

Mrtfa Cas9-CKO Strategy

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Project Overview

Project Name

Mrtfa

Project type

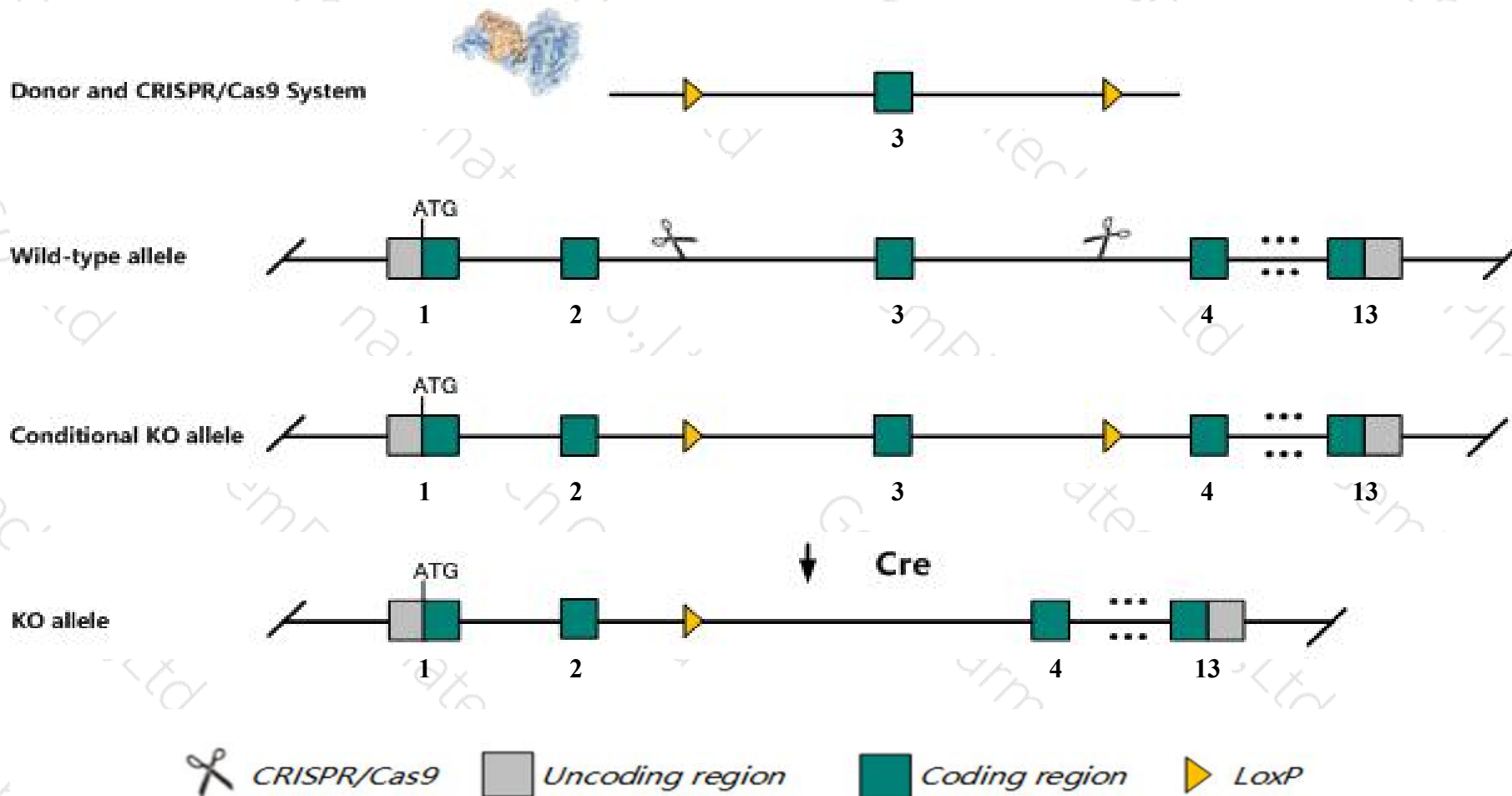
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Mrtfa* gene has 12 transcripts. According to the structure of *Mrtfa* gene, exon3 of *Mrtfa-201*(ENSMUST00000109579.8) transcript is recommended as the knockout region. The region contains 56bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Mrtfa* 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, mice homozygous for a null allele exhibit impaired mammary myoepithelial cell differentiation and fail to eject milk and productively nurse their offspring. Mice homozygous for another null allele show partial embryonic lethality caused by myocardial necrosis as well as mammary gland dysfunction.
- The *Mrtfa* gene is located on the Chr15. 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)

