

# ***Kcnu1* Cas9-KO Strategy**

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**Reviewer: Rui Xiong**

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# Project Overview

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**Project Name**

*Kcnu1*

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**Project type**

**Cas9-KO**

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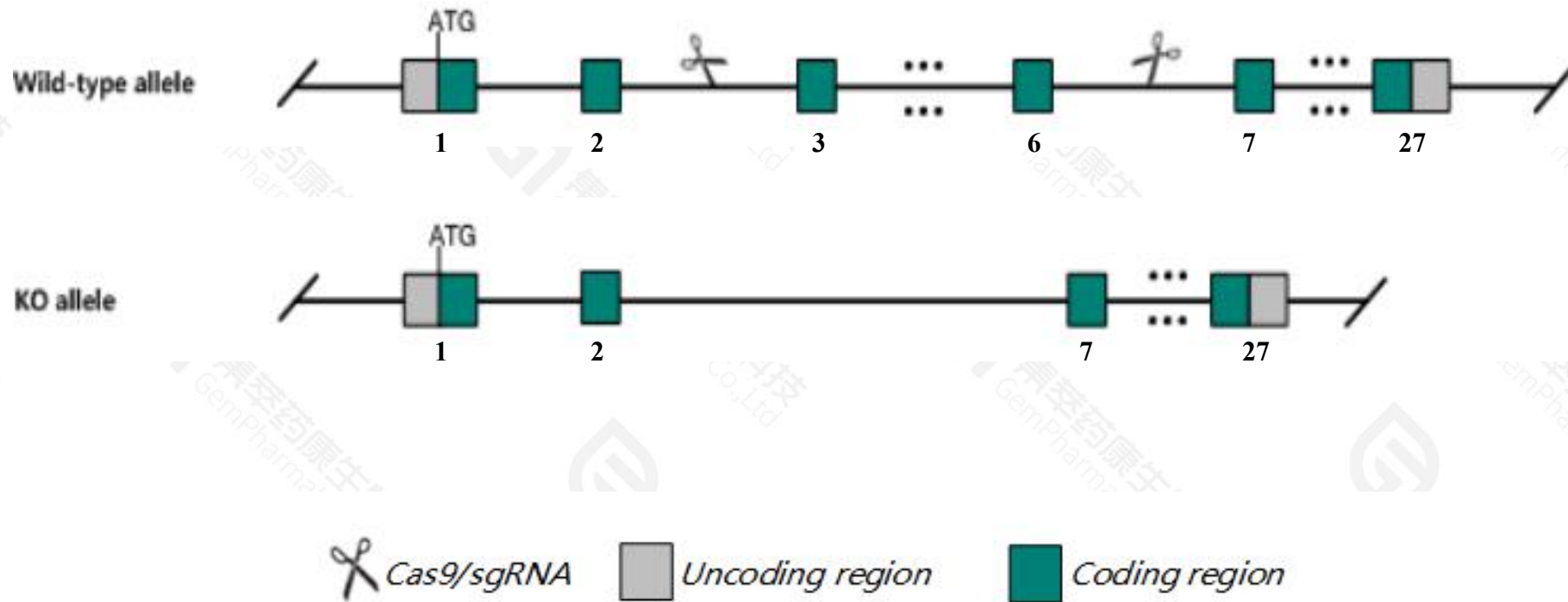
**Strain background**

**C57BL/6JGpt**

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# Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Kcnu1* gene. The schematic diagram is as follows:



- The *Kcnu1* gene has 4 transcripts. According to the structure of *Kcnu1* gene, exon3-exon6 of *Kcnu1*-201(ENSMUST00000098858.11) transcript is recommended as the knockout region. The region contains 341bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Kcnu1* gene. The brief process is as follows: sgRNA was transcribed in vitro. Cas9 and sgRNA 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.



- According to the existing MGI data, homozygous male mutants are infertile with impaired sperm capacitation.
- Transcripts of *Kcnu1-202,203* (protein coding) are not be affected.
- The *Kcnu1* 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.

