

Rasgrp1 Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Rasgrp1

Project type

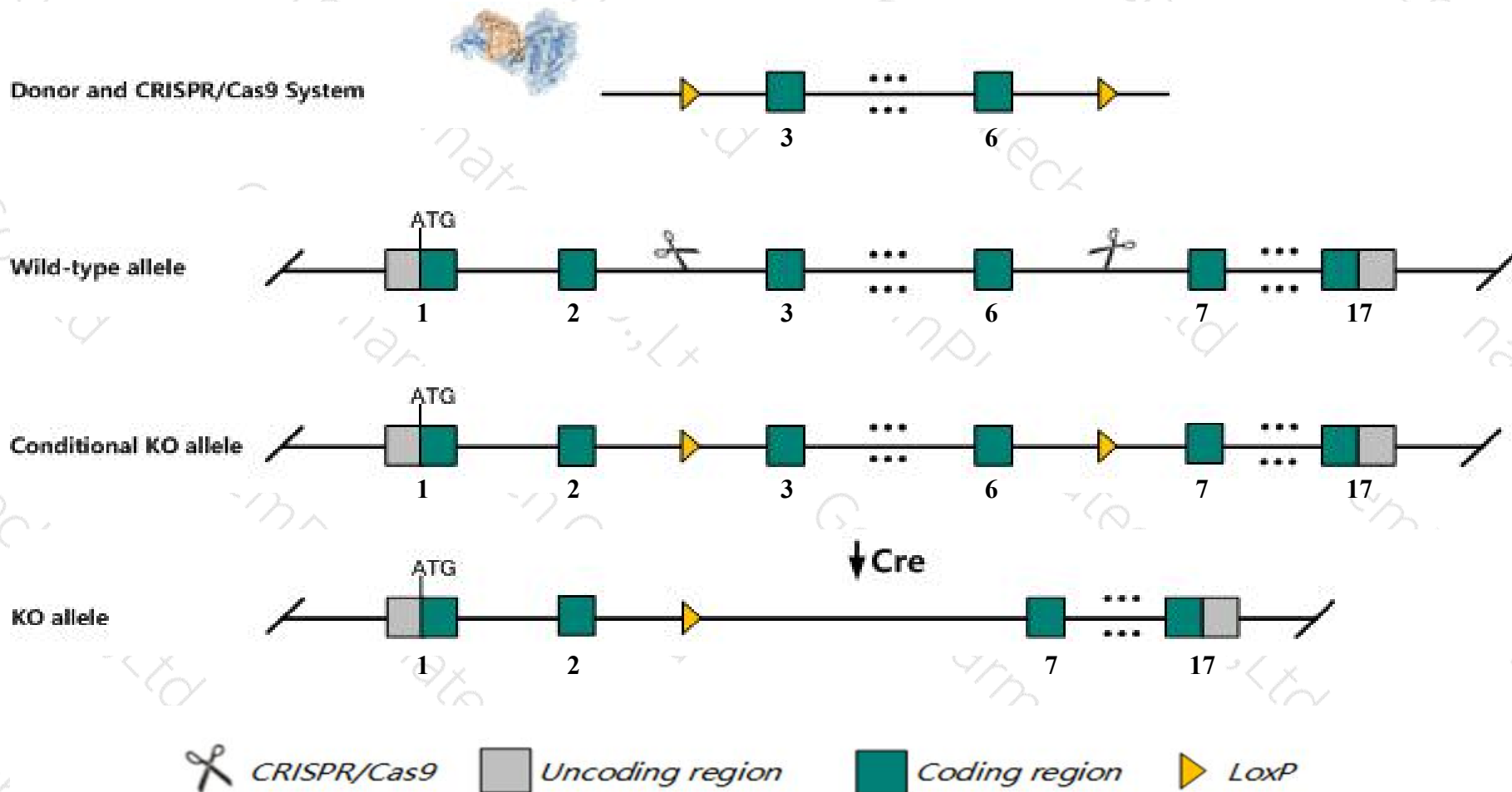
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Rasgrp1* gene. The schematic diagram is as follows:



- The *Rasgrp1* gene has 7 transcripts. According to the structure of *Rasgrp1* gene, exon3-exon6 of *Rasgrp1-201* (ENSMUST00000102534.10) transcript is recommended as the knockout region. The region contains 455bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rasgrp1* 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.

