Ppp1r3c Cas9-KO Strategy

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Reviewer: Daohua Xu

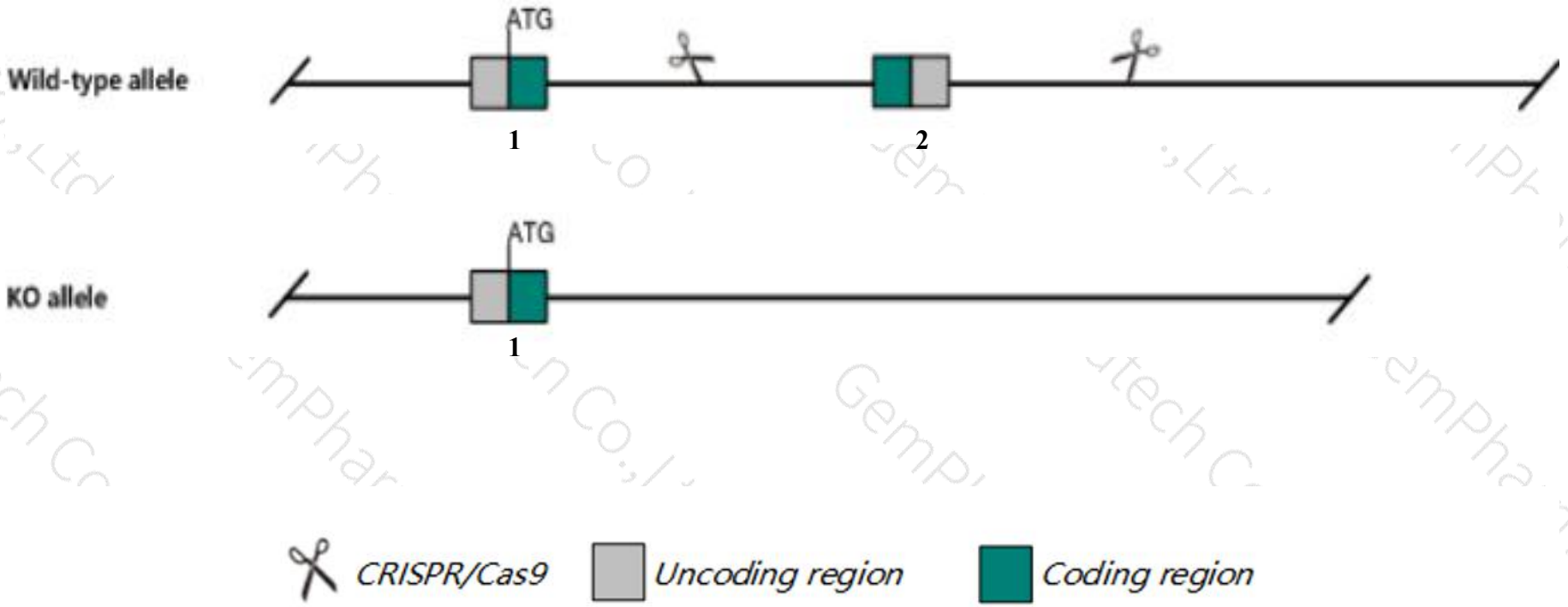
Design Date: 2020-7-24

Project Overview

Project Name	***Ppp1r3c***
Project type	**Cas9-KO**
Strain background	**C57BL/6JGpt**

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ppp1r3c* gene. The schematic diagram is as follows:



Technical routes

➢ The *Ppp1r3c* gene has 1 transcript. According to the structure of *Ppp1r3c* gene, exon2 of *Ppp1r3c-201*(ENSMUST00000087321.3) transcript is recommended as the knockout region.The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.

➢ In this project we use CRISPR/Cas9 technology to modify *Ppp1r3c* 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.

Notice

➢ According to the existing MGI data,homozygous null mice are embyronic lethal. Heterozygotes have reduced glycogen stores, attenuated glycogen synthesis, glucose intolerance, hyperinsulinemia and insulin resistance. Mice homozygous for a different knock-out allele exhibit normal lifespan with enhanced whole body insulin sensitivity.

➢ The *Ppp1r3c* gene is located on the Chr19. 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.

