

Prkn Cas9-CKO Strategy

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Reviewer:

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Project Overview

Project Name

Prkn

Project type

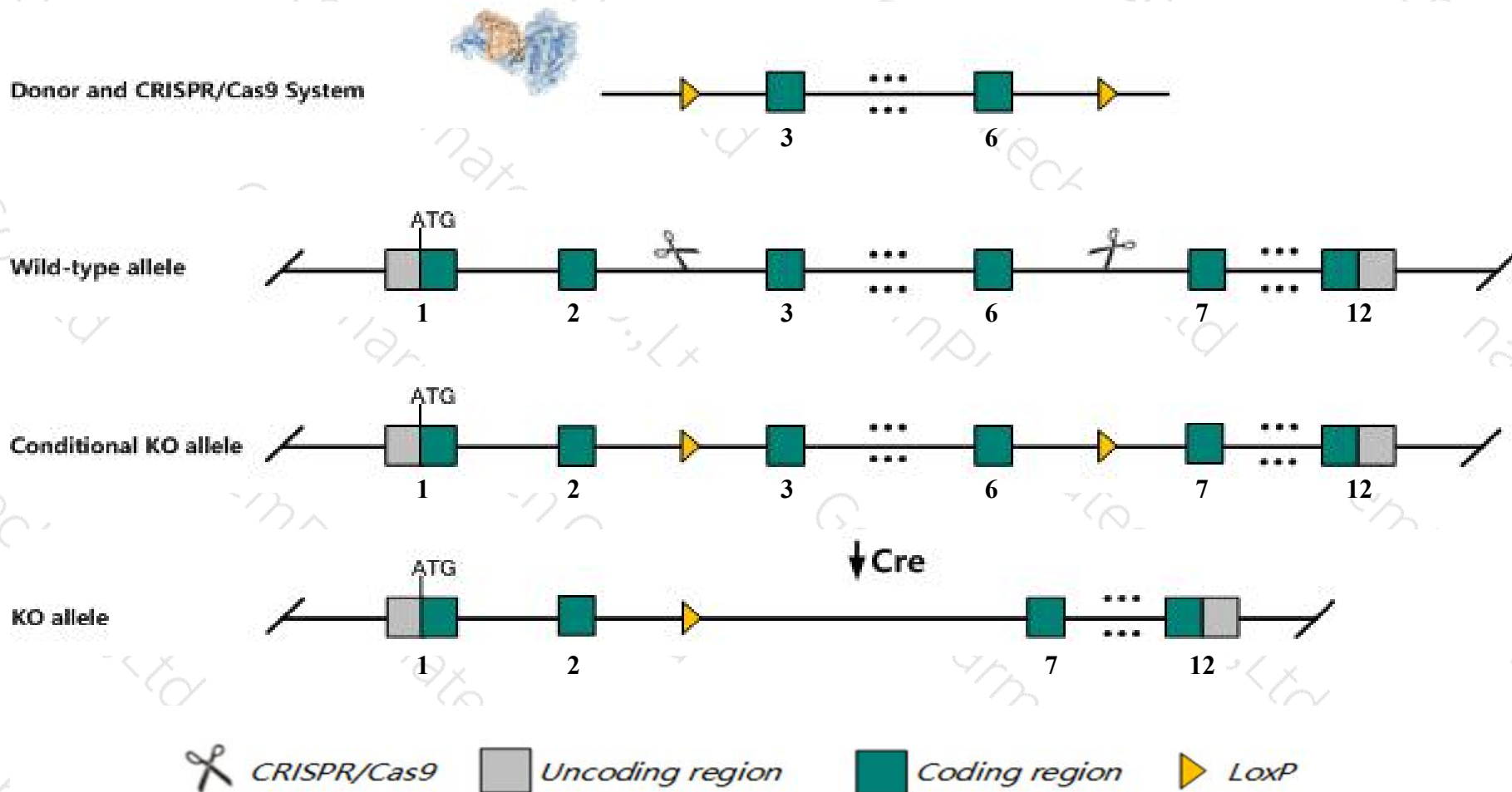
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

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



Technical routes

- The *Prkn* gene has 16 transcripts. According to the structure of *Prkn* gene, exon3-exon6 of *Prkn*-209 (ENSMUST00000191124.6) transcript is recommended as the knockout region. The region contains 560bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Prkn* 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, Dopamine and glutamate transmission are impaired in some targeted null mice, resulting in decreased exploratory behavior. These mice show decreased body weight and temperature. Park2 is inactivated as part of a large deletion in the quaking mouse, a dysmyelinating mutant with a pronounced tremor.
- Transcript *Prkn* -204 may not be affected.
- The *Prkn* gene is located on the Chr17. 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)

Prkn parkin RBR E3 ubiquitin protein ligase [Mus musculus (house mouse)]

