Polr1b Cas9-CKO Strategy

Designer: Daohua Xu
Reviewer: Huimin Su
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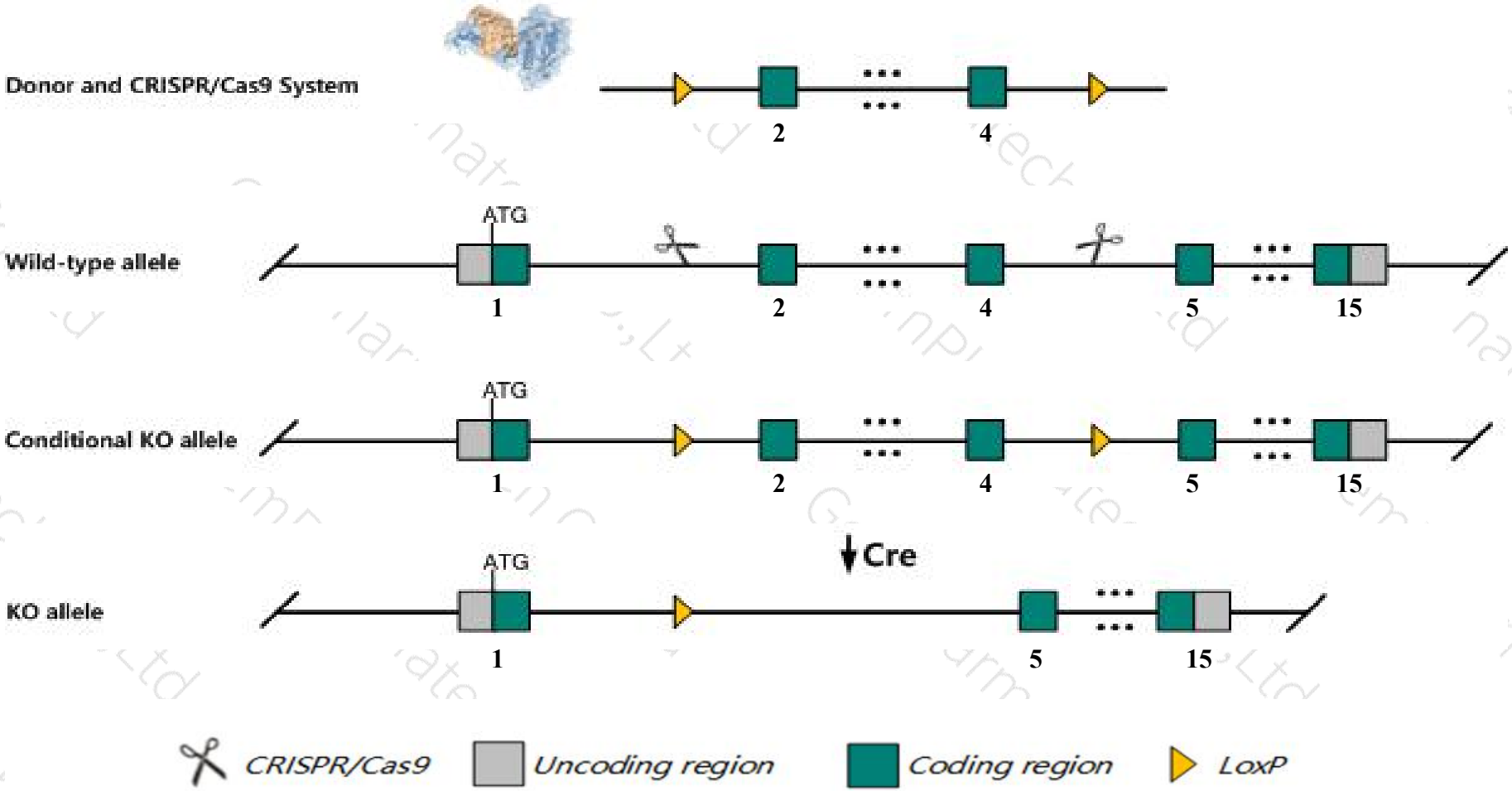
Project Overview

