

Rad50 Cas9-CKO Strategy

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Design Date: 2019/10/11

Project Overview



Project Name

Rad50

Project type

Cas9-CKO

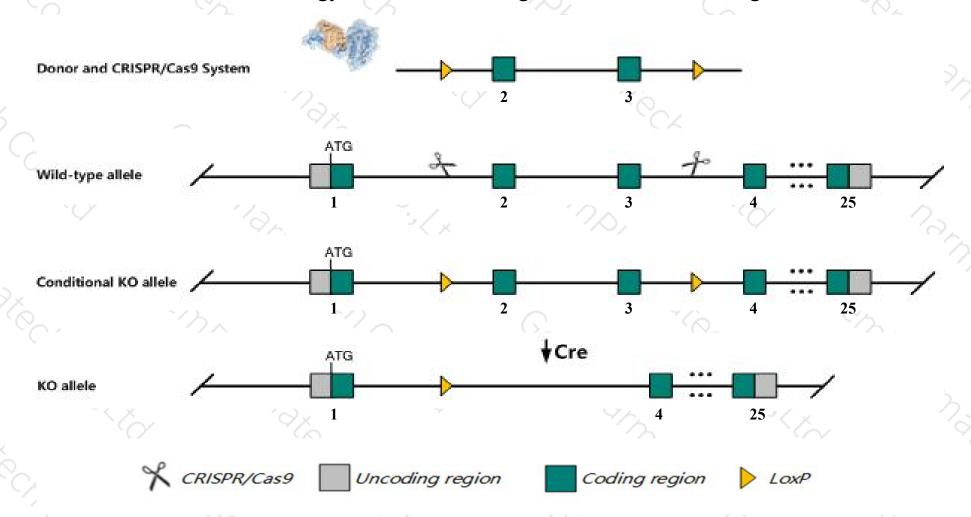
Strain background

C57BL/6JGpt

Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Rad50* gene. The schematic diagram is as follows:



Technical routes



- ➤ The *Rad50* gene has 5 transcripts. According to the structure of *Rad50* gene, exon2-exon3 of *Rad50-201* (ENSMUST00000020649.13) transcript is recommended as the knockout region. The region contains 236bp coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Rad50* 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, Homozygotes for a targeted hypomorphic mutation exhibit growth defects, predisposition toward cancer, progressive loss of hematopoietic and spermatogenic stem cells, and lethality due to bone marrow depletion. A null mutation results in embryonic death.
- The distance between exon 3 of Rad50 and Gm22275 is about 0.6kb, and the knockout of Rad50 may affect the functional of Gm22275.
- The *Rad50* 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)



Rad50 RAD50 double strand break repair protein [Mus musculus (house mouse)]

Gene ID: 19360, updated on 12-Aug-2019

Summary

△ ?

Official Symbol Rad50 provided by MGI

Official Full Name RAD50 double strand break repair protein provided by MGI

Primary source MGI:MGI:109292

See related Ensembl: ENSMUSG00000020380

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 Mrell; Rad50l

Expression Ubiquitous expression in bladder adult (RPKM 4.4), CNS E11.5 (RPKM 4.2) and 28 other tissues See more

Orthologs human all

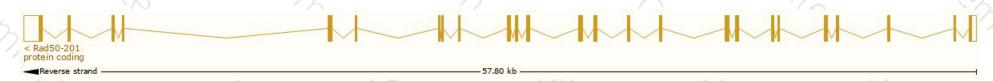
Transcript information (Ensembl)



The gene has 5 transcripts, all transcripts are shown below:

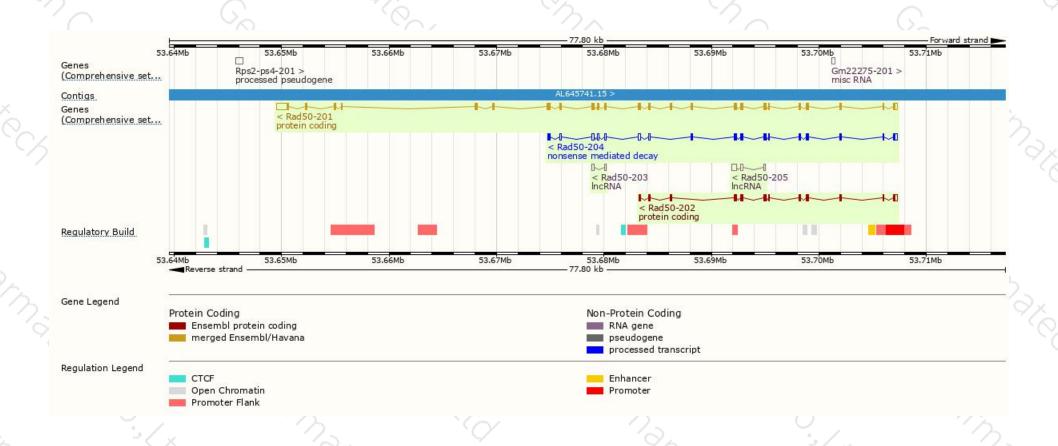
Name	Transcript ID	bp 🍦	Protein +	Biotype	CCDS .	UniProt 🍦	Flags
Rad50-201	ENSMUST00000020649.13	5153	<u>1312aa</u>	Protein coding	CCDS24684@	Q5SV02 ₽	TSL:1 GENCODE basic APPRIS P1
Rad50-202	ENSMUST00000124352.1	2211	<u>657aa</u>	Protein coding	-	<u>A8Y5I3</u> @	CDS 3' incomplete TSL:1
Rad50-204	ENSMUST00000128483.7	3085	<u>551aa</u>	Nonsense mediated decay	-	E9PUJ2₽	TSL:1
Rad50-205	ENSMUST00000152598.1	693	No protein	IncRNA	-	83	TSL:2
Rad50-203	ENSMUST00000126121.1	404	No protein	IncRNA	(44)	-	TSL:2

The strategy is based on the design of Rad50-201 transcript, The transcription is shown below



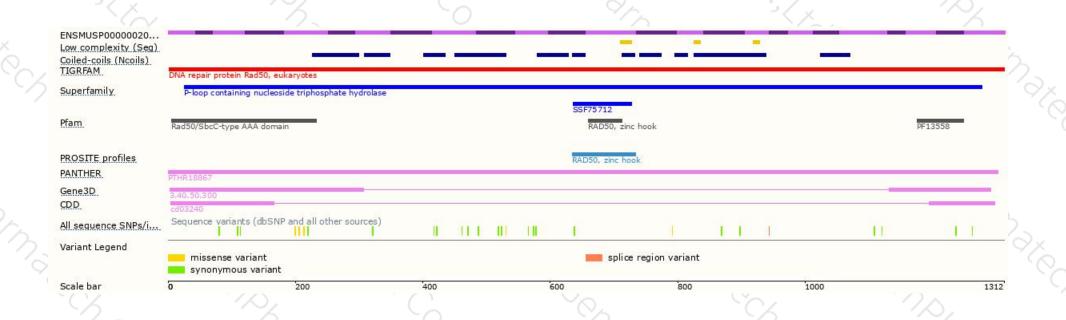
Genomic location distribution





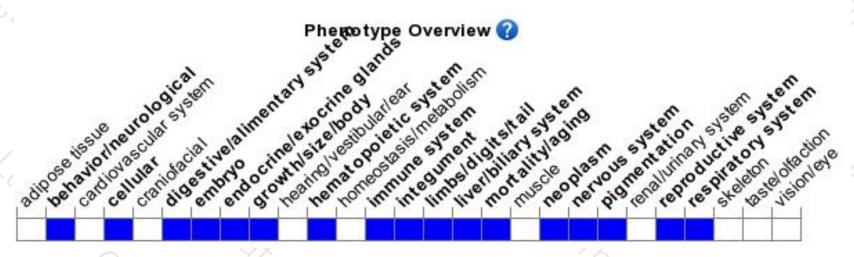
Protein domain





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 a targeted hypomorphic mutation exhibit growth defects, predisposition toward cancer, progressive loss of hematopoietic and spermatogenic stem cells, and lethality due to bone marrow depletion. A null mutation results in embryonic death.



If you have any questions, you are welcome to inquire. Tel: 400-9660890