# Gene information (NCBI)

## Kcnu1 potassium channel, subfamily U, member 1 [Mus musculus (house mouse)]

Gene ID: 16532, updated on 15-Mar-2020

### Summary



**Official Symbol** Kcnu1 provided by [MGI](#)

**Official Full Name** potassium channel, subfamily U, member 1 provided by [MGI](#)

**Primary source** [MGI:MGI:1202300](#)

**See related** [Ensembl:ENSMUSG00000031576](#)

**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** Kcnma3, Slo3, mSlo3

**Expression** Biased expression in testis adult (RPKM 17.3) and CNS E18 (RPKM 1.2)[See more](#)

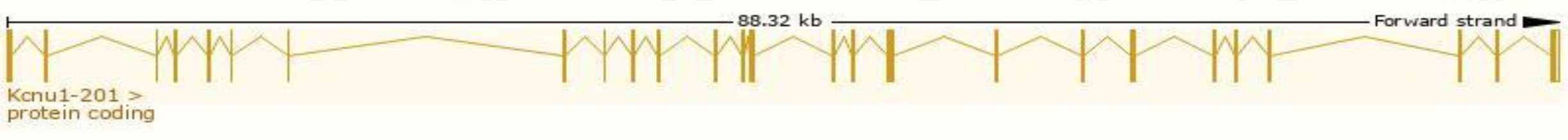
**Orthologs** [human](#) [all](#)

# Transcript information (Ensembl)

The gene has 4 transcripts,all transcripts are shown below:

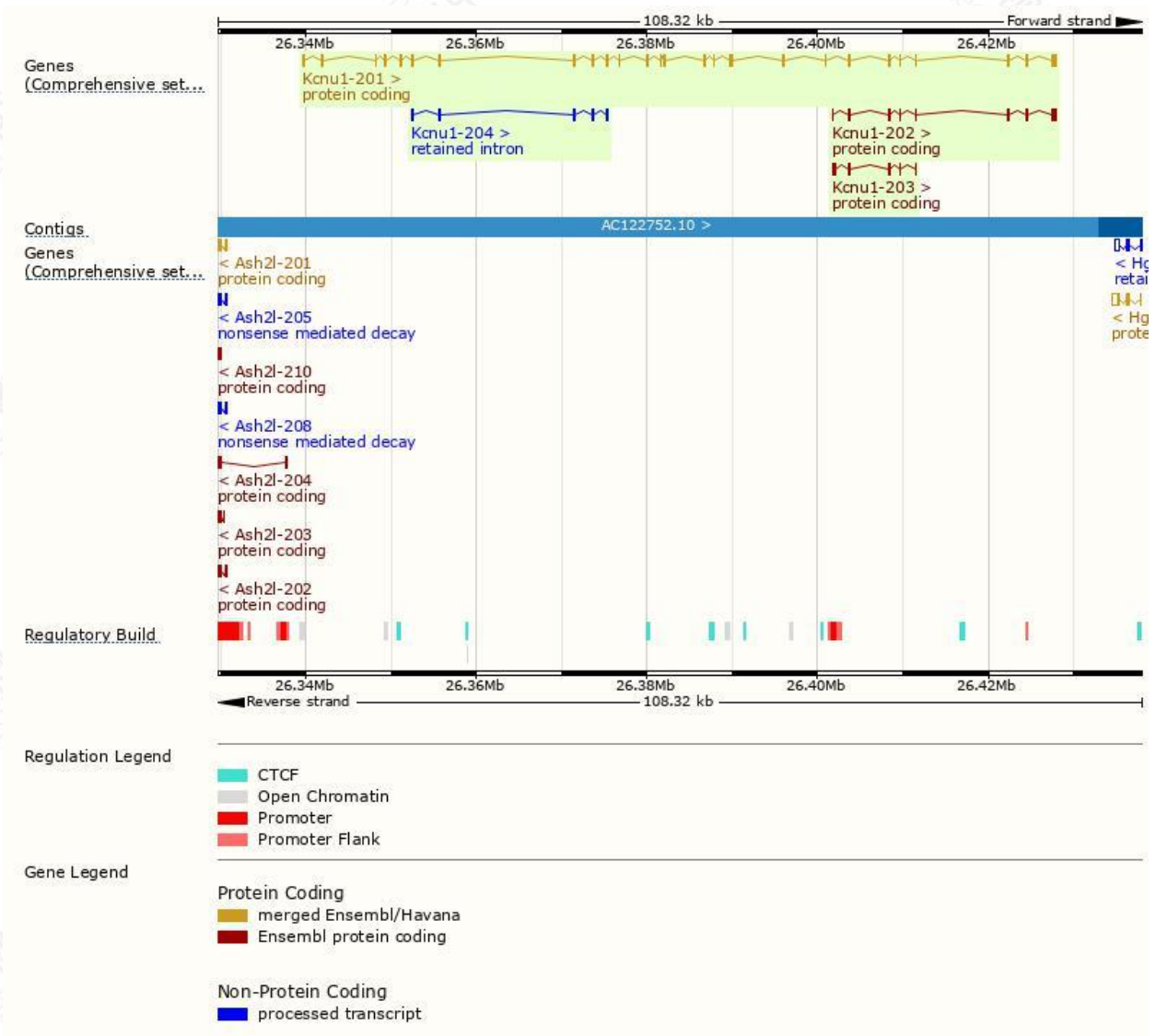
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Kcnu1-201	<a href="#">ENSMUST00000098858.10</a>	3536	<a href="#">1121aa</a>	Protein coding	<a href="#">CCDS52530</a>	<a href="#">G3X9P7</a>	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Kcnu1-202	<a href="#">ENSMUST00000120653.7</a>	1446	<a href="#">381aa</a>	Protein coding	-	<a href="#">D3Z5P2</a>	TSL:1 GENCODE basic
Kcnu1-203	<a href="#">ENSMUST00000126226.1</a>	697	<a href="#">160aa</a>	Protein coding	-	<a href="#">D3Z073</a>	CDS 3' incomplete TSL:3
Kcnu1-204	<a href="#">ENSMUST00000210273.1</a>	549	No protein	Retained intron	-	-	TSL:5

