

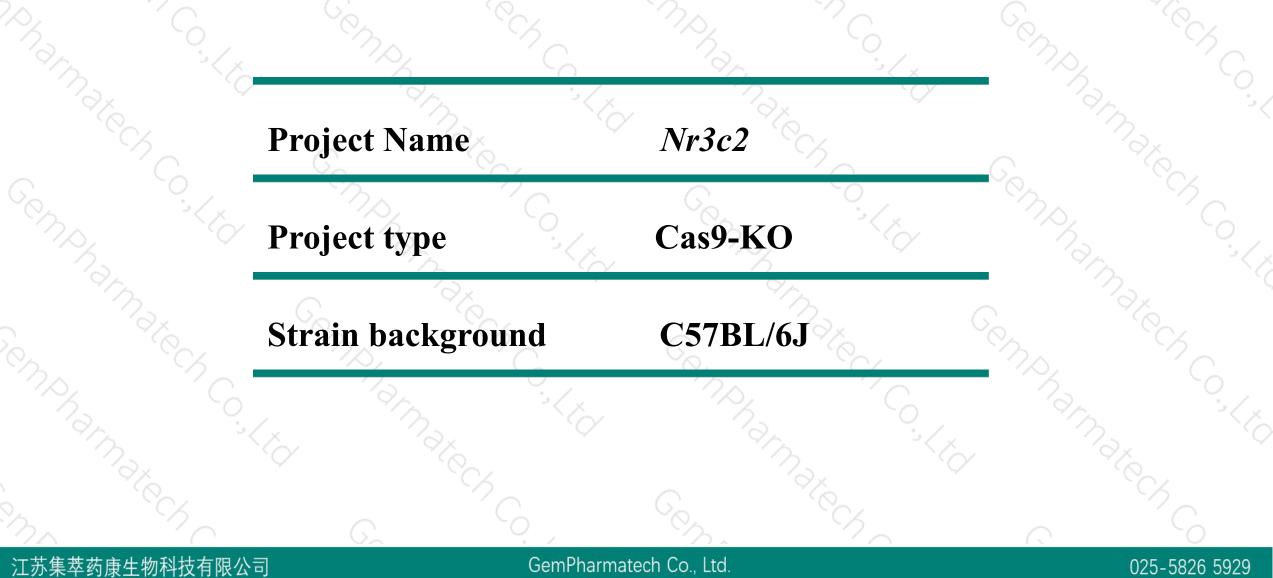
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Project Overview





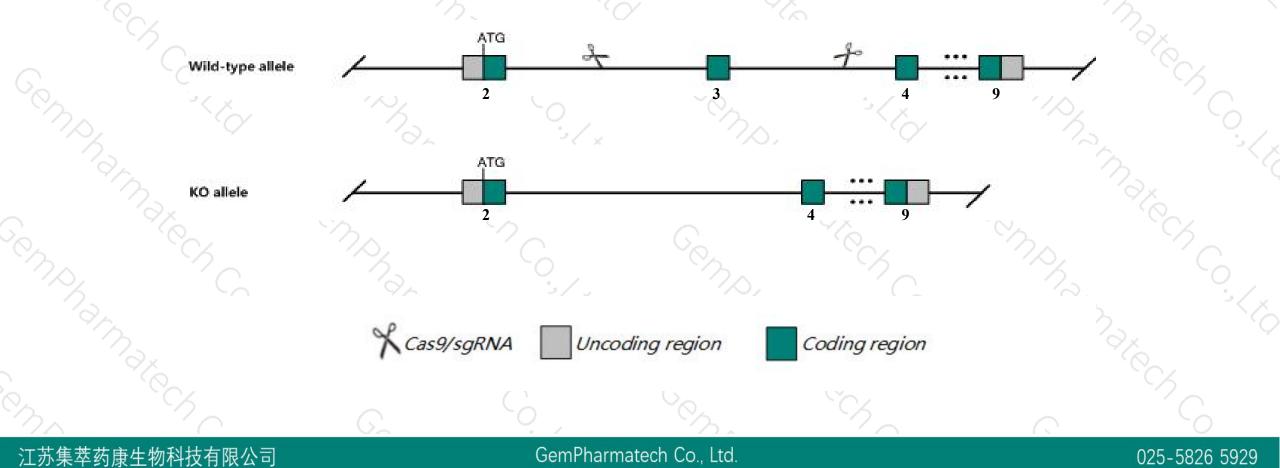
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Knockout strategy



This model will use CRISPR/Cas9 technology to edit the Nr3c2 gene. The schematic diagram is as follows:





- The Nr3c2 gene has 8 transcripts. According to the structure of Nr3c2 gene, exon3 of Nr3c2-203 (ENSMUST00000109912.7) transcript is recommended as the knockout region. The region contains 140bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify Nr3c2 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.



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- According to the existing MGI data, Mice homozygous for a targeted null mutation exhibit weight loss and symptoms of pseudohypoaldosteronism, and eventually die at around day 10 after birth from renal salt wasting and dehydration.
- ➤ Transcript *Nr3c2-206,207* may be unaffected.
- The Nr3c2 gene is located on the Chr8. 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.

