Bcas3 Cas9-CKO Strategy

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Project Overview
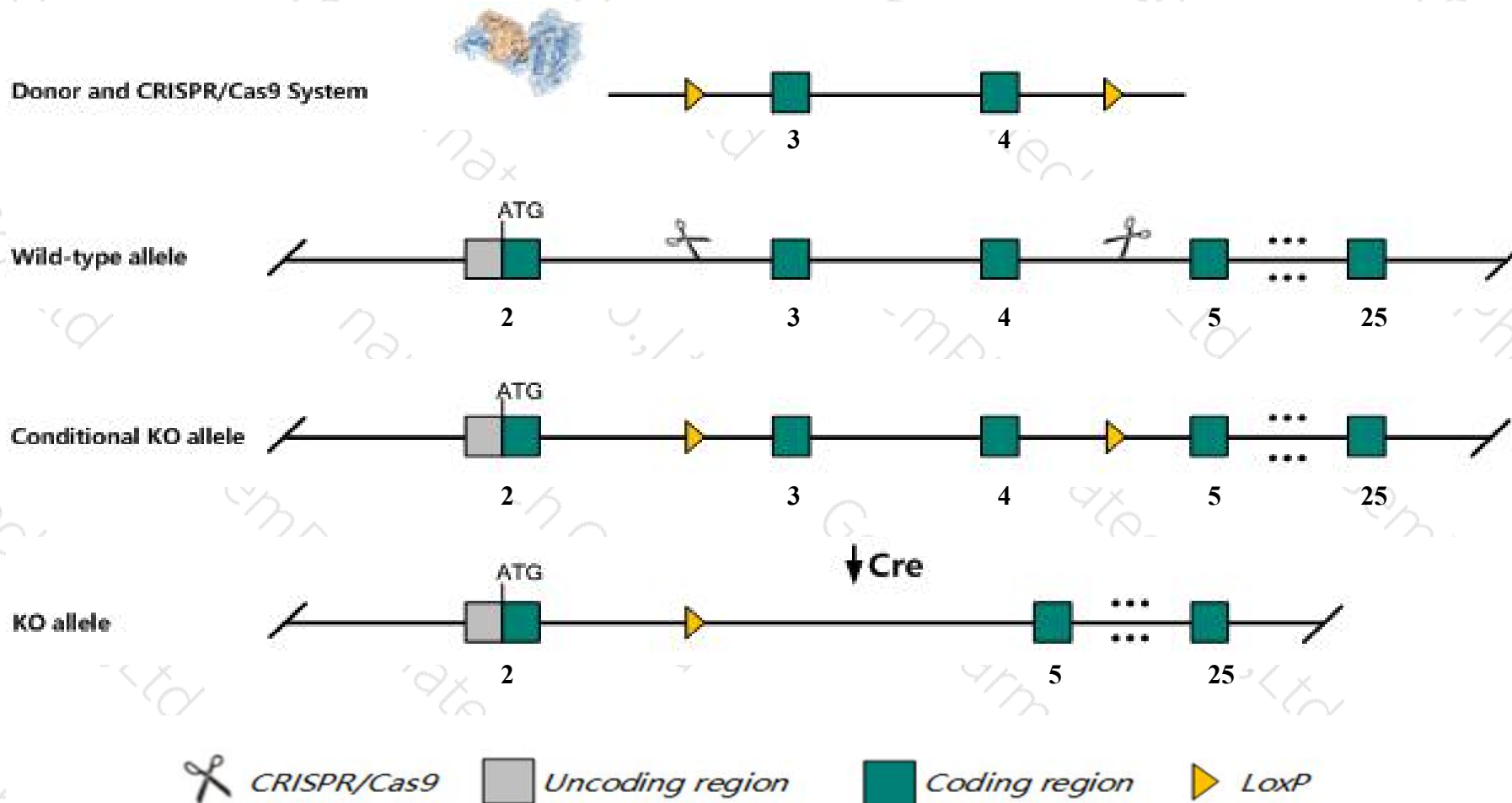
Project Name	*Bcas3*
Project type	Cas9-CKO
Strain background	C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Bcas3* gene. The schematic diagram is as follows:

Technical routes

➤ The *Bcas3* gene has 16 transcripts. According to the structure of *Bcas3* gene, exon3-exon4 of *Bcas3-201* (ENSMUST00000074875.10) transcript is recommended as the knockout region.The region contains 131bp coding sequence. Knock out the region will result in disruption of protein function.

➤ In this project we use CRISPR/Cas9 technology to modify *Bcas3* 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,Mice homozygous for a knock-out allele or a conditional allele activated

  in endothelial cells exhibit embryonic lethality with abnormal development and cardiovascular patterning.