Gene ID: 50873, updated on 9-Apr-2019

Summary



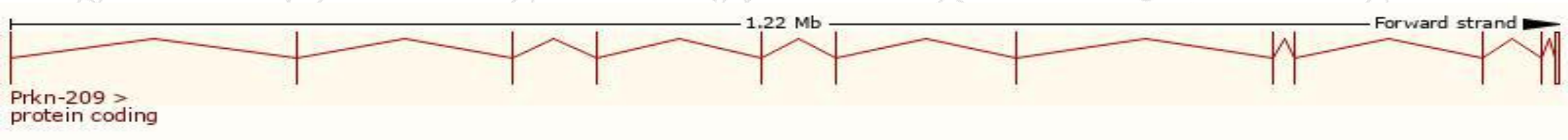
Official Symbol	Prkn provided by MGI
Official Full Name	parkin RBR E3 ubiquitin protein ligase provided by MGI
Primary source	MGI:MGI:1355296
See related	Ensembl:ENSMUSG00000023826
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Park2
Expression	Broad expression in frontal lobe adult (RPKM 1.6), cortex adult (RPKM 1.5) and 19 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

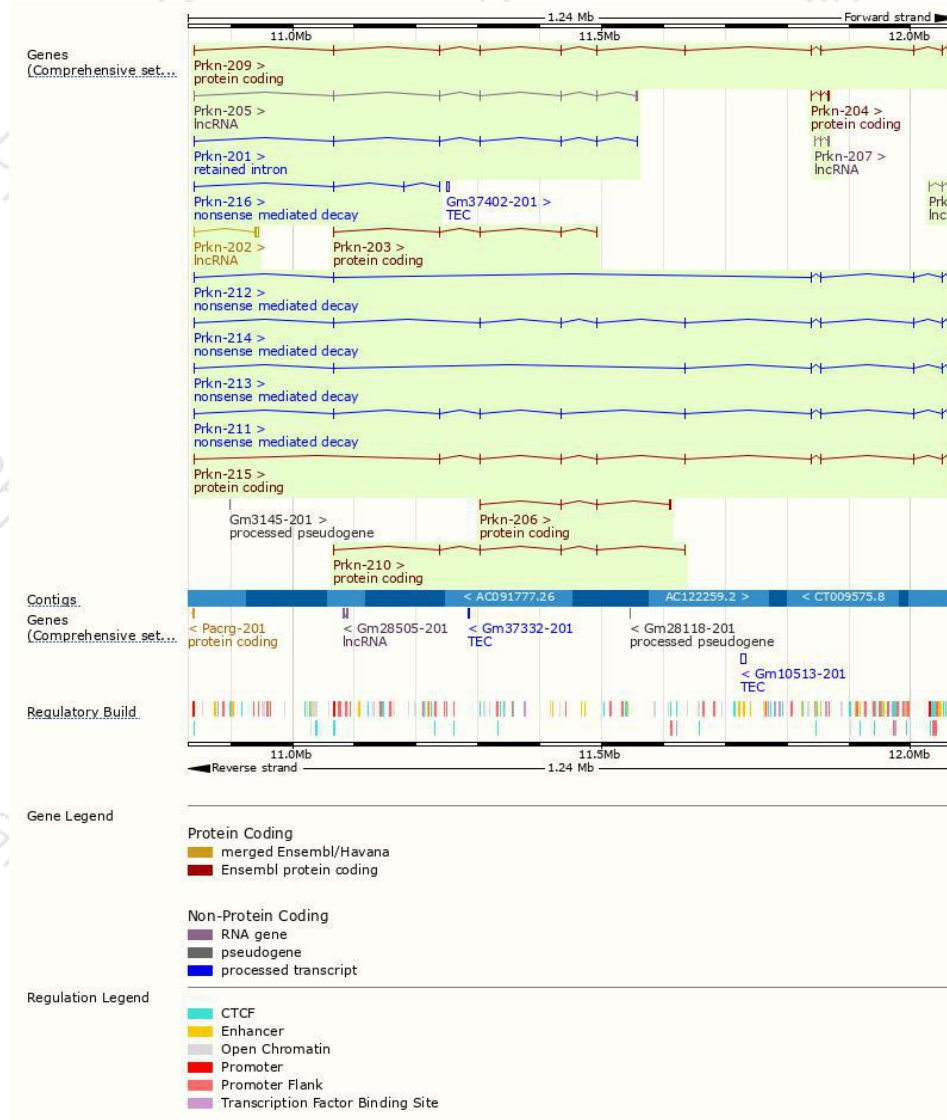
The gene has 16 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Prkn-209	ENSMUST00000191124.6	3202	464aa	Protein coding	CCDS79506	Q9WVS6	TSL:1 GENCODE basic APPRIS P1
Prkn-215	ENSMUST00000233706.1	1272	274aa	Protein coding	-	A0A3B2W489	GENCODE basic
Prkn-204	ENSMUST00000179624.7	1108	71aa	Protein coding	-	J3QMF8	TSL:5 GENCODE basic
Prkn-210	ENSMUST00000218435.1	865	288aa	Protein coding	-	A0A1W2P7W2	TSL:5 GENCODE basic
Prkn-203	ENSMUST00000168593.8	735	245aa	Protein coding	-	F6TNF2	TSL:5 GENCODE basic
Prkn-206	ENSMUST00000186406.1	607	104aa	Protein coding	-	A0A087WQJ2	CDS 5' incomplete TSL:3
Prkn-211	ENSMUST00000233294.1	1377	214aa	Nonsense mediated decay	-	B8YP90	
Prkn-214	ENSMUST00000233634.1	1252	106aa	Nonsense mediated decay	-	B8YP89	
Prkn-213	ENSMUST00000233436.1	930	65aa	Nonsense mediated decay	-	B8YP88	
Prkn-212	ENSMUST00000233411.1	793	63aa	Nonsense mediated decay	-	D3JSV6	
Prkn-216	ENSMUST00000233782.1	434	59aa	Nonsense mediated decay	-	A0A3B2W3P9	
Prkn-201	ENSMUST00000066658.12	1153	No protein	Retained intron	-	-	TSL:1
Prkn-202	ENSMUST00000097413.4	4759	No protein	lncRNA	-	-	TSL:1
Prkn-208	ENSMUST00000190894.1	2182	No protein	lncRNA	-	-	TSL:1
Prkn-205	ENSMUST00000186167.1	1420	No protein	lncRNA	-	-	TSL:1
Prkn-207	ENSMUST00000190849.1	1200	No protein	lncRNA	-	-	TSL:1

