Cfi Cas9-CKO Strategy

Designer: Daohua Xu
Reviewer: Huimin Su

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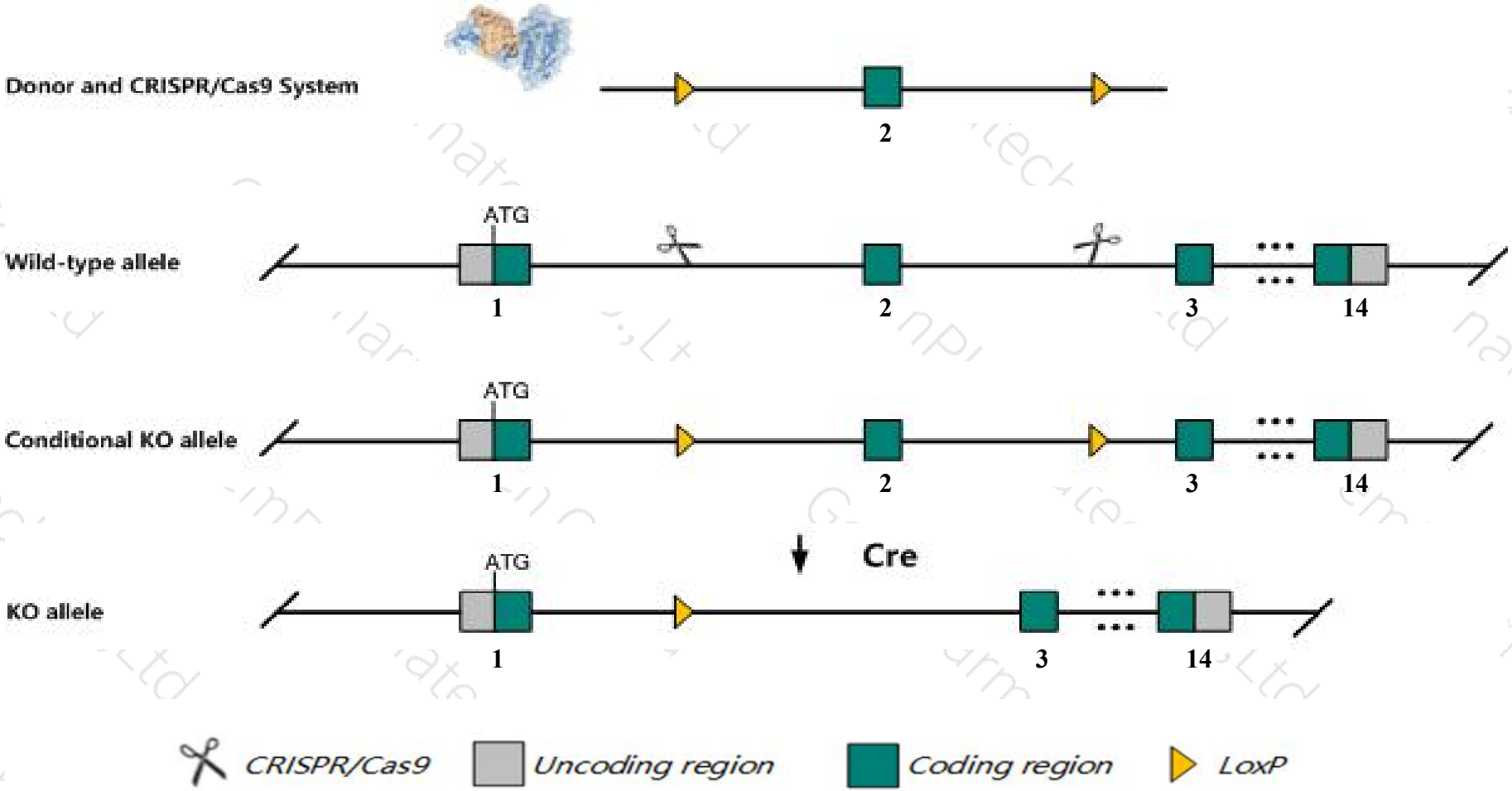
Project Overview

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

Conditional Knockout strategy

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



Technical routes

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- The *Cfi* gene has 2 transcripts. According to the structure of *Cfi* gene, exon2 of *Cfi-201* (ENSMUST00000077918.6) transcript is recommended as the knockout region.The region contains 280bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Cfi* 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,Homozygous null mice display uncontrolled alternative pathway activation as shown by reduced complement C3, factor B, and factor H levels, but do not develop C3 deposition along the glomerular basement membrane or membranoproliferative glomerulonephritistype II. Plasma C3 circulates as C3b.