The strategy is based on the design of *Kcnu1-201* transcript,the transcription is shown below:



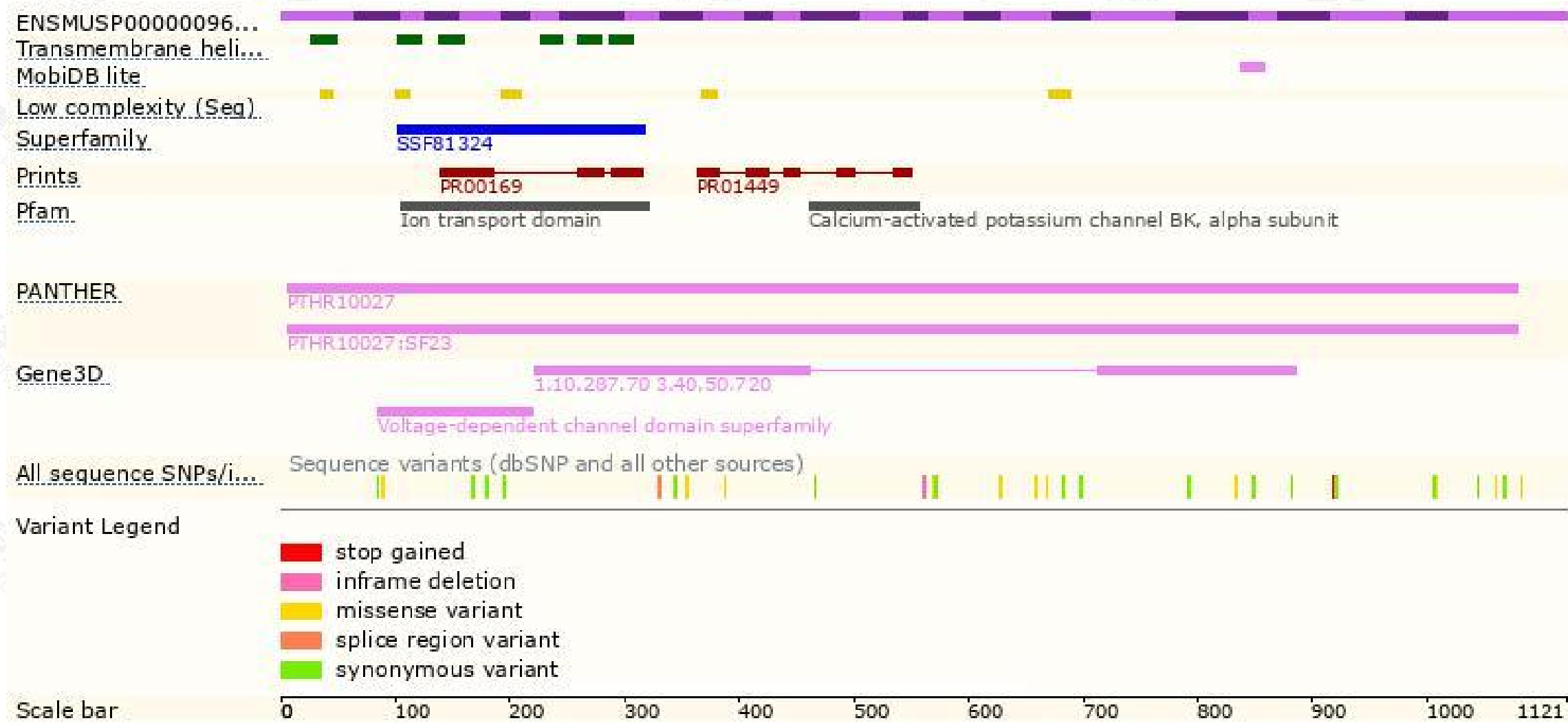


# Genomic location distribution

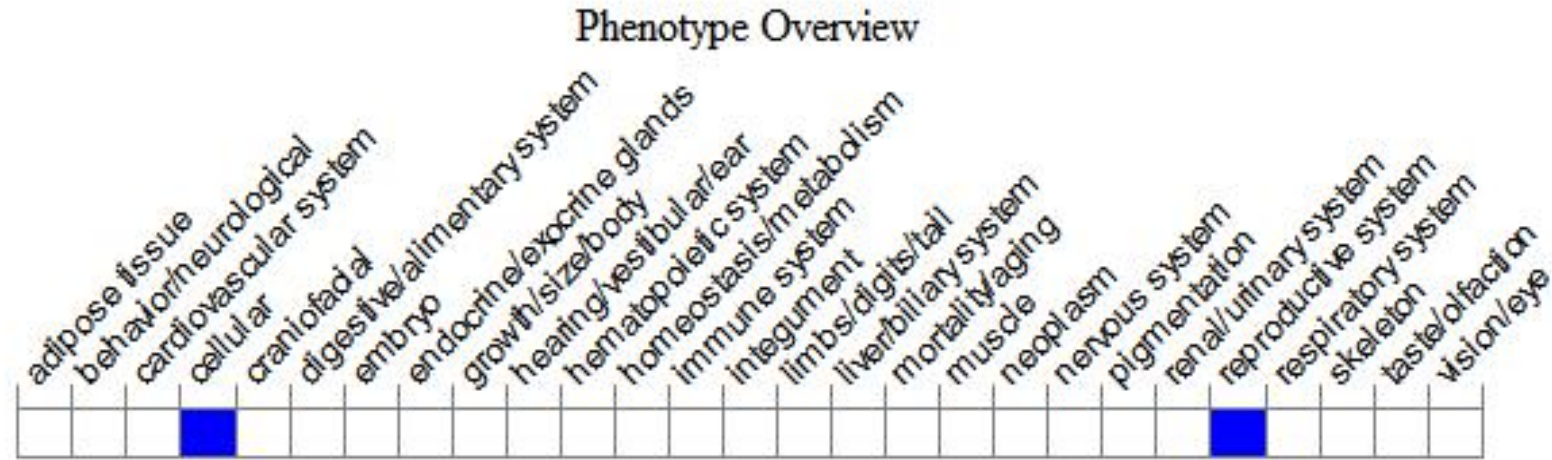




# Protein domain



# 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 male mutants are infertile with impaired sperm capacitation.

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

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