Map3k20 Cas9-CKO Strategy

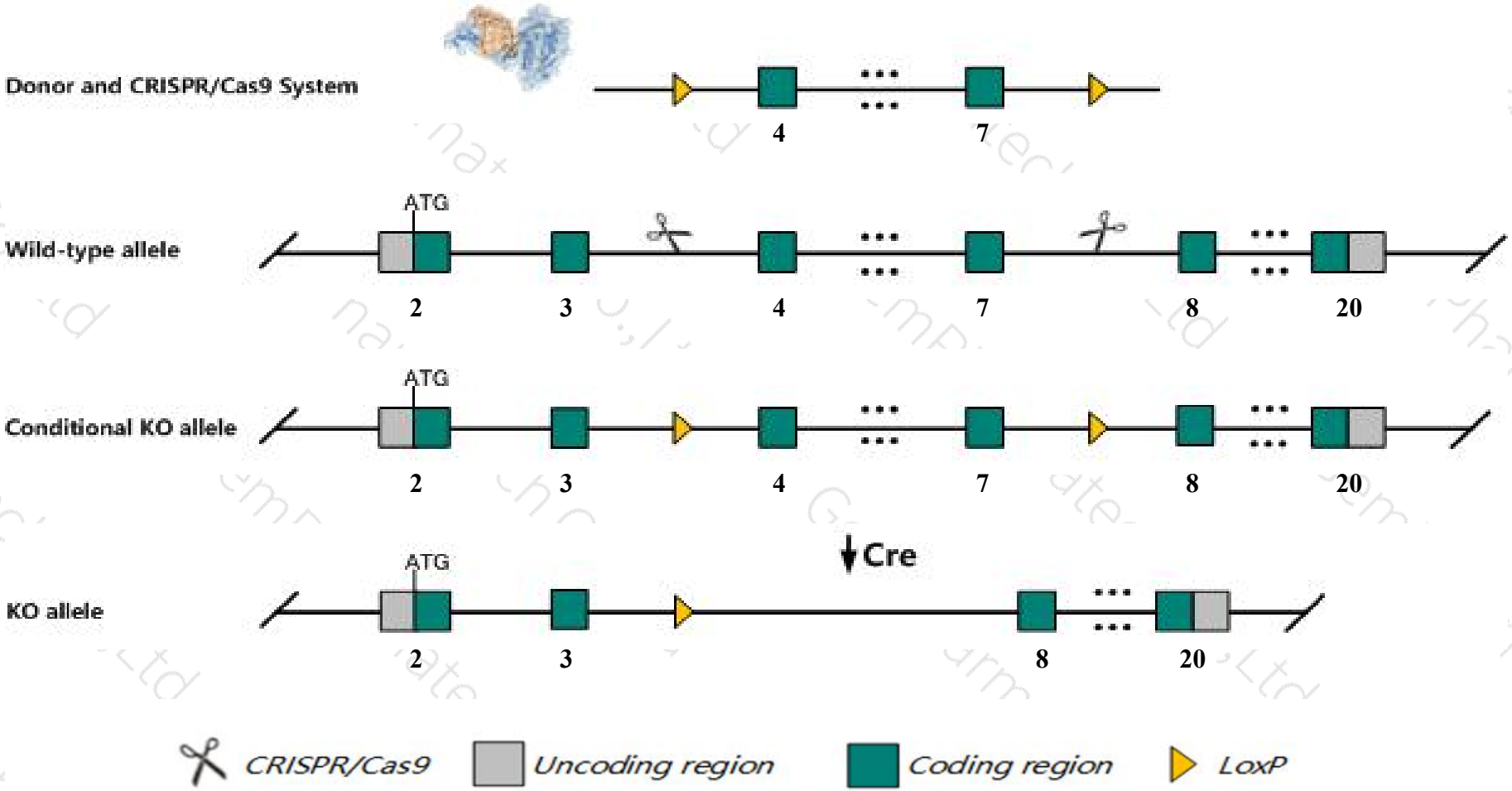
Designer: Huan Wang
Reviewer: Huan Fan
Design Date: 2020-5-15

Project Overview

Project Name	***Map3k20***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Map3k20* gene. The schematic diagram is as follows:



Technical routes

- The *Map3k20* gene has 6 transcripts. According to the structure of *Map3k20* gene, exon4-exon7 of *Map3k20-201* (ENSMUST00000090824.11) transcript is recommended as the knockout region.The region contains 335bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Map3k20* 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.

