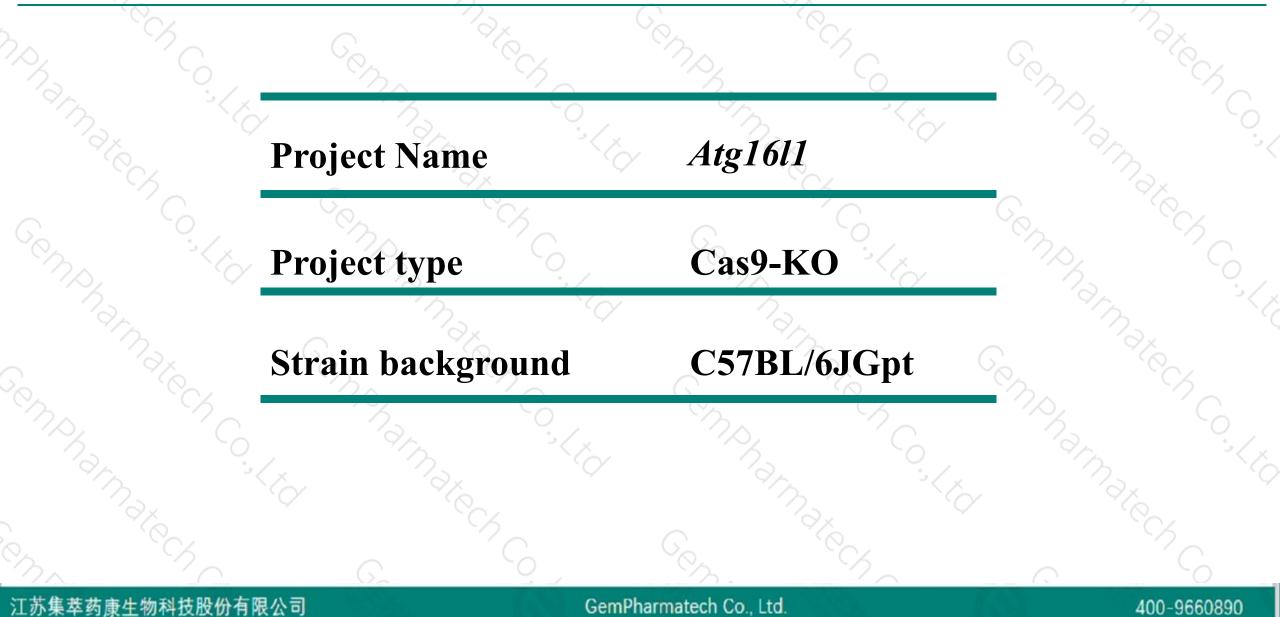
Atg16l1 Cas9-KO Strategy

Designer: Design Date: Jinling Wang 2019-7-24

Project Overview

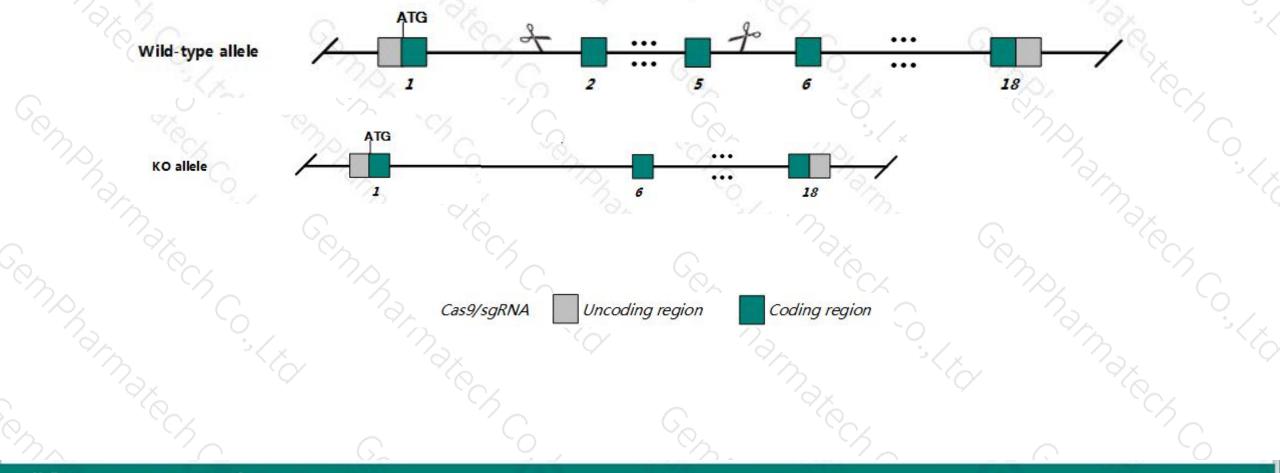




Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Atg16l1* gene. The schematic diagram is as follows:



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- The *Atg1611* gene has 11 transcript. According to the structure of *Atg1611* gene, exon2-5 of *Atg1611*-201 (ENSMUST00000027512.12) transcript is recommended as the knockout region. The region contains 526bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Atg16l1* 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.



- Null homozygotes have a cellular defect in autophagy that results in lethality during the neonatal starvation period. Mice homozygous for hypomorphic alleles have Paneth cells with aberrant, disorganized granules similar to those found in patients with Crohn's disease.
- The *Atg16l1* 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)



Atg16l1 autophagy related 16-like 1 (S. cerevisiae) [Mus musculus (house mouse)]

Gene ID: 77040, updated on 8-Dec-2018

Summary

Official Symbol Atg1611 provided by MGI Official Full Name autophagy related 16-like 1 (S. cerevisiae) provided by MGI Primary source MGI:MGI:1924290 Ensembl:ENSMUSG0000026289 See related 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 WDR30; Apg16I; Atg16I; 1500009K01Rik Ubiguitous expression in CNS E18 (RPKM 18.9), CNS E14 (RPKM 16.2) and 28 other tissues See more Expression Orthologs human all