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Gene information (NCBI)



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Nr3c2 nuclear receptor subfamily 3, group C, member 2 [Mus musculus (house mouse)]

Gene ID: 110784, updated on 19-Mar-2019

Summary

Official Symbol	Nr3c2 provided by MGI
Official Full Name	nuclear receptor subfamily 3, group C, member 2 provided by MGI
Primary source	MGI:MGI:99459
See related	Ensembl:ENSMUSG0000031618
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	MR, Mir
Expression	Broad expression in colon adult (RPKM 12.0), frontal lobe adult (RPKM 3.4) and 20 other tissues See more
Orthologs	human all

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Transcript information (Ensembl)



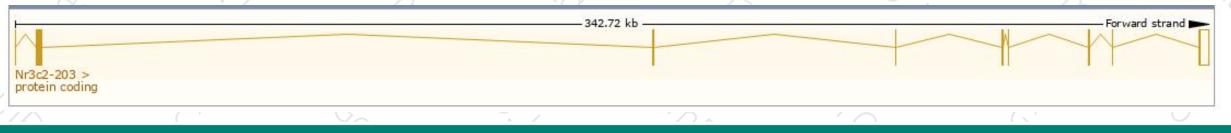
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The gene has 8 transcripts, all transcripts are shown below:

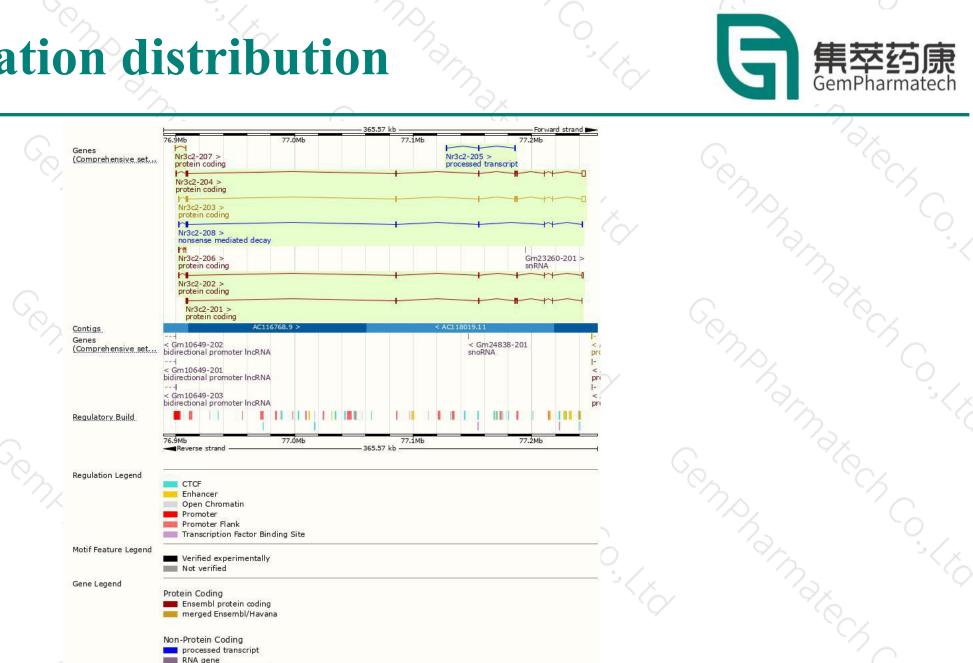
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Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nr3c2-204	ENSMUST00000109913.8	5916	<u>980aa</u>	Protein coding	CCDS40393	A3KN90	TSL:5 GENCODE basic APPRIS P2
Nr3c2-203	ENSMUST00000109912.7	5675	<u>980aa</u>	Protein coding	CCDS40393	<u>A3KN90</u>	TSL:1 GENCODE basic APPRIS P2
Nr3c2-202	ENSMUST00000109911.7	3804	<u>867aa</u>	Protein coding	19450	D3Z473	TSL:5 GENCODE basic
Nr3c2-201	ENSMUST0000034031.5	2955	<u>984aa</u>	Protein coding	14 <u>1</u> 23	<u>E9Q8M8</u>	TSL:5 GENCODE basic APPRIS ALT2
Nr3c2-206	ENSMUST00000128862.1	612	<u>128aa</u>	Protein coding	1753	D3Z7F2	CDS 3' incomplete TSL:2
Nr3c2-207	ENSMUST00000143284.1	270	<u>47aa</u>	Protein coding	(1 7)	<u>D3Z7J4</u>	CDS 3' incomplete TSL:5
Nr3c2-208	ENSMUST00000148106.7	3180	<u>698aa</u>	Nonsense mediated decay	(<u>1</u> 2)	D6RIL1	TSL:5
Nr3c2-205	2-205 ENSMUST00000126697.1 350 No protein		Processed transcript	1920	12	TSL:2	

The strategy is based on the design of Nr3c2-203 transcript, The transcription is shown below



Genomic location distribution



Bidirectional promoter IncRNA

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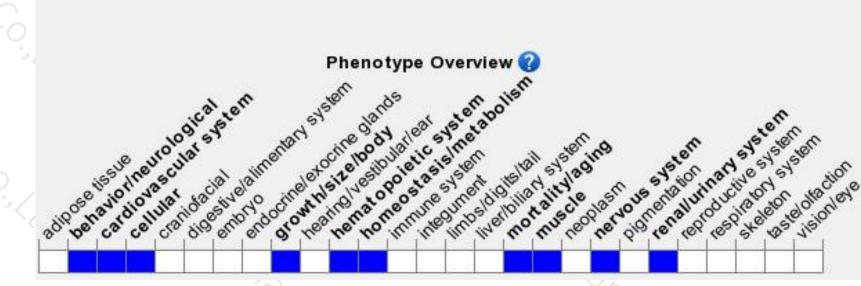
Protein domain



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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/).

According to the existing MGI data, Mice homozygous for a targeted null mutation exhibit weight loss and symptoms of pseudohypoaldosteronism, and eventually die at around day 10 after birth from renal salt wasting and dehydration.

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If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