- According to the existing MGI data, Homozygotes for spontaneous and targeted null mutations exhibit a lymphoproliferative autoimmune syndrome in which T cells fail to activate Ras or proliferate after antigen exposure, defects in positive selection, and enlarged spleen and lymph nodes.
- The *Rasgrp1* 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)

Rasgrp1 RAS guanyl releasing protein 1 [*Mus musculus* (house mouse)]

Gene ID: 19419, updated on 10-Oct-2019

Summary

Official Symbol	Rasgrp1 provided by MGI
Official Full Name	RAS guanyl releasing protein 1 provided by MGI
Primary source	MGI:MGI:1314635
See related	Ensembl:ENSMUSG000000027347
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	Rasgrp; calDAG-GEFII
Expression	Biased expression in frontal lobe adult (RPKM 49.5), cortex adult (RPKM 48.8) and 5 other tissues See more
Orthologs	human all

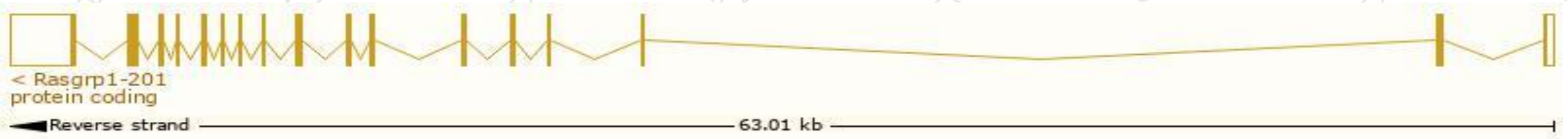


Transcript information (Ensembl)

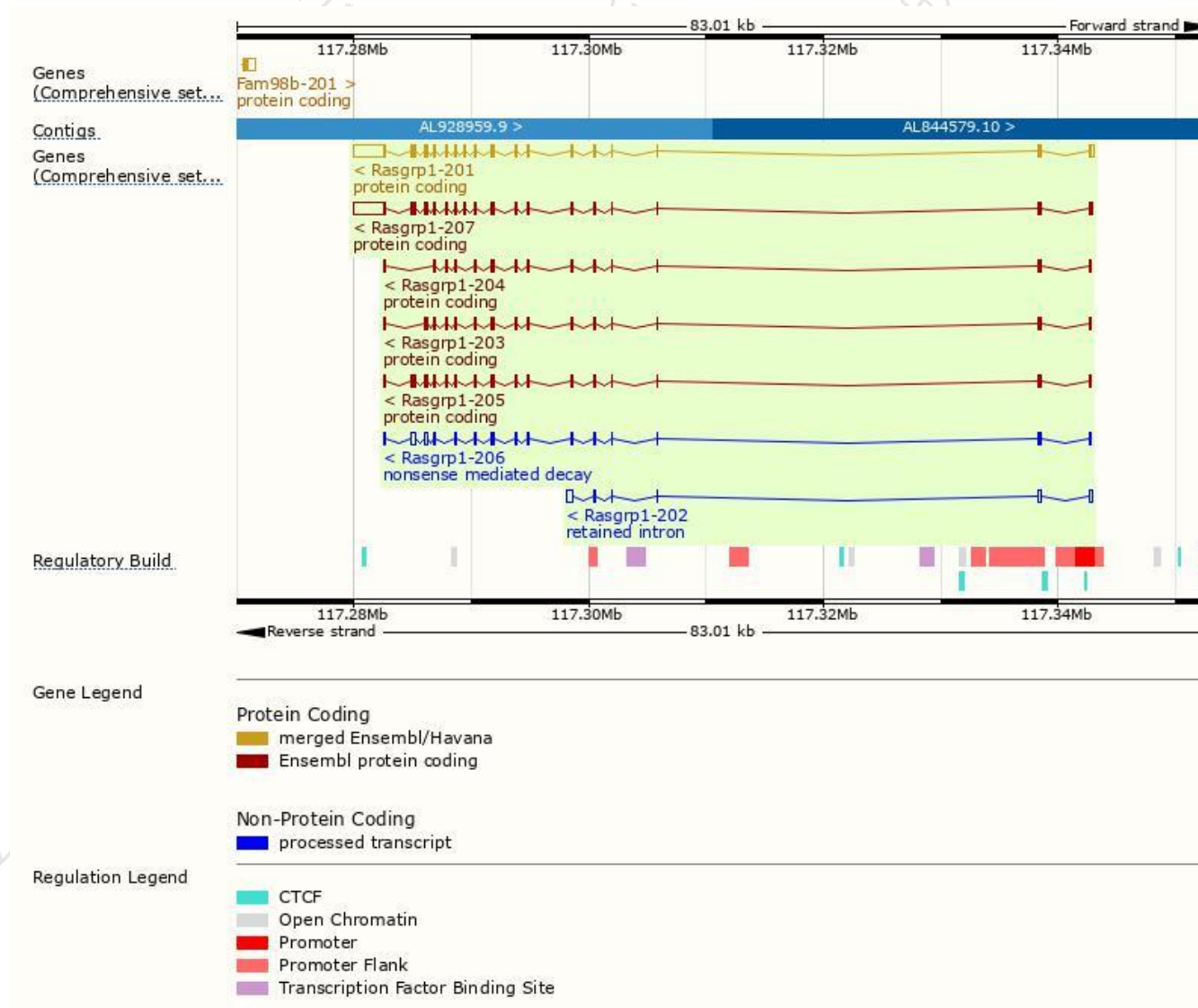
The gene has 7 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rasgrp1-201	ENSMUST00000102534.10	5229	795aa	Protein coding	CCDS16572	Q9Z1S3	TSL:5 GENCODE basic APPRIS P1
Rasgrp1-207	ENSMUST00000178884.7	5104	795aa	Protein coding	-	Q9Z1S3	TSL:1 GENCODE basic APPRIS P1
Rasgrp1-205	ENSMUST00000173541.7	2283	760aa	Protein coding	-	G3UYC3	TSL:5 GENCODE basic
Rasgrp1-203	ENSMUST00000172901.7	1903	597aa	Protein coding	-	G3UWW4	TSL:5 GENCODE basic
Rasgrp1-204	ENSMUST00000173252.1	1750	546aa	Protein coding	-	G3UZQ7	TSL:5 GENCODE basic
Rasgrp1-206	ENSMUST00000174770.7	2138	481aa	Nonsense mediated decay	-	G3UYP5	TSL:5
Rasgrp1-202	ENSMUST00000110898.1	1260	No protein	Retained intron	-	-	TSL:5

