

Bad Cas9-CKO Strategy

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Reviewer:

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Design Date:

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Project Overview

Project Name

Bad

Project type

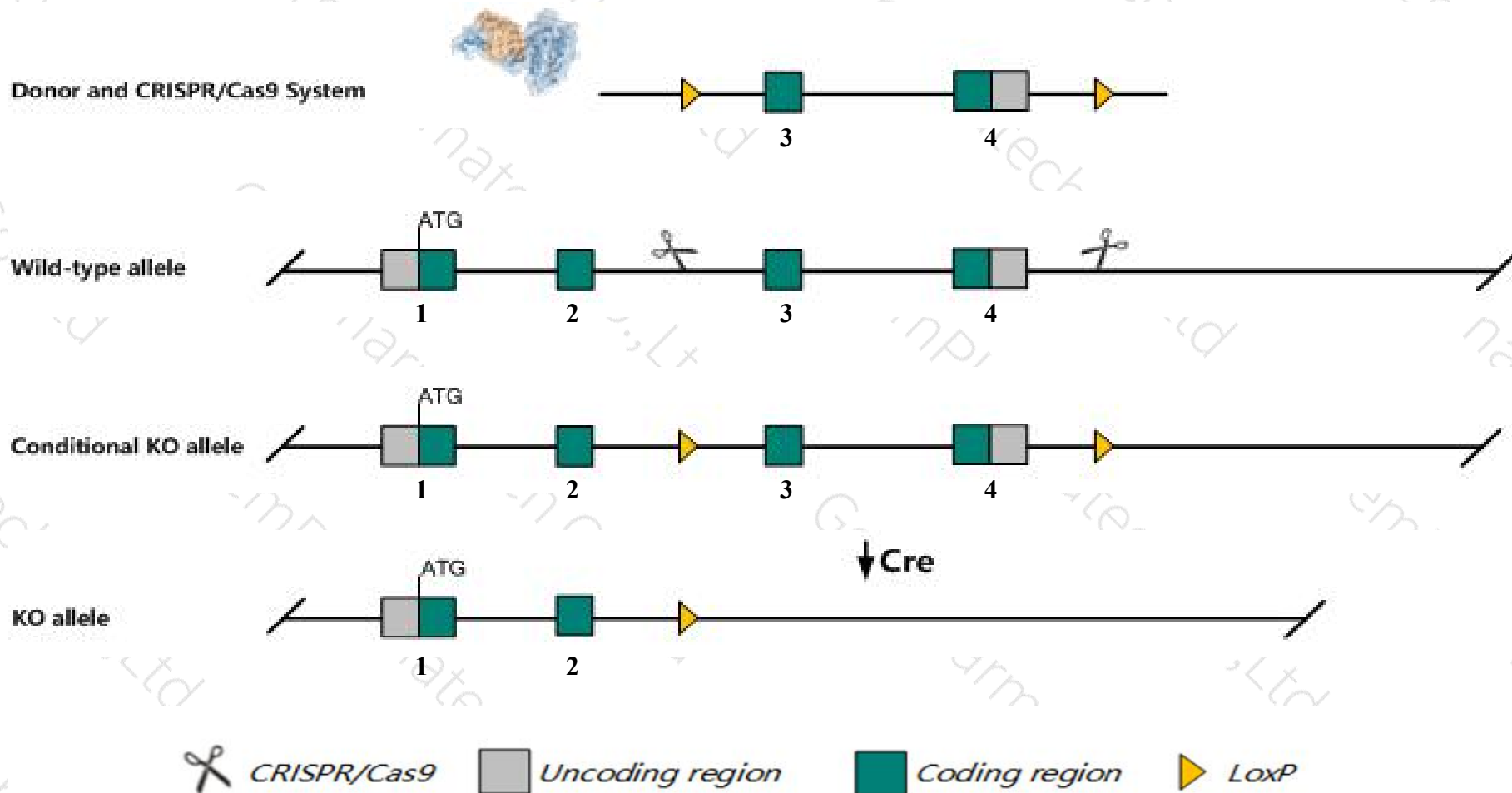
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Bad* gene has 9 transcripts. According to the structure of *Bad* gene, exon3-exon4 of *Bad-201* (ENSMUST00000025910.12) transcript is recommended as the knockout region. The region contains 317bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Bad* 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 disruptions in this gene are grossly normal and fertile. Both T and B cells show increased sensitivity to gamma irradiation.
- The *Bad* gene is located on the Chr19. 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)

Bad BCL2-associated agonist of cell death [Mus musculus (house mouse)]