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Transcript information (Ensembl)



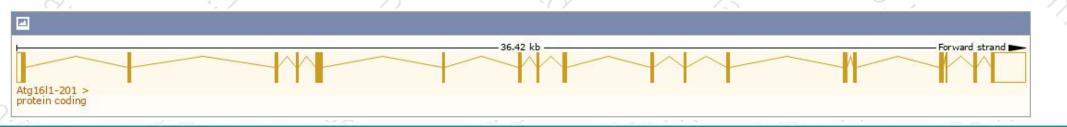
400-9660890

The gene has 11 transcripts, and all transcripts are shown below:

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Name 🖕	Transcript ID 👙	bp 🖕	Protein 🖕	Biotype 🍦	CCDS 🍦	UniProt 🖕	RefSeq 🖕	Flags		
Atg1611-201	ENSMUST0000027512.12	3130	<u>607aa</u>	Protein coding	<u>CCDS15136</u> &	Q8C0J2	<u>NM_029846</u> & NP_084122 &	TSL:1 GENCODE basic APPRIS P3		
Atg1611-203	ENSMUST00000113190.2	3102	<u>623aa</u>	Protein coding	<u>CCDS56637</u> &	<u>G9M4M6</u> & <u>Q8C0J2</u> &	NM_001205391& NP_001192320&	TSL:1 GENCODE basic APPRIS ALT1		
Atg1611-202	ENSMUST00000113186.7	3004	<u>588aa</u>	Protein coding	<u>CCDS56638</u> &	<u>Q3TDQ5</u> & <u>Q8C0J2</u> &	NM_001205392& NP_001192321&	TSL:1 GENCODE basic APPRIS ALT1		
Atg1611-210	ENSMUST00000144047.7	749	<u>154aa</u>	Protein coding	28	<u>D3YZW7</u> &		CDS 3' incomplete TSL:5		
Atg1611-206	ENSMUST00000133072.7	3751	No protein	Retained intron	23	2	121	TSL:2		
Atg1611-204	ENSMUST00000127884.1	3400	No protein	Retained intron	23	2		TSL:2		
Atg1611-211	ENSMUST00000151037.7	<u>3344</u>	No protein	Retained intron		2 2	12	TSL:2		
Atg1611-207	ENSMUST00000134603.1	1674	No protein	Retained intron		2	127	TSL:2		
Atg1611-208	ENSMUST00000137638.7	<mark>1</mark> 594	No protein	Retained intron		2	1.27	TSL:5		
Atg1611-205	ENSMUST00000129431.1	640	No protein	Retained intron	2	2		TSL:3		
Atg1611-209	ENSMUST00000139628.1	332	No protein	Retained intron	<u>1</u>	2	122	TSL:3		

