Usp20 Cas9-CKO Strategy

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Project Overview

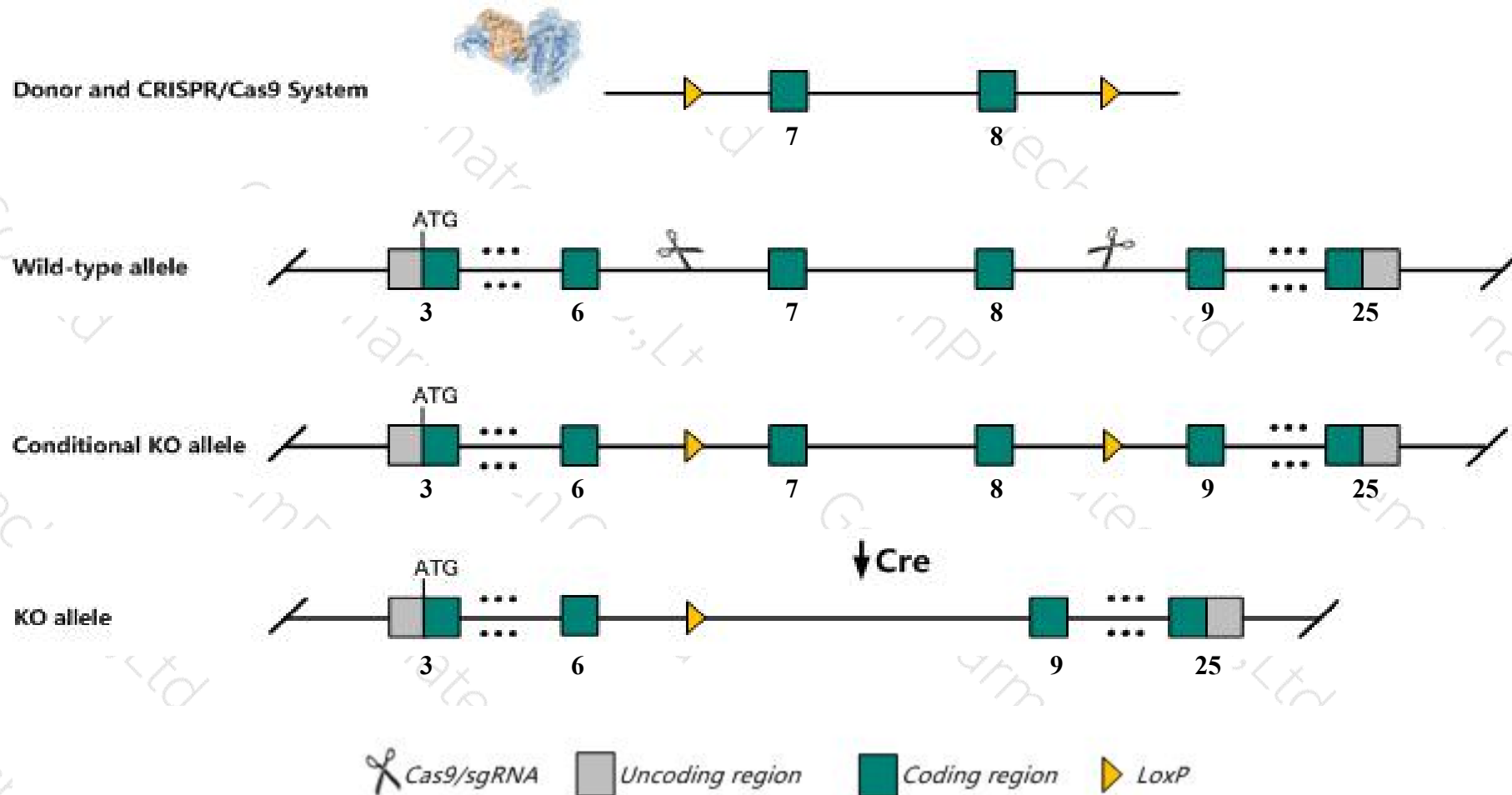|	
Project Name	*Usp20*
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Usp20* gene. The schematic diagram is as follows:

Technical routes

➤ The *Usp20* gene has 12 transcripts. According to the structure of *Usp20* gene, exon7-exon8 of *Usp20-202* (ENSMUST00000102849.10) transcript is recommended as the knockout region.The region contains 167bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Usp20* gene. The brief process is as follows:sgRNA was transcribed in vitro, donor vector was constructed.Cas9, sgRNA 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 was 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

➢ The *Usp20* 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.