➢ The *Cfi* gene is located on the Chr3. 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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Cfi complement component factor i [Mus musculus (house mouse)]

Gene ID: 12630, updated on 31-Jan-2019

Summary

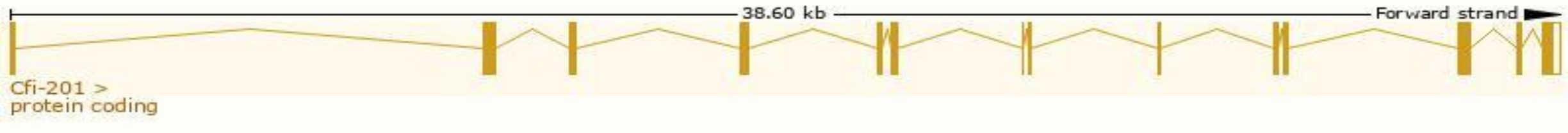
Official Symbol	Cfi provided by MGI
Official Full Name	complement component factor i provided byMGI
Primary source	MGI:MGI:105937
See related	Ensembl:ENSMUSG00000058952
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Summary	This gene encodes a serine protease that plays an important role in the classical and alternative complement pathways where it cleaves C4b and C3b components of C3 and C5 convertases. The encoded preproprotein undergoes proteolytic processing to generate an active, disulfide-linked heterodimeric enzyme comprised of heavy and light chains. [provided by RefSeq, Jul 2016]
Expression	Biased expression in liver E18 (RPKM 164.6), liver adult (RPKM 83.6) and 3 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

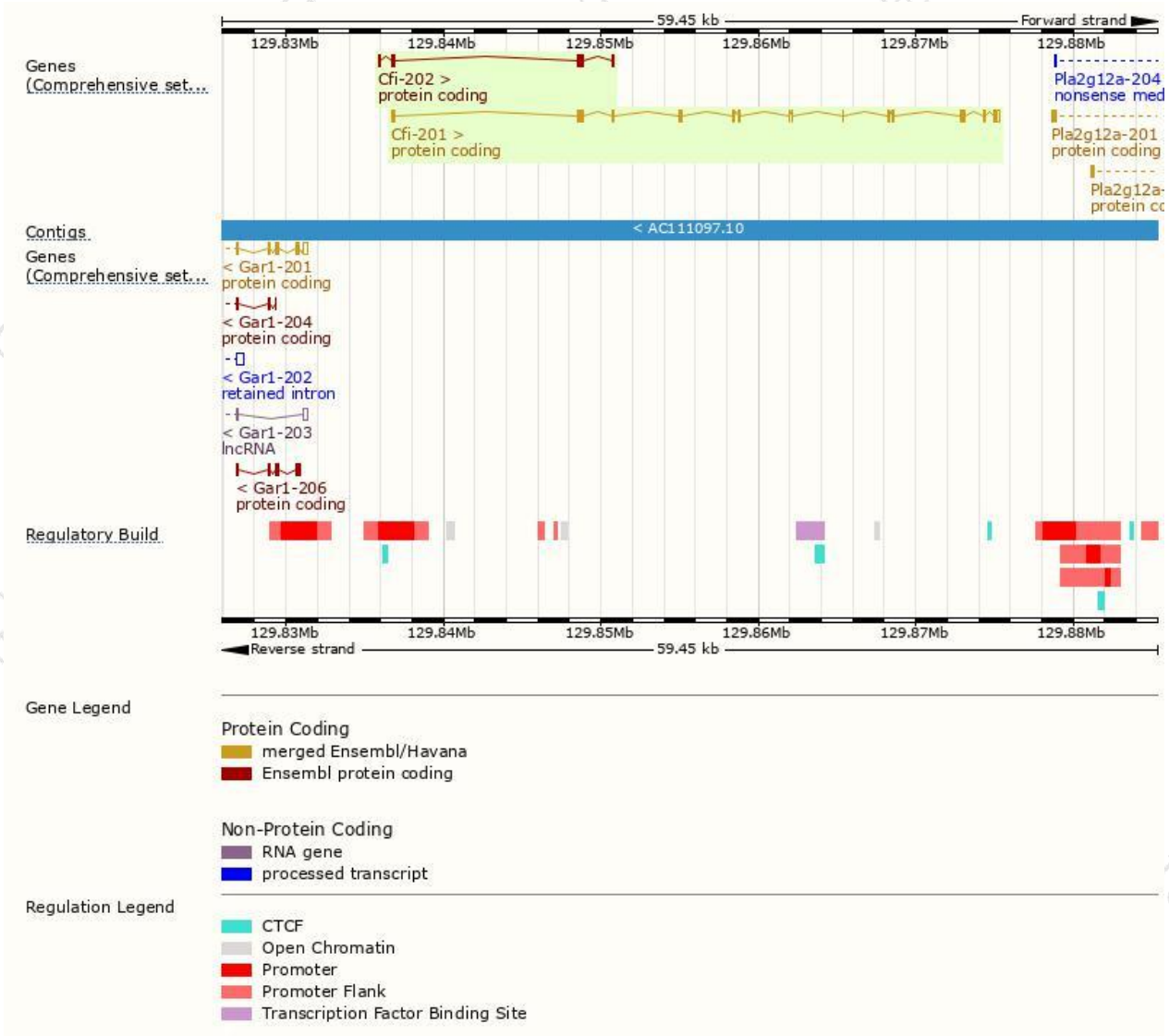
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cfi-201	ENSMUST00000077918.6	2079	603aa	Protein coding	CCDS17835	Q61129	TSL:1 GENCODE basic APPRIS P1
Cfi-202	ENSMUST00000200206.4	651	145aa	Protein coding	-	A0A0G2JF07	CDS 3' incomplete TSL:3

