Kcnma1 Cas9-KO Strategy

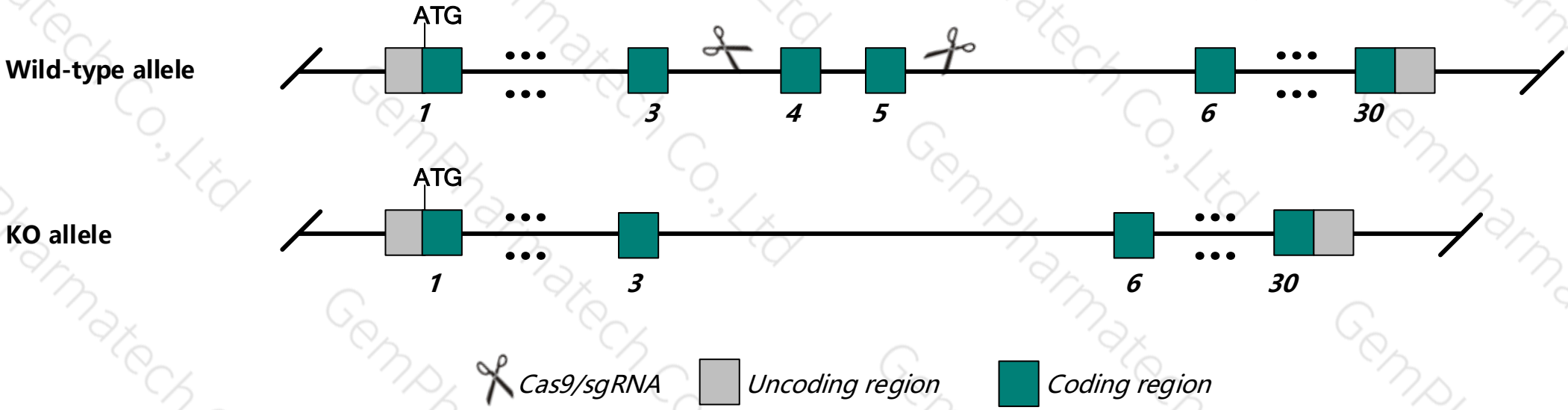
Designer: Ruirui Zhang

Project Overview

Project Name	***Kcnma1***
Project type	**Cas9-KO**
Strain background	**C57BL/6J**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Kcnma1* gene. The schematic diagram is as follows:



Technical routes

- The *Kcnma1* gene has 38 transcripts. According to the structure of *Kcnma1* gene, exon4-exon5 of *Kcnma1-212* (ENSMUST00000188285.6) transcript is recommended as the knockout region.The region contains 206bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Kcnma1* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6J 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/6J mice.

Notice

➢ According to the existing MGI data,Homozygous inactivation of this gene leads to cerebellar ataxia, Purkinje cell dysfunction, uneven gait patterns, bladder hyperactivity, urinary incontinence, abnormal colonic K+ secretion, and hearing impairment.

➢ The *Kcnma1* gene is located on the Chr14. 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 gene transcription and translation processes, all risks cannot be predicted under existing information.

Gene information（NCBI）

集萃药康 GemPharmatech

Kcnma1 potassium large conductance calcium-activated channel, subfamily M, alpha member 1 [Mus musculus (house mouse)]

Gene ID: 16531, updated on 31-Jan-2019

Summary

Official Symbol	Kcnma1 provided by MGI
Official Full Name	potassium large conductance calcium-activated channel, subfamily M, alpha member 1 provided by MGI
Primary source	MGI:MGI:99923
See related	Ensembl:ENSMUSG00000063142
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	5730414M22Rik, BKCa, MaxiK, Slo, Slo1, mSlo, mSlo1
Expression	Biased expression in cortex adult (RPKM 10.4), frontal lobe adult (RPKM 9.3) and 12 other tissues See more
Orthologs	human all