➢ Transcript *Usp20-208* is not be affected.

➢ 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）

Usp20 ubiquitin specific peptidase 20 [Mus musculus (house mouse)]

Gene ID: 74270, updated on 31-Jan-2019

Summary

|	
Official Symbol	Usp20 provided by MGI
Official Full Name	ubiquitin specific peptidase 20 provided by MGI
Primary source	MGI:MGI:1921520
See related	Ensembl:ENSMUSG00000026854
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	1700055M05Rik, AI467231, Vdu2
Expression	Ubiquitous expression in testis adult (RPKM 28.5), genital fat pad adult (RPKM 18.2) and 28 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

The gene has 12 transcripts,all transcripts are shown below:

| Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Usp20-202	ENSMUST00000102849.10	5088	916aa	Protein coding	CCDS15893	Q8C6M1	TSL:1 GENCODE basic APPRIS P1
Usp20-211	ENSMUST00000170476.7	4970	916aa	Protein coding	CCDS15893	Q8C6M1	TSL:1 GENCODE basic APPRIS P1
Usp20-201	ENSMUST00000061544.10	1360	210aa	Protein coding	-	Q8C9A6	TSL:1 GENCODE basic
Usp20-205	ENSMUST00000128295.7	639	77aa	Protein coding	-	A2AQ35	CDS 3' incomplete TSL:2
Usp20-203	ENSMUST00000125601.7	467	66aa	Protein coding	-	A2AQ34	CDS 3' incomplete TSL:5
Usp20-207	ENSMUST00000138161.7	462	77aa	Protein coding	-	A2AQ35	CDS 3' incomplete TSL:3
Usp20-208	ENSMUST00000142232.1	438	100aa	Protein coding	-	B0R093	CDS 3' incomplete TSL:5
Usp20-206	ENSMUST00000136588.2	430	143aa	Protein coding	-	F6QWP5	5' and 3' truncations in transcript evidence prevent annotation of the start and the end of the CDS. CDS 5' and 3' incomplete TSL:5
Usp20-209	ENSMUST00000154351.1	526	No protein	Processed transcript	-	-	TSL:5
Usp20-212	ENSMUST00000192882.1	1901	No protein	Retained intron	-	-	TSL:NA
Usp20-210	ENSMUST00000154634.1	869	No protein	Retained intron	-	-	TSL:3
Usp20-204	ENSMUST00000127776.1	622	No protein	Retained intron	-	-	TSL:3

