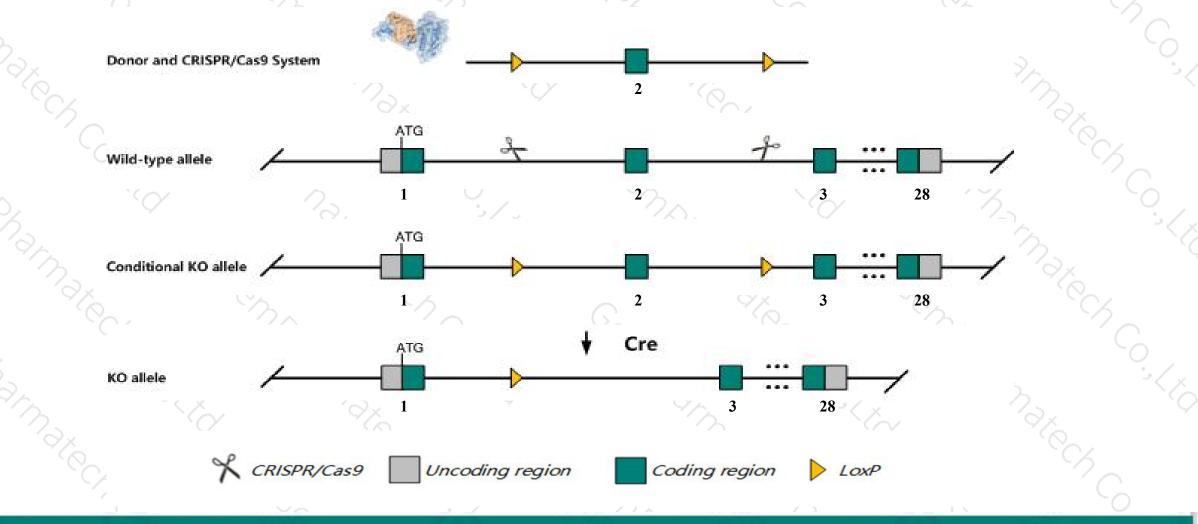
GemPharmatech Co., Ltd.

Conditional Knockout strategy



400-9660890

This model will use CRISPR/Cas9 technology to edit the *Asap1* gene. The schematic diagram is as follows:



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 The Asap1 gene has 19 transcripts. According to the structure of Asap1 gene, exon2 of Asap1-217 (ENSMUST00000177371.7) transcript is recommended as the knockout region. The region contains 73bp coding sequence. Knock out the region will result in disruption of protein function.

In this project we use CRISPR/Cas9 technology to modify *Asap1* 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.



- The Asap1 gene is located on the Chr15. 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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Asap1 ArfGAP with SH3 domain, ankyrin repeat and PH domain1 [Mus musculus (house mouse)]

Gene ID: 13196, updated on 7-Apr-2019

Summary

Official Symbol	Asap1 provided by MGI					
Official Full Name	e ArfGAP with SH3 domain, ankyrin repeat and PH domain1 provided by MGI					
Primary source	MGI:MGI:1342335					
See related	Ensembl:ENSMUSG0000022377					
Gene type	protein coding					
RefSeq status	VALIDATED					
Organism	Mus musculus					
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;					
	Muroidea; Muridae; Murinae; Mus; Mus					
Also known as	AV239055, DEF-1, Ddef1, PAP, mKIAA1249, s19					
Expression	Ubiquitous expression in testis adult (RPKM 21.3), cortex adult (RPKM 15.3) and 28 other tissues See more					
Orthologs	human all					

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Transcript information (Ensembl)



The gene has 19 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Asap1-217	ENSMUST00000177371.7	6164	<u>1124aa</u>	Protein coding	CCDS70631	H3BL41	TSL:1 GENCODE basic APPRIS ALT2
Asap1-218	ENSMUST00000177374.7	4582	<u>1147aa</u>	Protein coding	CCDS56986		TSL:1 GENCODE basic APPRIS P3
Asap1-216	ENSMUST00000177083.7	4415	<u>1112aa</u>	Protein coding	CCDS70632	E9QMJ1	TSL:1 GENCODE basic APPRIS ALT2
Asap1-206	ENSMUST00000175799.7	4237	<u>1087aa</u>	Protein coding	CCDS70633	H3BKE6	TSL:5 GENCODE basic APPRIS ALT2
Asap1-210	ENSMUST00000176384.7	4149	<u>1090aa</u>	Protein coding	CCDS70634	Q9QWY8	TSL:1 GENCODE basic APPRIS ALT2
Asap1-201	ENSMUST00000023008.15	4580	<u>1147aa</u>	Protein coding	₹3	E9QN63	TSL:5 GENCODE basic APPRIS ALT2
Asap1-203	ENSMUST00000110115.8	4535	<u>1132aa</u>	Protein coding	2 5		TSL:5 GENCODE basic APPRIS ALT2
Asap1-202	ENSMUST00000110114.9	4409	<u>1090aa</u>	Protein coding	20	E9QMI7	TSL:5 GENCODE basic APPRIS ALT2
Asap1-205	ENSMUST00000175793.7	4314	<u>1135aa</u>	Protein coding	50		TSL:5 GENCODE basic APPRIS ALT2
Asap1-208	ENSMUST00000176014.7	3435	<u>1144aa</u>	Protein coding	•	H3BJY2	TSL:5 GENCODE basic APPRIS ALT2
Asap1-215	ENSMUST00000177035.7	3228	<u>1075aa</u>	Protein coding	25	H3BKD4	TSL:5 GENCODE basic APPRIS ALT2
Asap1-209	ENSMUST00000176358.1	631	<u>211aa</u>	Protein coding	20	<u>H3BK40</u>	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Asap1-207	ENSMUST00000175963.1	466	<u>98aa</u>	Protein coding	74	H3BJM7	CDS 3' incomplete TSL:2
Asap1-204	ENSMUST00000175710.7	985	No protein	Retained intron	-1	100	TSL1
Asap1-214	ENSMUST00000176909.1	474	No protein	Retained intron	-1	0.20	TSL:3
Asap1-219	ENSMUST00000177475.1	339	No protein	Retained intron	20	1021	TSL:3
Asap1-213	ENSMUST00000176821.7	396	No protein	IncRNA	54	85	TSL:5
Asap1-211	ENSMUST00000176427.7	339	No protein	IncRNA	₹3	390	TSL:5
Asap1-212	ENSMUST00000176516.1	238	No protein	IncRNA	48	820	TSL:3
	19				1	1	