➤ Transcript 203,212,213,215,216 CDS 5' incomplete the influences is unknown. Transcript 209 CDS 3' incomplete the

influences is unknown.

➤ The *Bcas3* gene is located on the Chr11. 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）

Bcas3 breast carcinoma amplified sequence 3 [Mus musculus (house mouse)]

Gene ID: 192197, updated on 13-Mar-2020

▲Summary

Official Symbol	Bcas3 provided by MGI
Official Full Name	breast carcinoma amplified sequence 3 provided by MGI
Primary source	MGI:MGI:2385848
See related	Ensembl:ENSMUSG00000059439
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	1500019F07Rik, 2610028P08Rik, AU021018, BC028339, K20D4, rudhira
Expression	Ubiquitous expression in lung adult (RPKM 3.3), kidney adult (RPKM 3.3) and 28 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

The gene has 16 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Bcas3-201	ENSMUST00000074875.10	2814	928aa	Protein coding	CCDS25195	Q8CCN5	TSL:5 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 P3
Bcas3-202	ENSMUST00000092821.9	2769	913aa	Protein coding	CCDS48873	Q0VF62	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 ALT2
Bcas3-205	ENSMUST00000108061.7	3810	943aa	Protein coding	-	B1AR74	TSL:5 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 ALT2
Bcas3-206	ENSMUST00000108062.7	3724	943aa	Protein coding	-	B1AR74	TSL:5 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 ALT2
Bcas3-204	ENSMUST00000108056.7	2925	527aa	Protein coding	-	Q8CCN5	TSL:1 GENCODE basic
Bcas3-216	ENSMUST00000154396.7	2789	693aa	Protein coding	-	F7C0F2	CDS 5' incomplete TSL:5
Bcas3-212	ENSMUST00000142596.7	2141	477aa	Protein coding	-	F7CPM1	CDS 5' incomplete TSL:5
Bcas3-203	ENSMUST00000092822.4	2085	458aa	Protein coding	-	B1AR76	CDS 5' incomplete TSL:5
Bcas3-213	ENSMUST00000144276.1	604	165aa	Protein coding	-	Q5SWI1	CDS 5' incomplete TSL:3
Bcas3-209	ENSMUST00000138423.7	414	120aa	Protein coding	-	B1B0B3	CDS 3' incomplete TSL:5
Bcas3-215	ENSMUST00000149662.1	412	86aa	Protein coding	-	F6VJ09	CDS 5' incomplete TSL:1
Bcas3-207	ENSMUST00000130343.7	1252	No protein	Processed transcript	-	-	TSL:1
Bcas3-214	ENSMUST00000145281.1	620	No protein	Processed transcript	-	-	TSL:3
Bcas3-208	ENSMUST00000133194.1	594	No protein	Processed transcript	-	-	TSL:3
Bcas3-210	ENSMUST00000140215.1	391	No protein	Processed transcript	-	-	TSL:2
Bcas3-211	ENSMUST00000140562.1	253	No protein	Processed transcript	-	-	TSL:2