Gene ID: 12015, updated on 31-Jan-2019

Summary



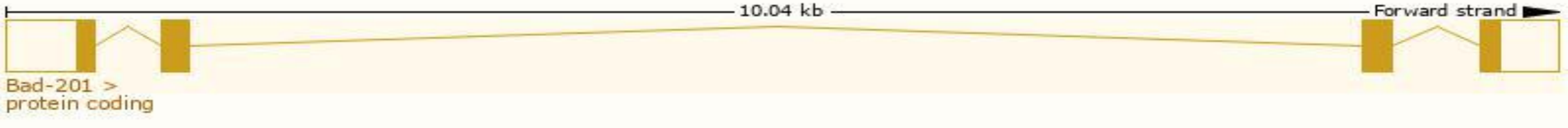
Official Symbol	Bad provided by MGI
Official Full Name	BCL2-associated agonist of cell death provided by MGI
Primary source	MGI:MGI:1096330
See related	Ensembl:ENSMUSG000000024959
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI325008, Bbc2
Expression	Ubiquitous expression in colon adult (RPKM 22.0), duodenum adult (RPKM 20.4) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

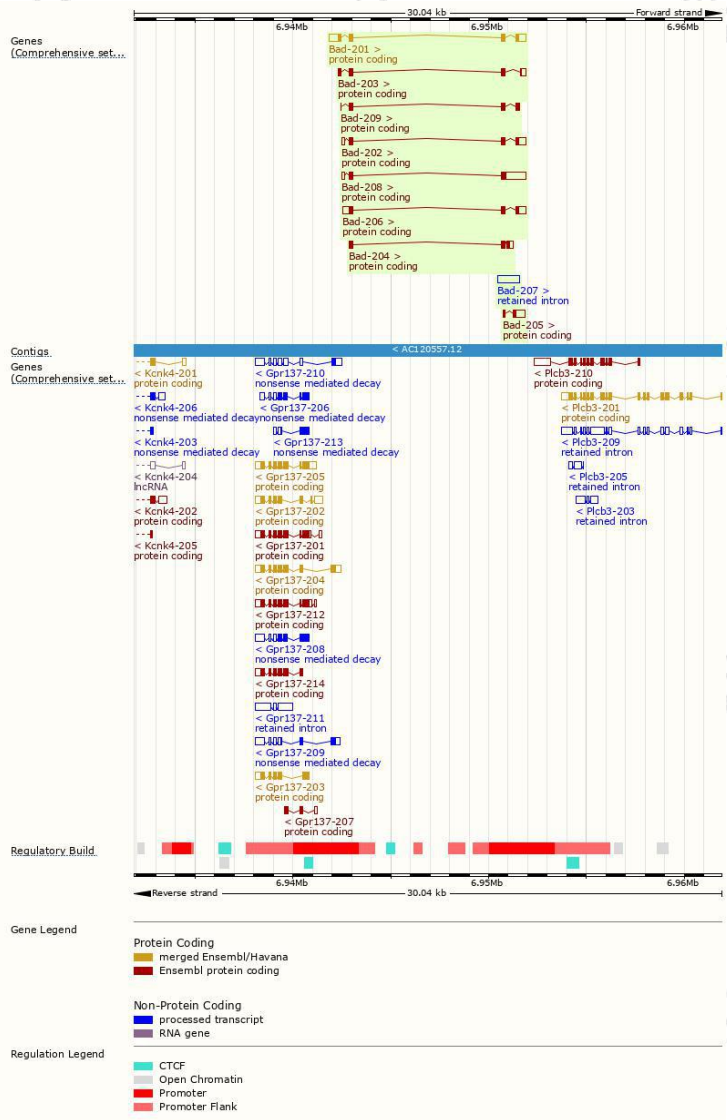
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Bad-201	ENSMUST00000025910.12	1452	204aa	Protein coding	CCDS29513	Q61337	TSL:1 GENCODE basic APPRIS P3
Bad-206	ENSMUST00000235240.1	1169	162aa	Protein coding	CCDS70924	-	GENCODE basic APPRIS ALT2
Bad-202	ENSMUST00000113423.9	990	162aa	Protein coding	CCDS70924	Q3U9H3	TSL:1 GENCODE basic APPRIS ALT2
Bad-209	ENSMUST00000238184.1	540	162aa	Protein coding	CCDS70924	-	GENCODE basic APPRIS ALT2
Bad-208	ENSMUST00000237334.1	1533	131aa	Protein coding	-	-	GENCODE basic
Bad-203	ENSMUST00000113426.7	740	178aa	Protein coding	-	D3YZR8	TSL:5 GENCODE basic
Bad-205	ENSMUST00000145463.1	668	63aa	Protein coding	-	F6S493	CDS 5' incomplete TSL:3
Bad-204	ENSMUST00000141410.1	663	161aa	Protein coding	-	F7ABX5	CDS 5' incomplete TSL:2
Bad-207	ENSMUST00000237244.1	1107	No protein	Retained intron	-	-	