Project Name	***Polr1b***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Polr1b* gene. The schematic diagram is as follows:



Technical routes

- The *Polr1b* gene has 5 transcripts. According to the structure of *Polr1b* gene, exon2-exon4 of *Polr1b-202* (ENSMUST00000103205.10) transcript is recommended as the knockout region.The region contains 448bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Polr1b* 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 gene trapped allele exhibit embryonic lethality prior to implantation.

- The *Polr1b* 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）

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Polr1b polymerase (RNA) I polypeptide B [Mus musculus (house mouse)]

Gene ID: 20017, updated on 31-Jan-2019

Summary

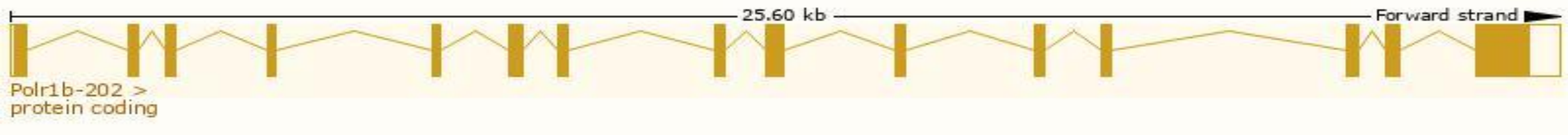
Official Symbol	Polr1b provided by MGI
Official Full Name	polymerase (RNA) I polypeptide B provided by MGI
Primary source	MGI:MGI:108014
See related	Ensembl:ENSMUSG00000027395
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	128kDa, D630020H17Rik, RPA116, RPA135, RPA2, Rpo1-2
Expression	Ubiquitous expression in limb E14.5 (RPKM 5.9), CNS E11.5 (RPKM 5.4) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

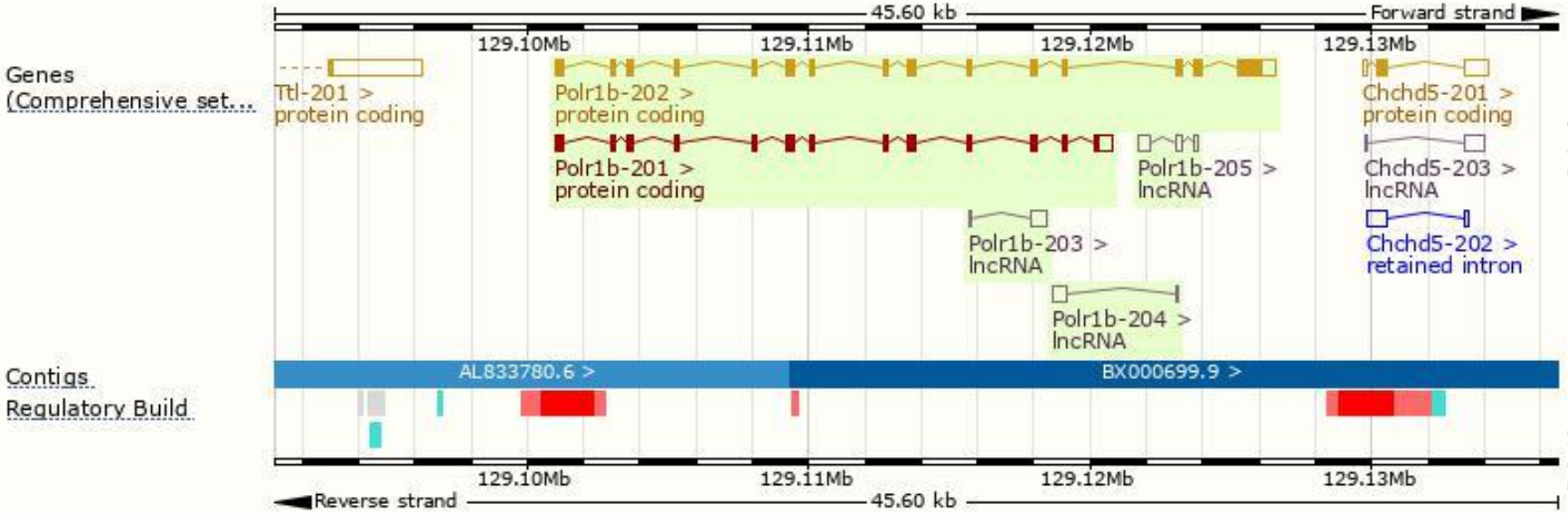
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Polr1b-202	ENSMUST00000103205.10	3998	1135aa	Protein coding	CCDS16720	P70700	TSL:1 GENCODE basic APPRIS P1
Polr1b-201	ENSMUST00000028874.7	2769	754aa	Protein coding	-	A2AP84	TSL:1 GENCODE basic
Polr1b-205	ENSMUST00000147727.1	720	No protein	lncRNA	-	-	TSL:3
Polr1b-203	ENSMUST00000133345.1	663	No protein	lncRNA	-	-	TSL:2
Polr1b-204	ENSMUST00000144004.1	612	No protein	lncRNA	-	-	TSL:2