Notice

- According to the existing MGI data,mice homozygous for a knock-out allele exhibit complete lethality at e9.5 with growth retardation. mice homozygous for an allele lacking the sam domain exhibit low penetrant unilateral complex hindlimb duplication phenotype.
- The *Map3k20* gene is located on the Chr2. 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）

集萃药康 GemPharmatech

Map3k20 mitogen-activated protein kinase kinase kinase 20 [Mus musculus (house mouse)]

Gene ID: 65964, updated on 20-Mar-2020

Summary

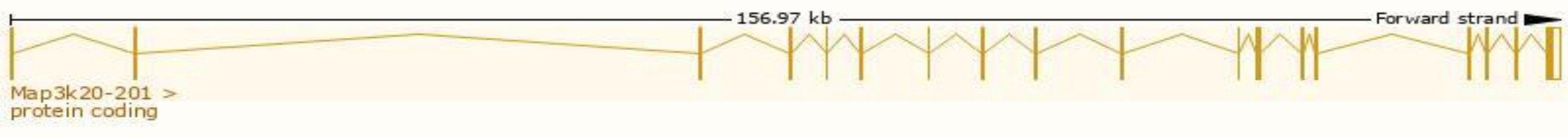
Official Symbol	Map3k20 provided by MGI
Official Full Name	mitogen-activated protein kinase kinase kinase 20 provided by MGI
Primary source	MGI:MGI:2443258
See related	Ensembl:ENSMUSG00000004085
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AV006891, B230120H23Rik, HCCS-4, MLTK, MLTKalpha, MLTKbeta, Zak
Expression	Broad expression in heart adult (RPKM 20.9), bladder adult (RPKM 15.7) and 20 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

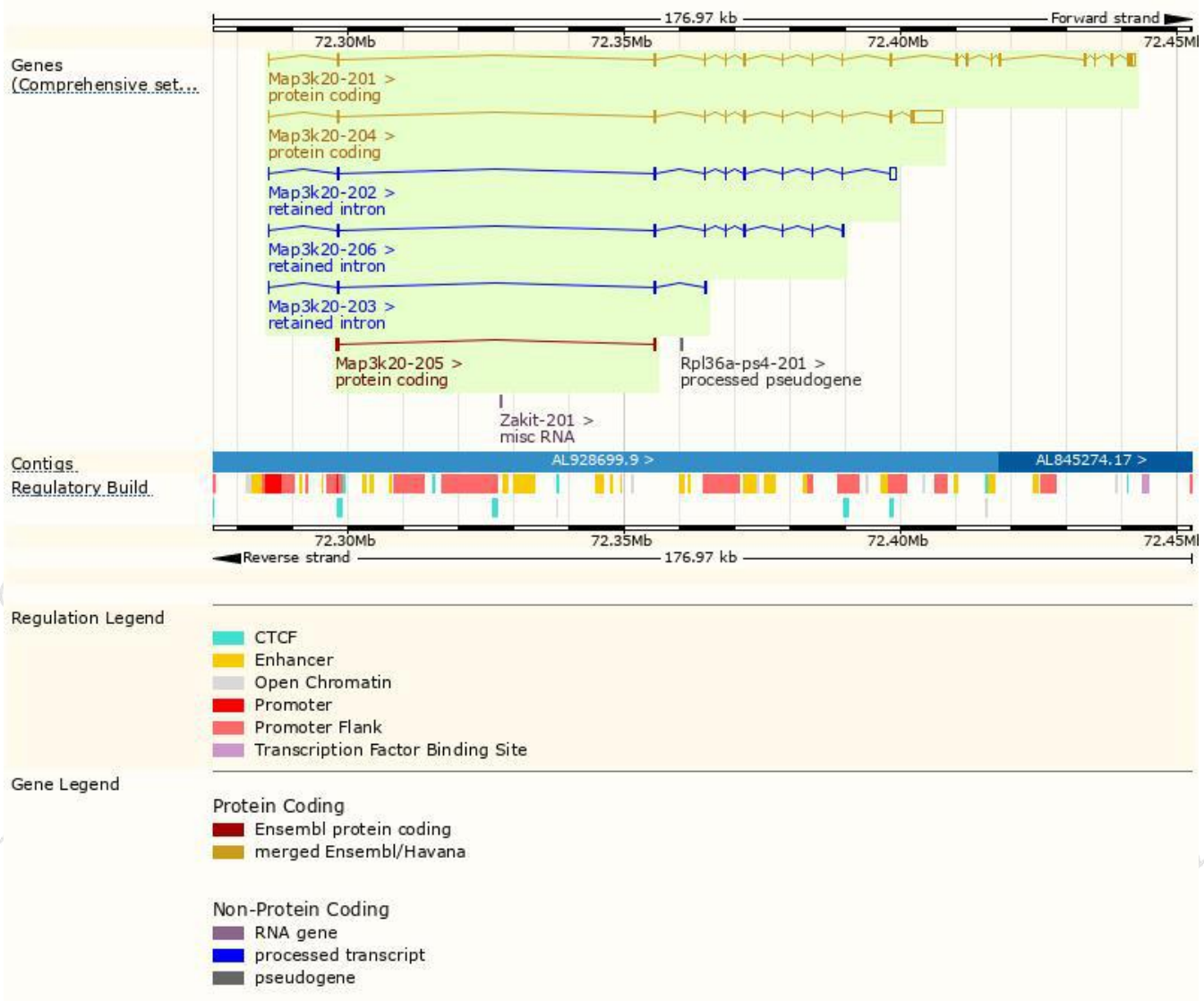
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Map3k20-204	ENSMUST00000135469.7	6663	454aa	Protein coding	CCDS50605	Q3TRG2 Q9ESL4	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS ALT2
Map3k20-201	ENSMUST00000090824.11	3334	802aa	Protein coding	CCDS38143	Q9ESL4	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P3
Map3k20-205	ENSMUST00000144111.1	396	74aa	Protein coding	-	A2ASW6	CDS 3' incomplete TSL:5
Map3k20-202	ENSMUST00000112073.8	1925	No protein	Retained intron	-	-	TSL:2
Map3k20-206	ENSMUST00000150126.7	1227	No protein	Retained intron	-	-	TSL:1
Map3k20-203	ENSMUST00000135204.1	559	No protein	Retained intron	-	-	TSL:2