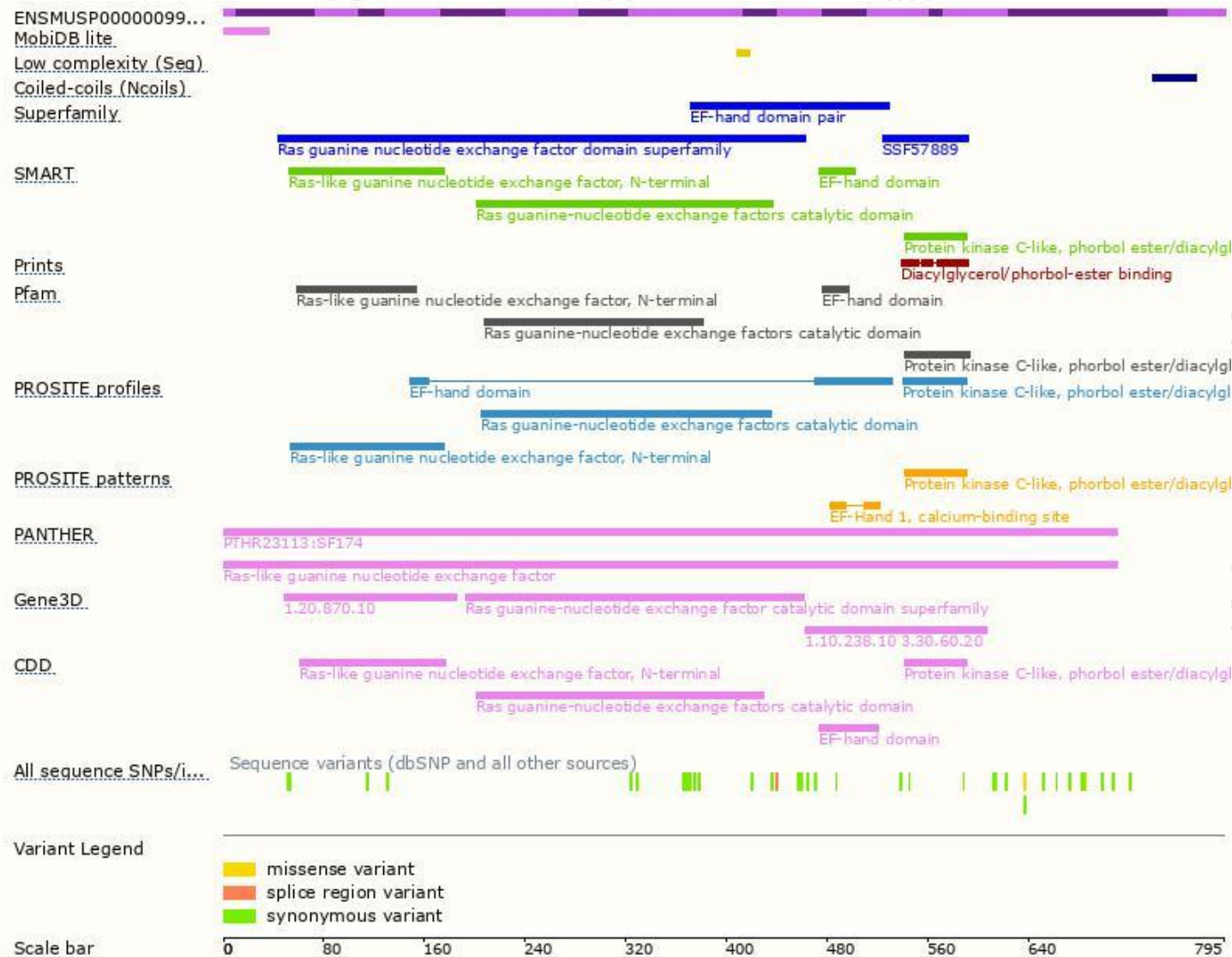
The strategy is based on the design of *Rasgrp1-201* transcript,The transcription is shown below