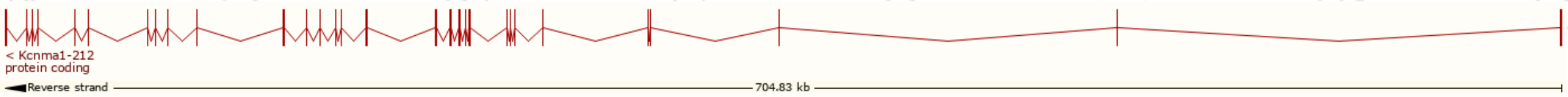
Transcript information（Ensembl）

The gene has 38 transcripts,all transcripts are shown below:

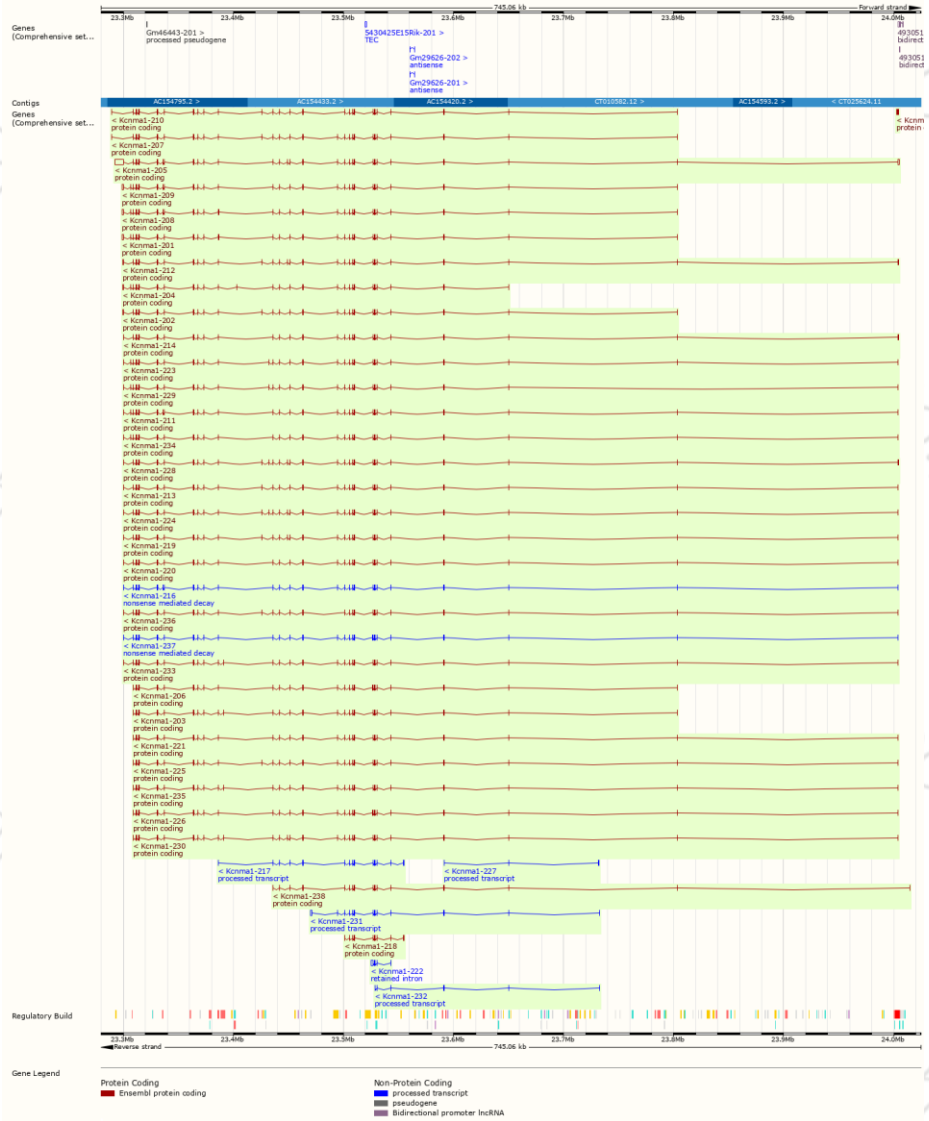
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Kcnma1-201	ENSMUST00000065788.14	4524	1108aa	Protein coding	-	F6ZSN2	TSL:5 GENCODE basic
Kcnma1-202	ENSMUST00000074983.12	3974	1107aa	Protein coding	-	F6XW53	TSL:5 GENCODE basic APPRIS ALT2
Kcnma1-203	ENSMUST00000100831.10	3261	1086aa	Protein coding	-	F6V0P5	TSL:5 GENCODE basic
Kcnma1-204	ENSMUST00000112423.9	3664	1055aa	Protein coding	-	F7BHL0	TSL:5 GENCODE basic
Kcnma1-205	ENSMUST00000145596.2	12094	1209aa	Protein coding	CCDS79277	A0A286YD35	TSL:NA GENCODE basic APPRIS ALT2
Kcnma1-206	ENSMUST00000163322.2	3162	1053aa	Protein coding	-	E9Q9P4	TSL:5 GENCODE basic
Kcnma1-207	ENSMUST00000172099.8	3354	1118aa	Protein coding	-	F6WSZ3	TSL:5 GENCODE basic APPRIS ALT2
Kcnma1-208	ENSMUST00000177634.7	4605	1135aa	Protein coding	-	J3QMT8	TSL:5 GENCODE basic
Kcnma1-209	ENSMUST00000179097.7	4596	1132aa	Protein coding	-	J3QN27	TSL:5 GENCODE basic
Kcnma1-210	ENSMUST00000179836.7	3177	1059aa	Protein coding	-	J3QP84	TSL:5 GENCODE basic
Kcnma1-211	ENSMUST00000188210.7	3579	1192aa	Protein coding	-	E3VRY9	CDS 5' incomplete TSL:1
Kcnma1-212	ENSMUST00000188285.6	4440	1239aa	Protein coding	CCDS79278	A0A087WQN5	TSL:1 GENCODE basic APPRIS P4
Kcnma1-213	ENSMUST00000188991.6	3711	1236aa	Protein coding	CCDS79276	A0A087WRS4	TSL:1 GENCODE basic APPRIS ALT2
Kcnma1-214	ENSMUST00000190044.6	3747	1178aa	Protein coding	CCDS79275	A0A087WQ41	TSL:1 GENCODE basic APPRIS ALT2
Kcnma1-215	ENSMUST00000190339.1	890	191aa	Protein coding	-	A0A087WSQ4	TSL:5 GENCODE basic
Kcnma1-216	ENSMUST00000190985.7	3429	634aa	Nonsense mediated decay	-	A0A087WQE1	TSL:1
Kcnma1-217	ENSMUST00000212542.1	1600	No protein	Processed transcript	-	-	TSL:1