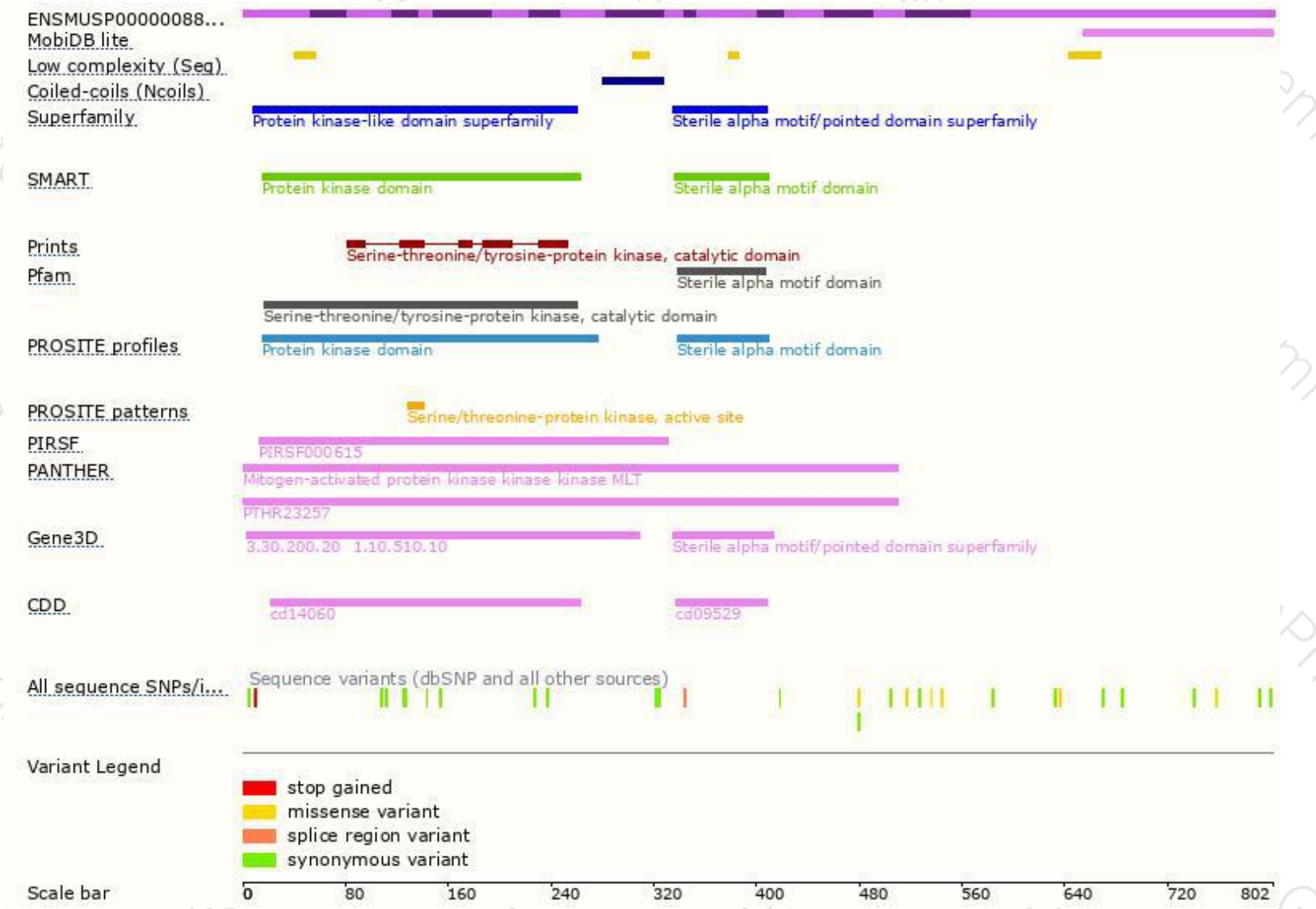
The strategy is based on the design of *Map3k20-201* transcript,the transcription is shown below