The strategy is based on the design of *Polr1b-202* transcript,The transcription is shown below



Genomic location distribution



Gene Legend

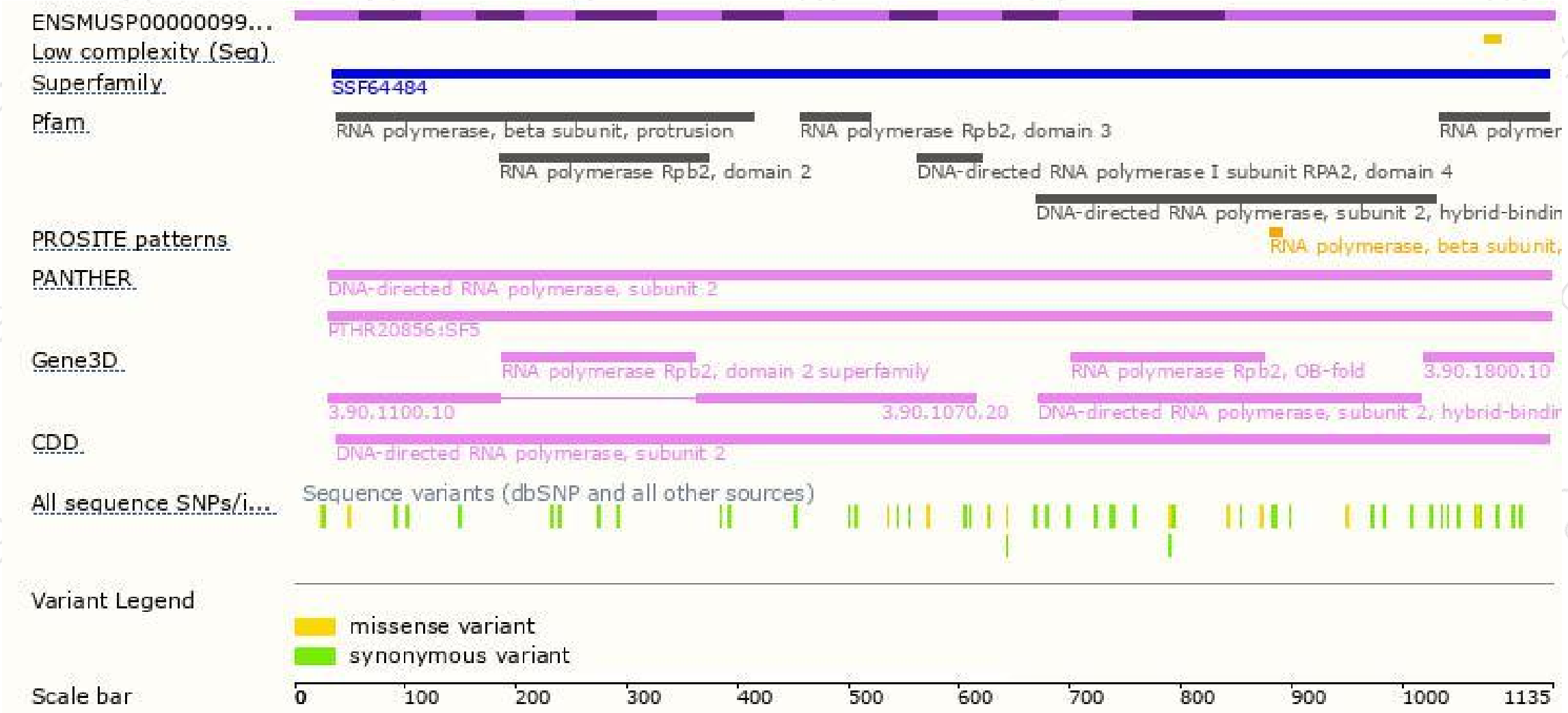
Protein Coding
- merged Ensembl/Havana
- Ensembl protein coding