Mrtfa myocardin related transcription factor A [Mus musculus (house mouse)]

Gene ID: 223701, updated on 13-Mar-2020

Summary



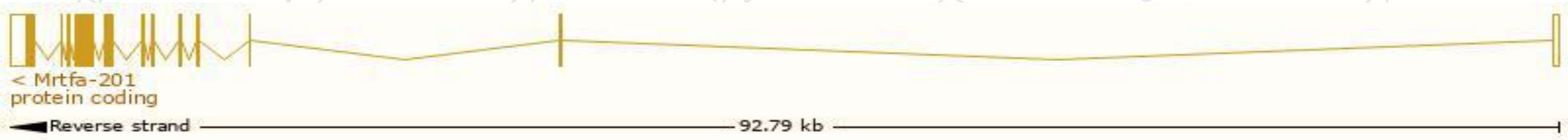
Official Symbol	Mrtfa provided by MGI
Official Full Name	myocardin related transcription factor A provided by MGI
Primary source	MGI:MGI:2384495
See related	Ensembl:ENSMUSG00000042292
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AMKL, Bsac, Mal, Mkl1, Mrtf-A
Expression	Ubiquitous expression in spleen adult (RPKM 34.3), thymus adult (RPKM 29.0) and 27 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

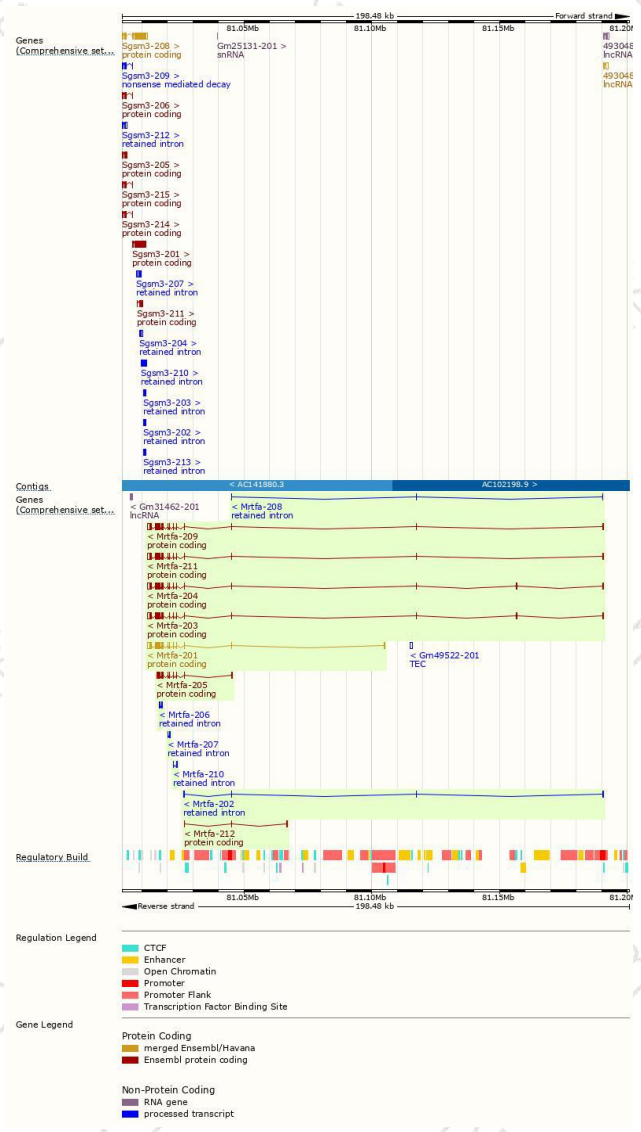
The gene has 12 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Mrtfa-209	ENSMUST00000149582.7	4366	929aa	Protein coding	CCDS49674	Q8K4J6	TSL:1 GENCODE basic
Mrtfa-201	ENSMUST00000109579.8	4154	964aa	Protein coding	CCDS37147	Q8K4J6	TSL:1 GENCODE basic APPRIS P1
Mrtfa-204	ENSMUST00000134469.8	4392	1029aa	Protein coding	-	Q8K4J6	TSL:5 GENCODE basic
Mrtfa-211	ENSMUST00000239114.1	4366	1029aa	Protein coding	-	-	GENCODE basic
Mrtfa-203	ENSMUST00000131235.8	4222	979aa	Protein coding	-	D3YUI2	TSL:5 GENCODE basic
Mrtfa-205	ENSMUST00000135047.1	2367	705aa	Protein coding	-	D3YUG5	CDS 3' incomplete TSL:1
Mrtfa-212	ENSMUST00000239267.1	429	108aa	Protein coding	-	-	CDS 3' incomplete
Mrtfa-202	ENSMUST00000123243.7	849	No protein	Retained intron	-	-	TSL:2
Mrtfa-206	ENSMUST00000138769.1	702	No protein	Retained intron	-	-	TSL:2
Mrtfa-207	ENSMUST00000145779.1	627	No protein	Retained intron	-	-	TSL:3
Mrtfa-208	ENSMUST00000147921.1	500	No protein	Retained intron	-	-	TSL:3
Mrtfa-210	ENSMUST00000154797.1	499	No protein	Retained intron	-	-	TSL:3