The strategy is based on the design of *Asap1-217* transcript, The transcription is shown below

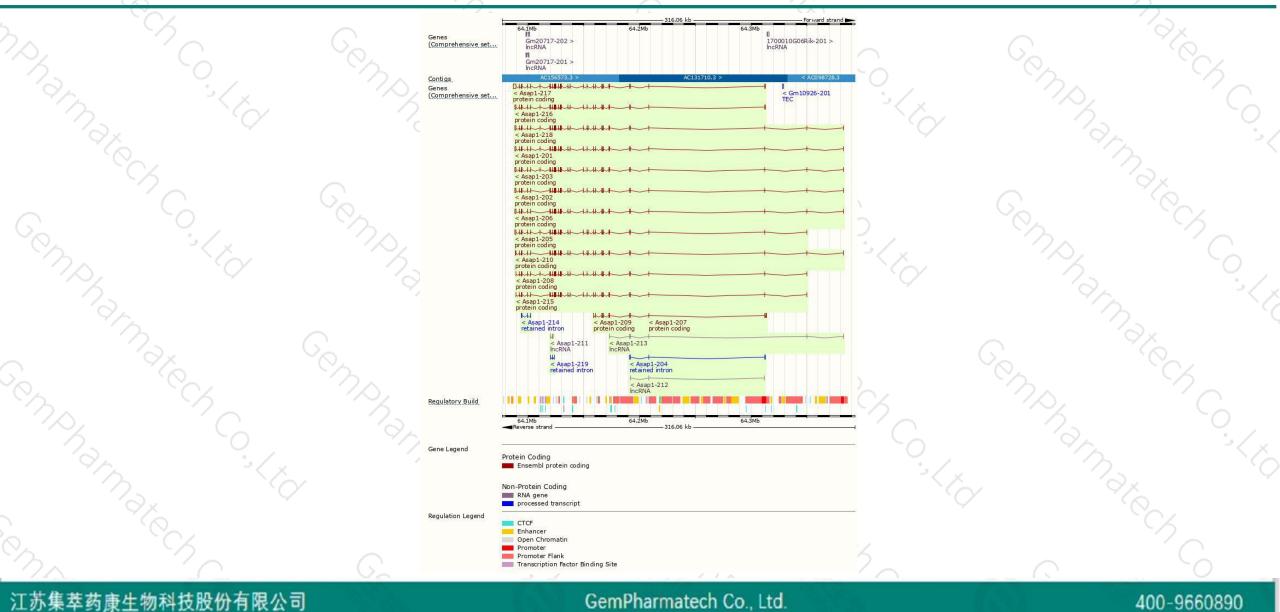
< Asap1-217 protein coding

Reverse strand -

- 225.82 kb -

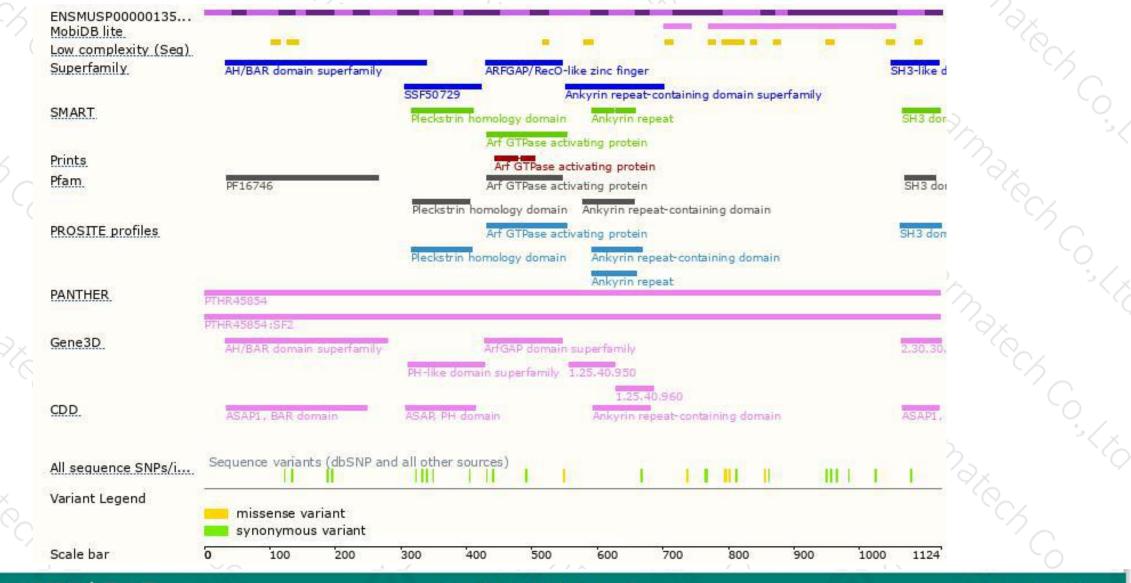
Genomic location distribution





Protein domain





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