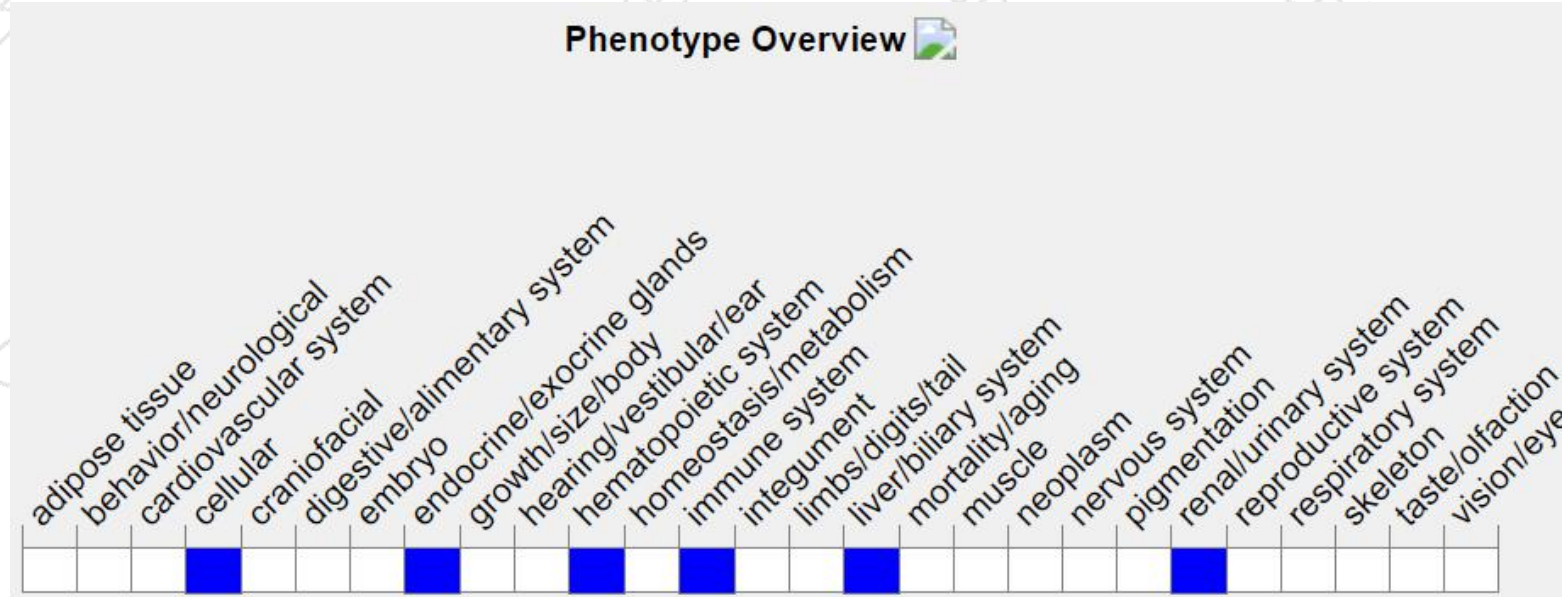
Genomic location distribution



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, Homozygotes for spontaneous and targeted null mutations exhibit a lymphoproliferative autoimmune syndrome in which T cells fail to activate Ras or proliferate after antigen exposure, defects in positive selection, and enlarged spleen and lymph nodes.

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

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