The strategy is based on the design of *Usp20-202* transcript,The transcription is shown below
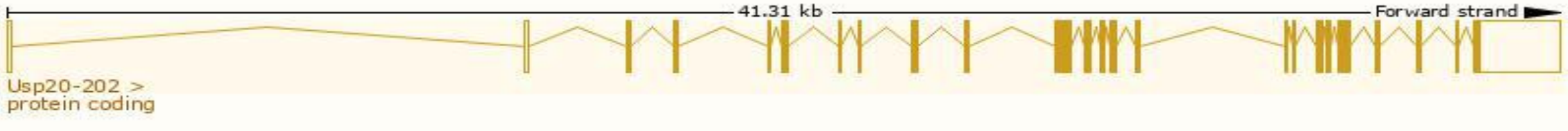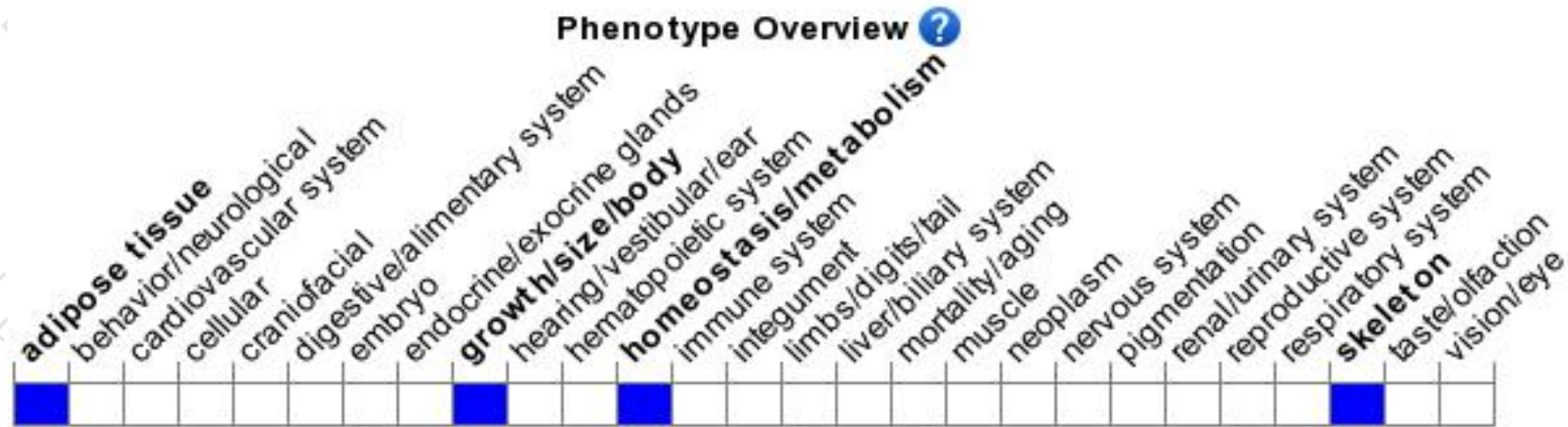
Genomic location distribution