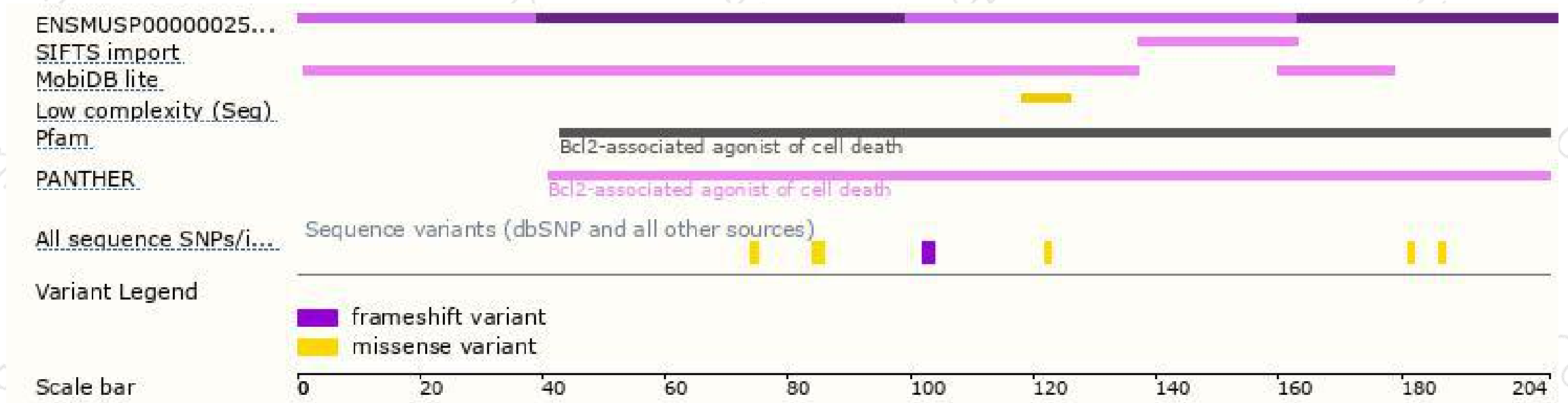
The strategy is based on the design of *Bad-201* transcript,The transcription is shown below



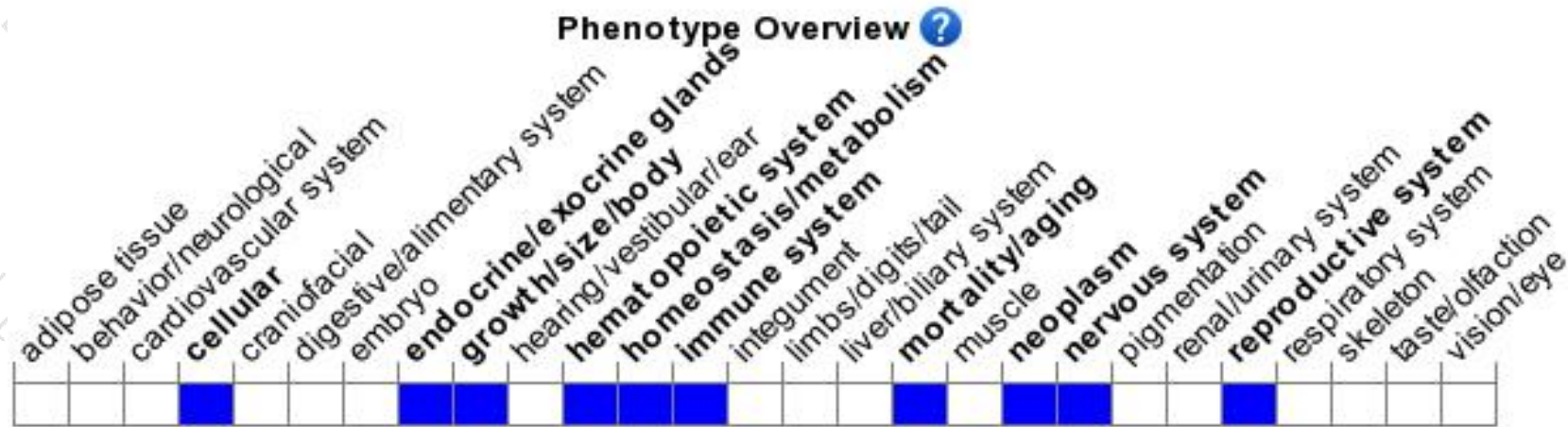
Genomic location distribution



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/>).

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

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