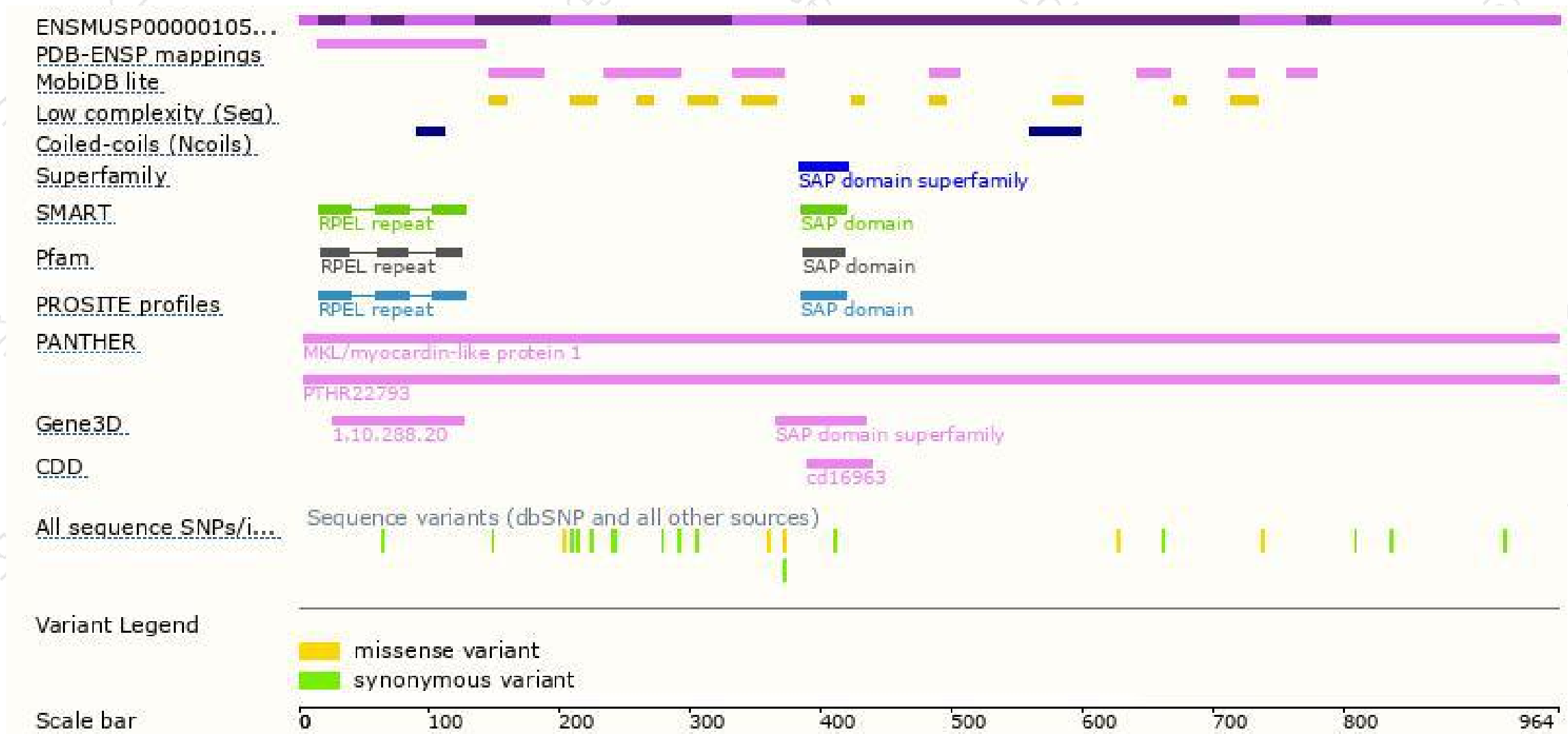
The strategy is based on the design of *Mrtfa-201* transcript,the transcription is shown below:



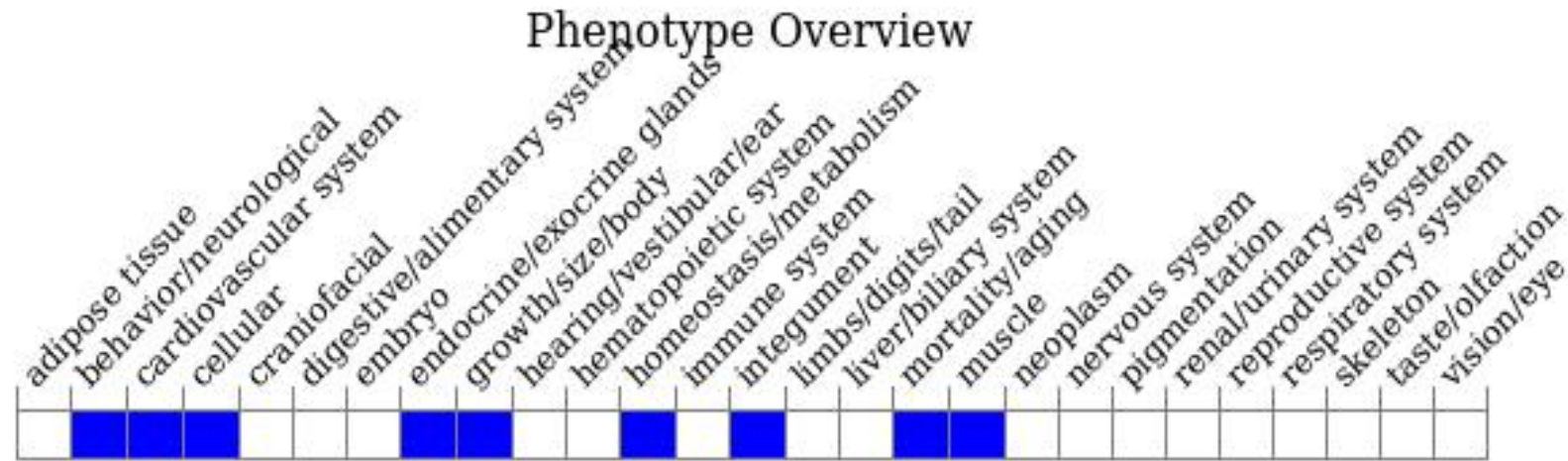
Genomic location distribution



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, mice homozygous for a null allele exhibit impaired mammary myoepithelial cell differentiation and fail to eject milk and productively nurse their offspring. Mice homozygous for another null allele show partial embryonic lethality caused by myocardial necrosis as well as mammary gland dysfunction.

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

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