Non-Protein Coding
- processed transcript
- RNA gene

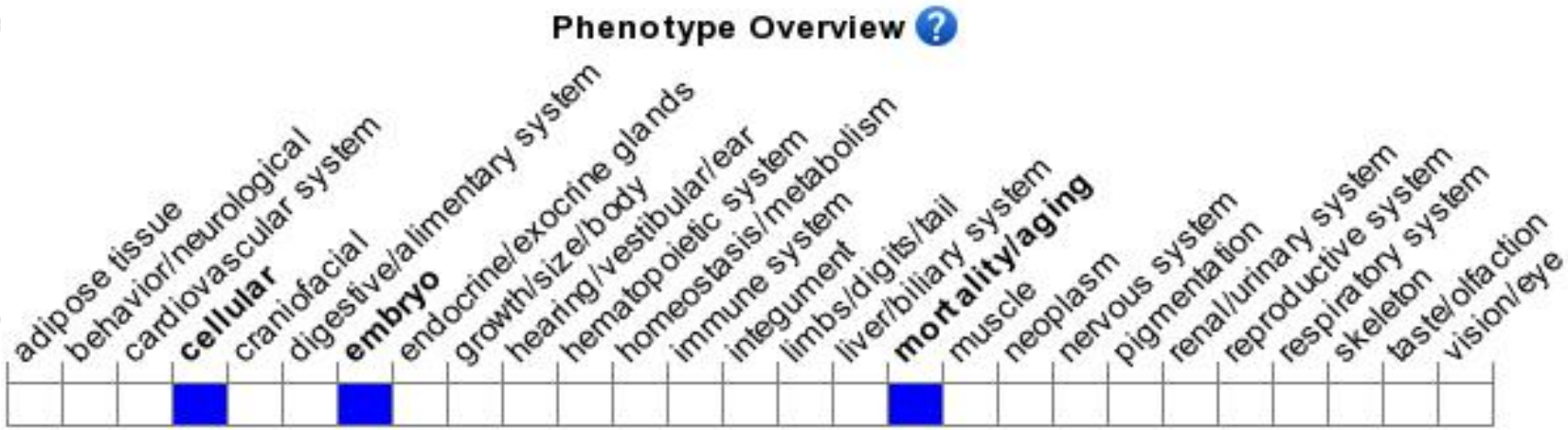
Regulation Legend

- CTCF
- Open Chromatin
- Promoter
- Promoter Flank

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/).

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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