Gene information（NCBI）

集萃药康 GemPharmatech

Ppp1r3c protein phosphatase 1, regulatory subunit 3C [Mus musculus (house mouse)]

Gene ID: 53412, updated on 13-Mar-2020

Summary

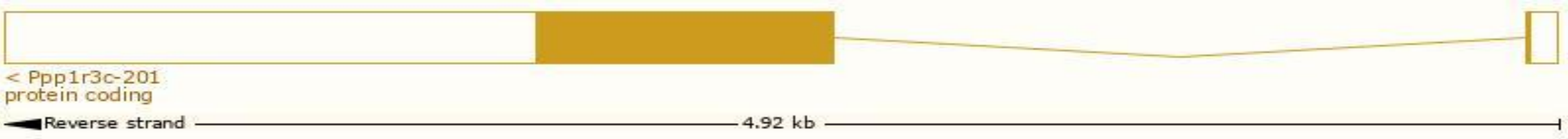
Official Symbol	Ppp1r3c provided by MGI
Official Full Name	protein phosphatase 1, regulatory subunit 3C provided byMGI
Primary source	MGI:MGI:1858229
See related	Ensembl:ENSMUSG00000067279
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	PTG, Ppp1r5
Expression	Broad expression in heart adult (RPKM 24.0), liver E18 (RPKM 21.6) and 18 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

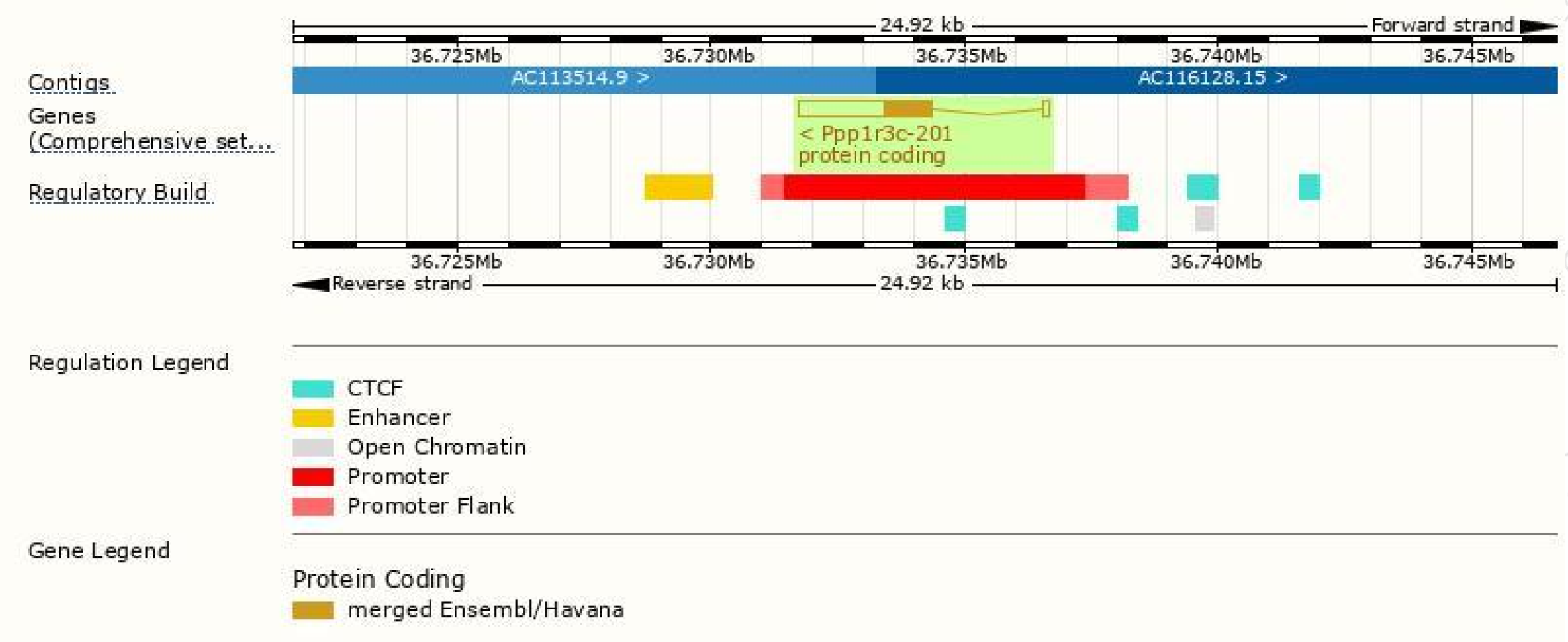
The gene has 1 transcript,and the transcript is shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ppp1r3c-201	ENSMUST00000087321.3	2725	317aa	Protein coding	CCDS37968	Q7TMB3	TSL:1 GENCODE basic APPRIS P1