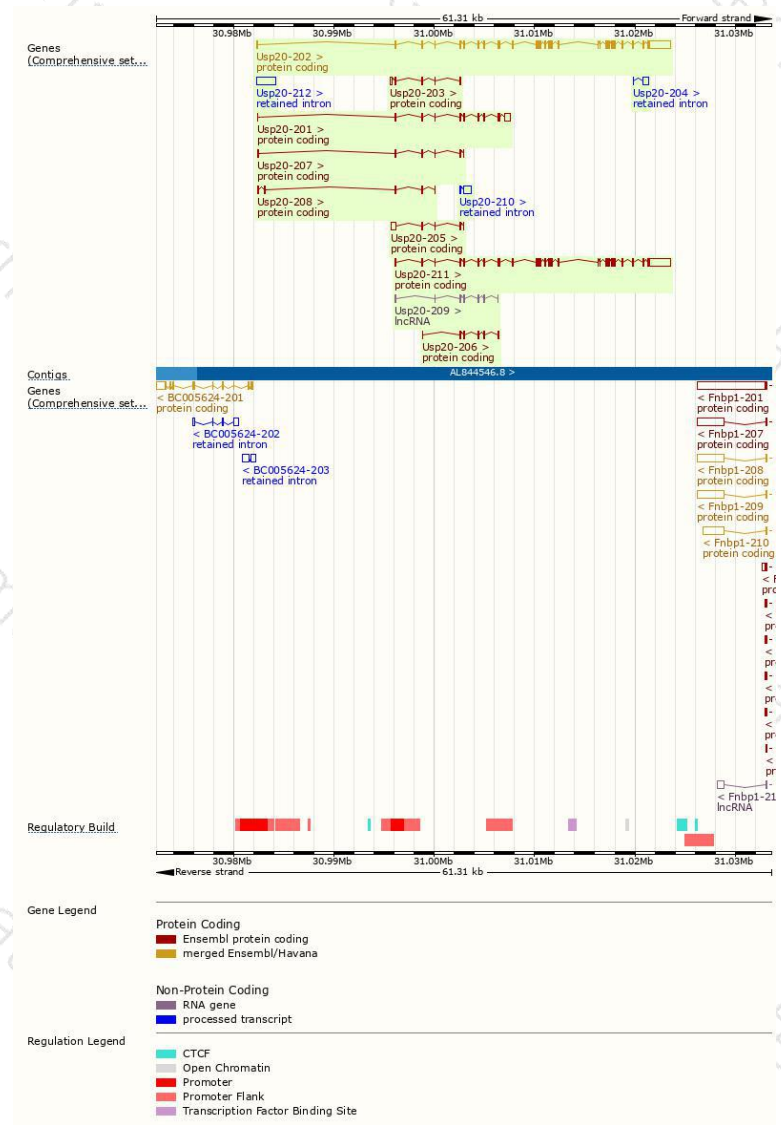
Protein domain
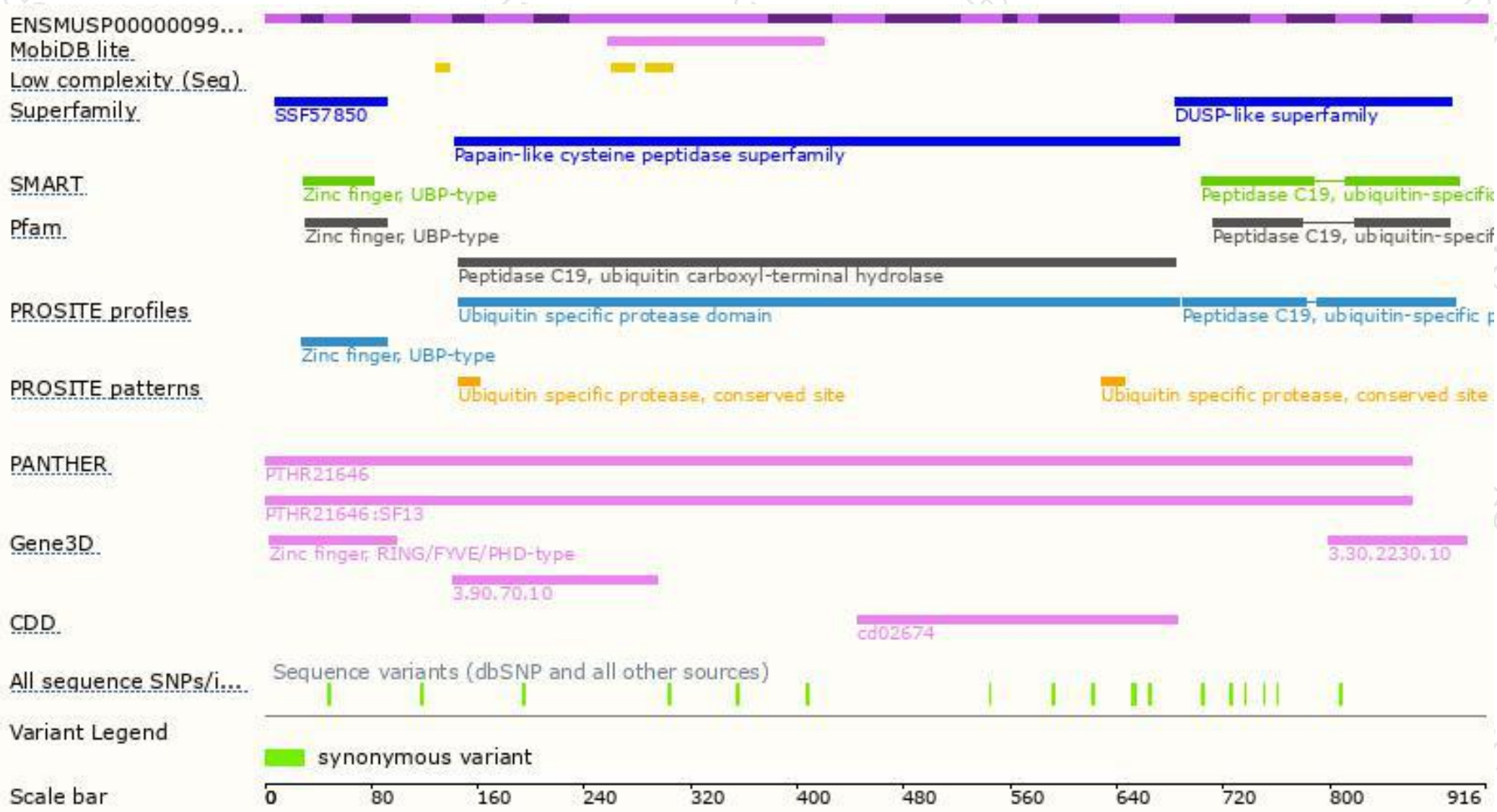
Mouse phenotype description(MGI)

Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

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