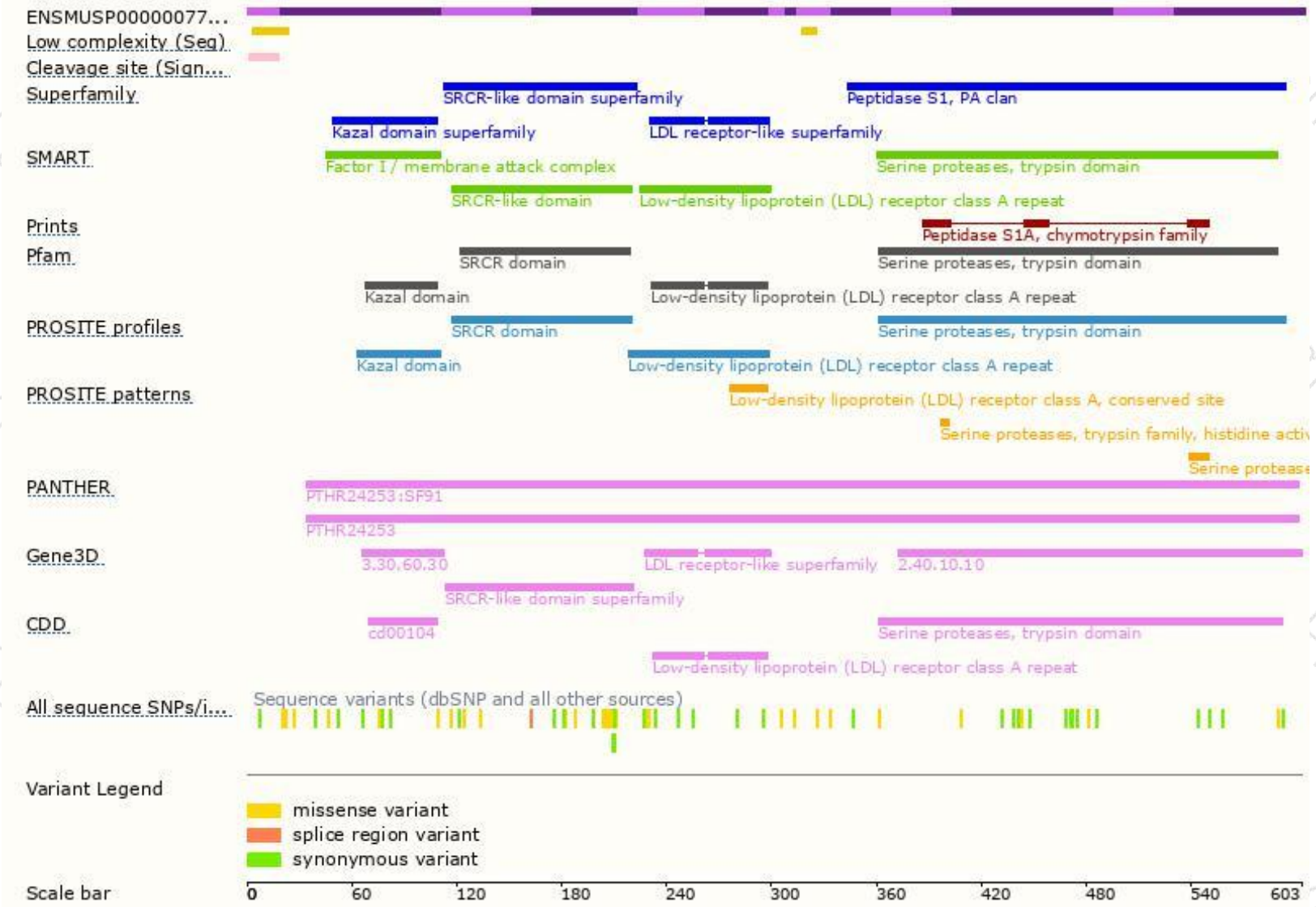
The strategy is based on the design of *Cfi-201* transcript,The transcription is shown below