The strategy is based on the design of *Prkn-209* transcript,The transcription is shown below



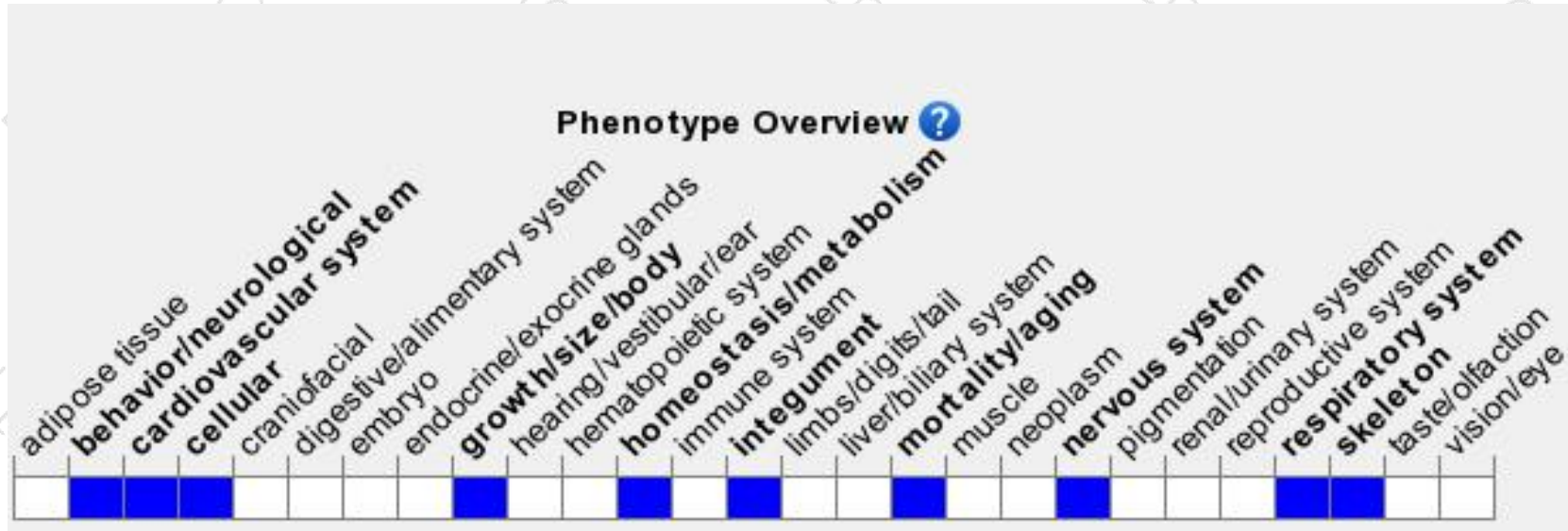
Genomic location distribution



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, Dopamine and glutamate transmission are impaired in some targeted null mice, resulting in decreased exploratory behavior. These mice show decreased body weight and temperature. Park2 is inactivated as part of a large deletion in the quaking mouse, a dysmyelinating mutant with a pronounced tremor.

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

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