Kcnma1-218	ENSMUST00000212576.1	1217	332aa	Protein coding	-	A0A1D5RMA3	CDS 3' incomplete TSL:5
Kcnma1-219	ENSMUST00000223655.1	3528	1175aa	Protein coding	-	A0A286YDC1	CDS 5' incomplete
Kcnma1-220	ENSMUST00000223727.1	3339	1112aa	Protein coding	-	A0A286YCW3	CDS 5' incomplete
Kcnma1-221	ENSMUST00000223749.1	3342	1113aa	Protein coding	-	E3VRZ4	CDS 5' incomplete
Kcnma1-222	ENSMUST00000223847.1	2133	No protein	Retained intron	-	-	-
Kcnma1-223	ENSMUST00000224025.1	3557	1185aa	Protein coding	-	A0A286YCP1	CDS 5' incomplete
Kcnma1-224	ENSMUST00000224077.1	3537	1178aa	Protein coding	-	A0A286YCZ9	GENCODE basic APPRIS ALT2
Kcnma1-225	ENSMUST00000224232.1	3516	1171aa	Protein coding	-	A0A286YD59	CDS 5' incomplete
Kcnma1-226	ENSMUST00000224285.1	3342	1113aa	Protein coding	-	E3VRY6	CDS 5' incomplete
Kcnma1-227	ENSMUST00000224465.1	447	No protein	Processed transcript	-	-	-
Kcnma1-228	ENSMUST00000224468.1	3753	1242aa	Protein coding	-	A0A286YDM6	GENCODE basic APPRIS ALT2
Kcnma1-229	ENSMUST00000224787.1	3348	1115aa	Protein coding	-	A0A286YCI9	CDS 5' incomplete
Kcnma1-230	ENSMUST00000224812.1	3441	1146aa	Protein coding	-	A0A286YDR7	CDS 5' incomplete
Kcnma1-231	ENSMUST00000224933.1	2705	No protein	Processed transcript	-	-	-
Kcnma1-232	ENSMUST00000225305.1	732	No protein	Processed transcript	-	-	-
Kcnma1-233	ENSMUST00000225315.1	3429	1142aa	Protein coding	-	A0A286YDU9	CDS 5' incomplete
Kcnma1-234	ENSMUST00000225431.1	3498	1165aa	Protein coding	-	A0A286YE30	CDS 5' incomplete
Kcnma1-235	ENSMUST00000225471.1	3429	1142aa	Protein coding	-	A0A286YCQ2	CDS 5' incomplete
Kcnma1-236	ENSMUST00000225556.1	3360	1119aa	Protein coding	-	A0A286YCF5	CDS 5' incomplete
Kcnma1-237	ENSMUST00000225794.1	3434	357aa	Nonsense mediated decay	-	A0A286YD36	CDS 5' incomplete
Kcnma1-238	ENSMUST00000226051.1	1641	477aa	Protein coding	-	A0A286YDB5	CDS 3' incomplete