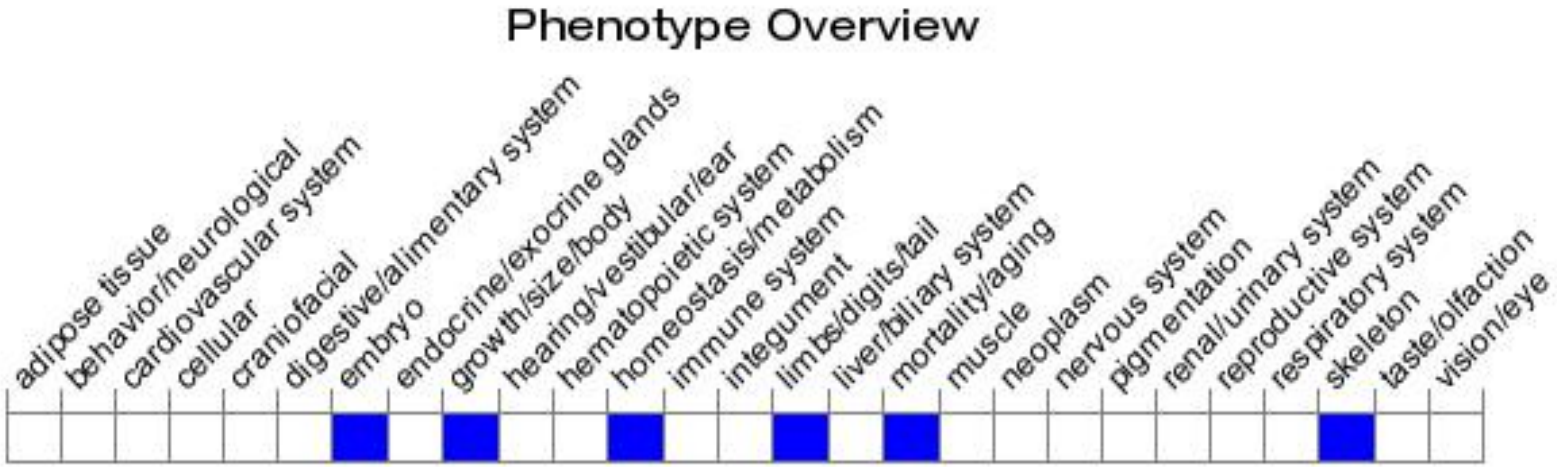
Genomic location distribution



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,mice homozygous for a knock-out allele exhibit complete lethality at E9.5 with growth retardation. Mice homozygous for an allele lacking the SAM domain exhibit low penetrant unilateral complex hindlim duplication phenotype.

集萃药康
GemPharmatech

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

Tel: 400-9660890

集萃药康生物科技
GemPharmatech Co.,Ltd