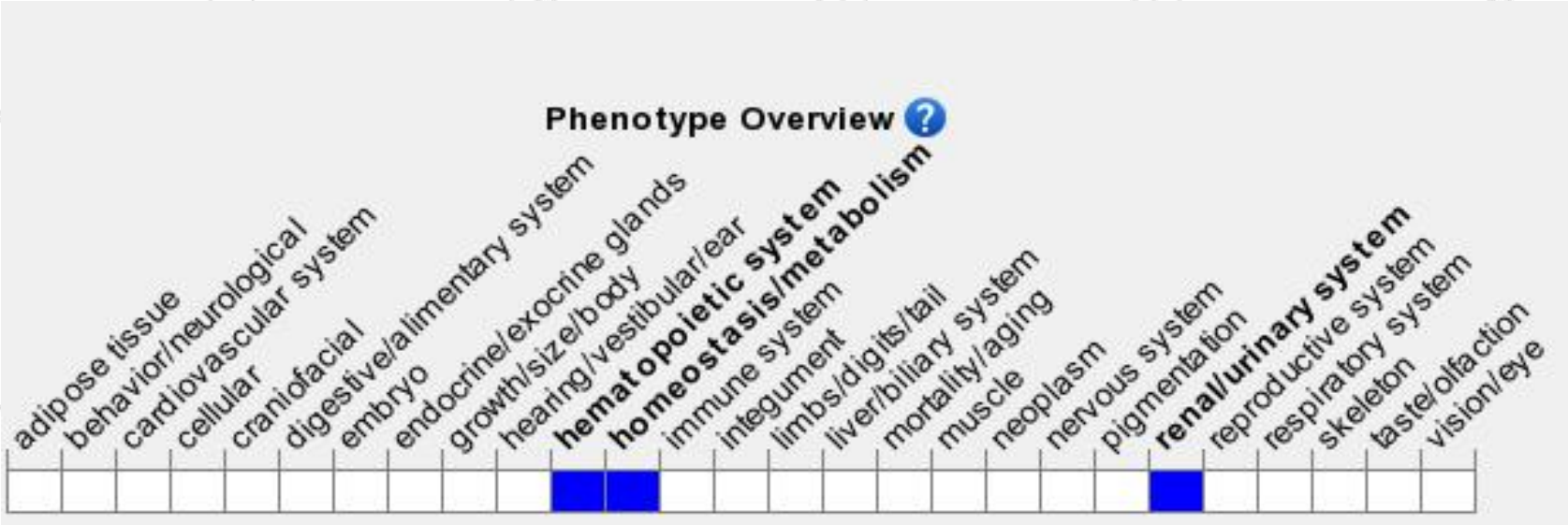
Genomic location distribution



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/).

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

Tel: 400-9660890

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