Transcript information（Ensembl）

The strategy is based on the design of *Kcnma1-212* transcript, the transcription is shown below:

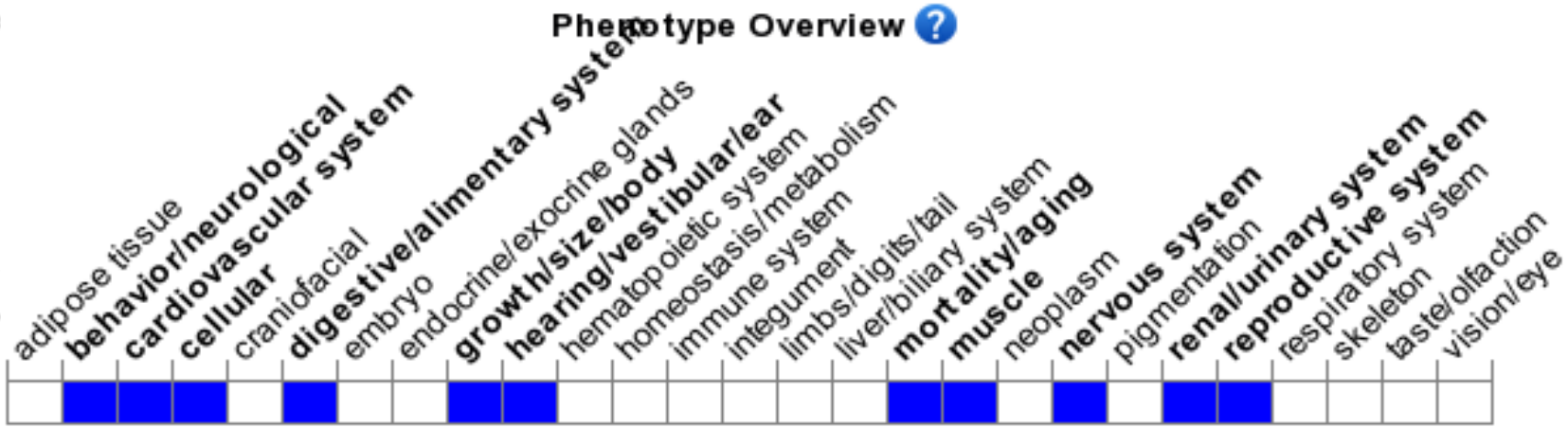


Genomic location distribution



Protein domain

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,homozygous inactivation of this gene leads to cerebellar ataxia, Purkinje cell dysfunction, uneven gait patterns, bladder hyperactivity, urinary incontinence, abnormal colonic K+ secretion, and hearing impairment.

集萃药康
GemPharmatech

If you have any questions, you are welcome to inquire.

Tel: 025-5864 1534

集萃药康生物科技
GemPharmatech Co.,Ltd