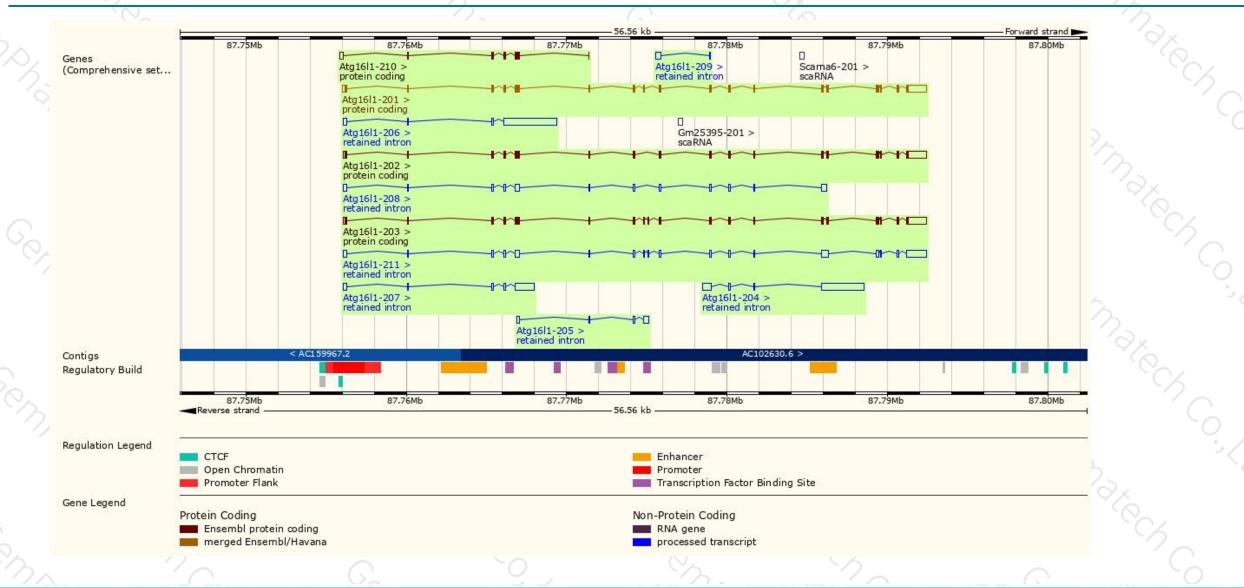
The strategy is based on the design of Atg1611-201 transcript, The transcription is shown below



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Genomic location distribution



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Protein domain



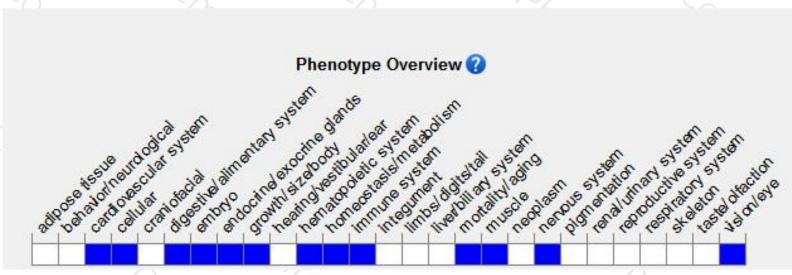
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	hmmpanther	PTHR19878 PTHR19878:SF6									
	Superfamily domains					WD40-repeat-containing of	domain superfamily				-~
	SMART domains					WD40 repeat		-		-	>
	Prints domain						a WD-40 repeat				
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	PROSITE profiles					WD40-repeat-containin	ng domain				
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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/).

Null homozygotes have a cellular defect in autophagy that results in lethality during the neonatal starvation period. Mice homozygous for hypomorphic alleles have Paneth cells with aberrant, disorganized granules similar to those found in patients with Crohn's disease.

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