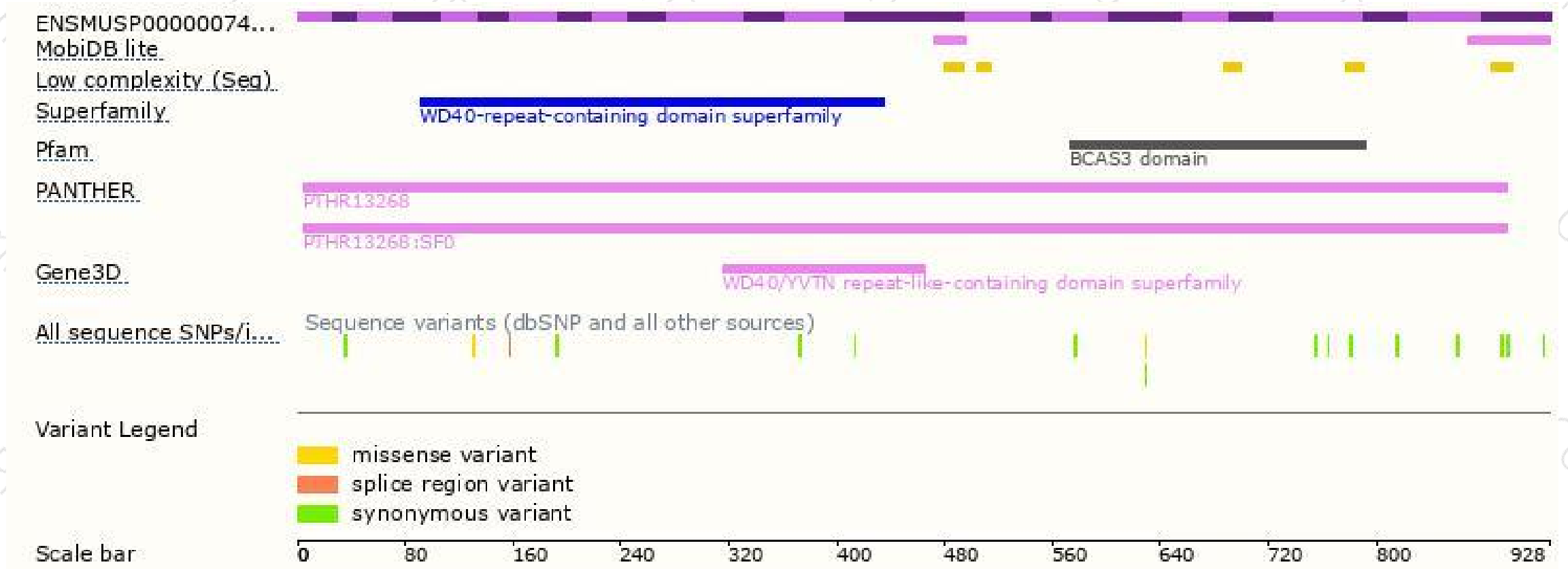The strategy is based on the design of *Bcas3-201* transcript,The transcription is shown below
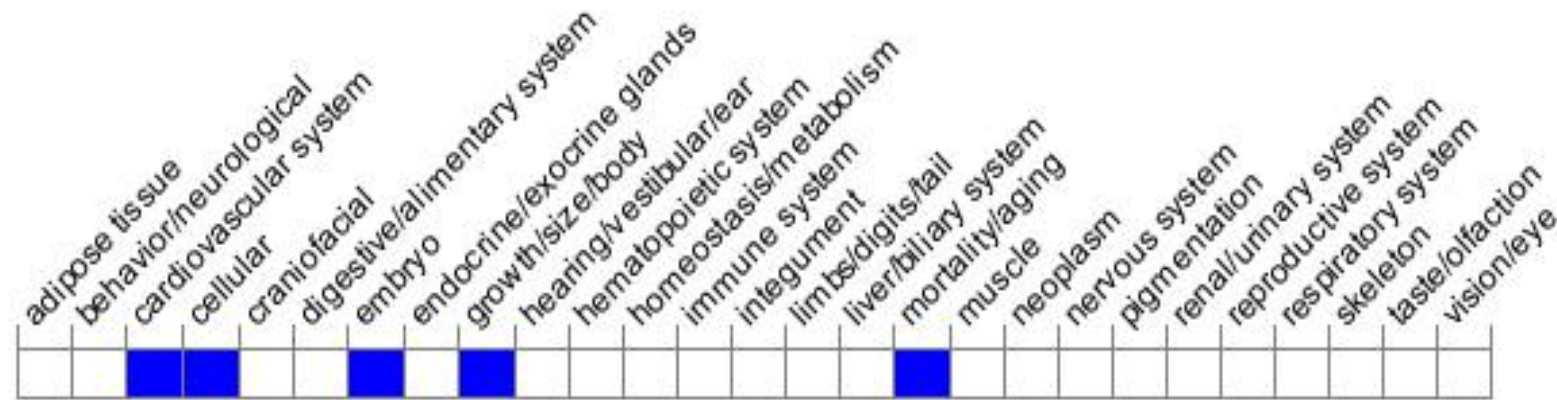
Genomic location distribution
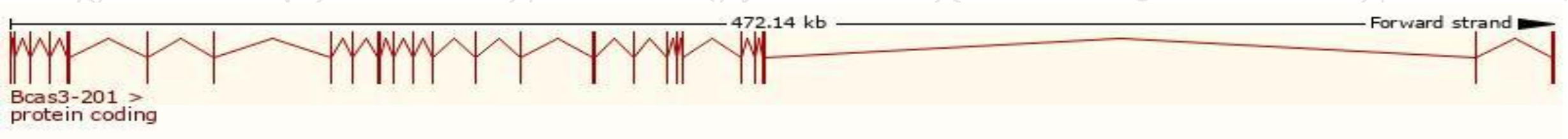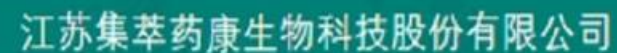
Protein domain

Mouse phenotype description(MGI)

Phenotype Overview

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 knock-out allele or a conditional allele activated in endothelial cells exhibit embryonic lethality with abnormal development and cardiovascular patterning.

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