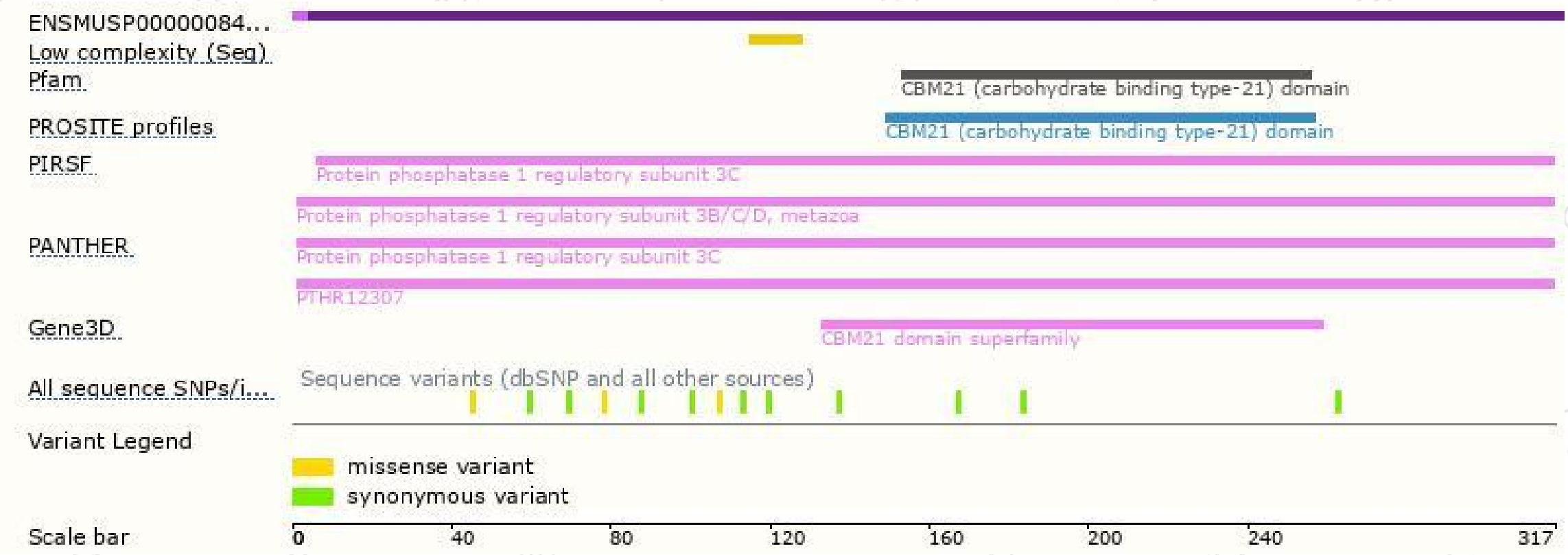
The strategy is based on the design of *Ppp1r3c-201* transcript,the transcription is shown below:



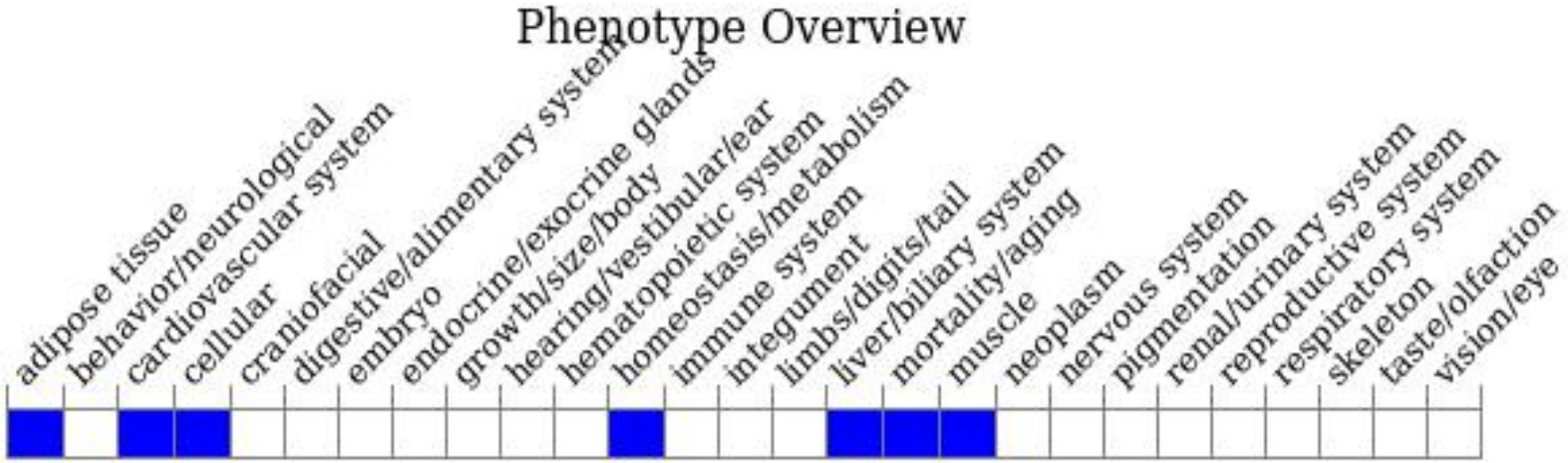
Genomic location distribution



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,homozygous null mice are embryonic lethal. Heterozygotes have reduced glycogen stores, attenuated glycogen synthesis, glucose intolerance, hyperinsulinemia and insulin resistance. Mice homozygous for a different knock-out allele exhibit normal lifespan with enhanced whole body insulin sensitivity.

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

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