Xpot Cas9-CKO Strategy

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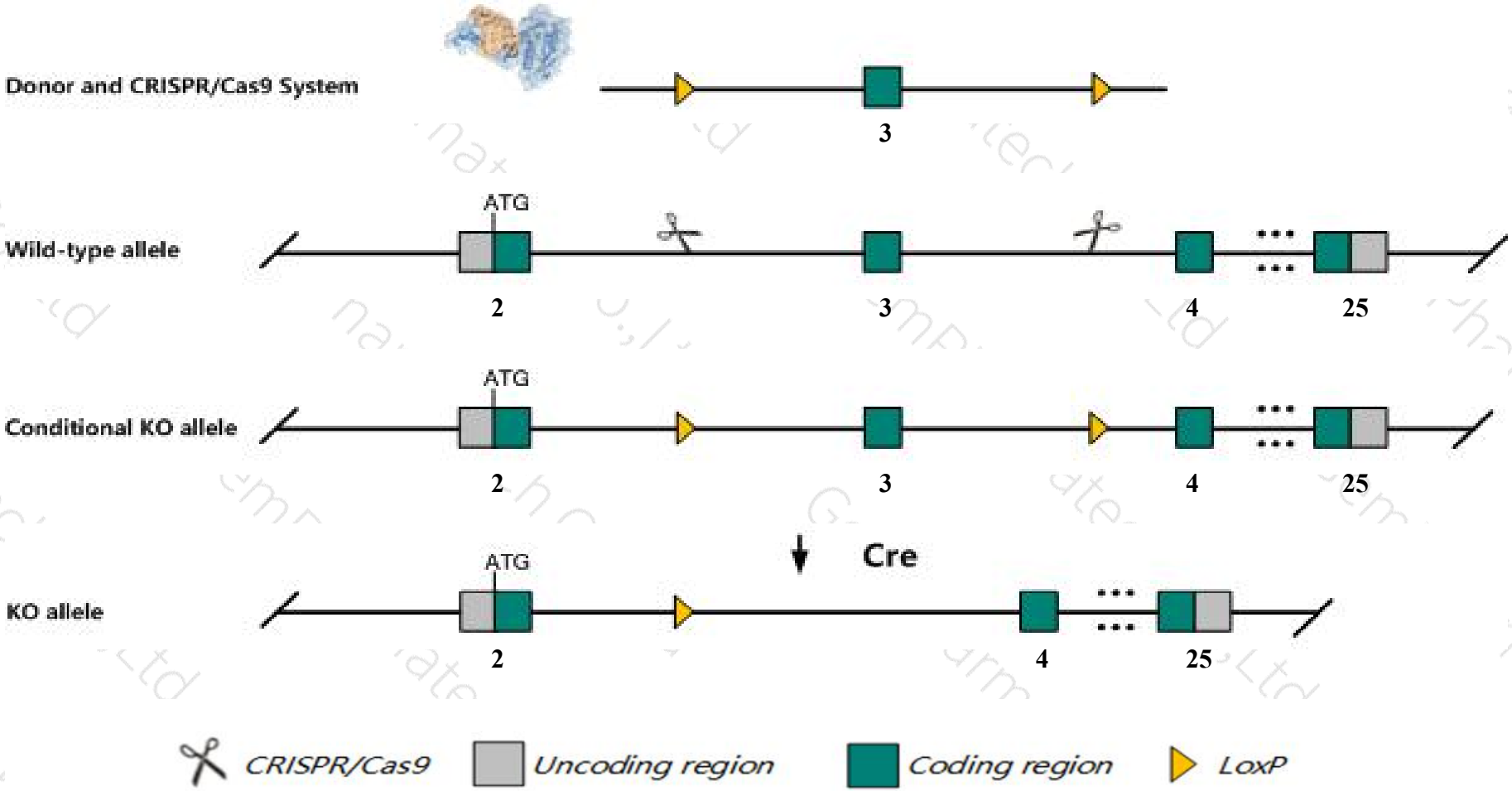
Project Overview

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

Conditional Knockout strategy

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



Technical routes

- The *Xpot* gene has 5 transcripts. According to the structure of *Xpot* gene, exon3 of *Xpot-201* (ENSMUST00000039810.7) transcript is recommended as the knockout region.The region contains 83bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Xpot* 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

➢ The *Xpot* gene is located on the Chr10. 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.

➢ The effect on transcript *Xpot*-2027205 is unknown.

➢ Transcript *Xpot*-204 may not be affected.

➢ *Gm24330* gene may be destroyed.

➢ 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）

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Xpot exportin, tRNA (nuclear export receptor for tRNAs) [*Mus musculus* (house mouse)]

Gene ID: 73192, updated on 13-Mar-2020

Summary

Official Symbol	Xpot provided by MGI
Official Full Name	exportin, tRNA (nuclear export receptor for tRNAs) provided by MGI
Primary source	MGI:MGI:1920442
See related	Ensembl:ENSMUSG00000034667
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	C79645; AI452076; EXPORTIN-T; 1110004L07Rik; 3110065H13Rik
Expression	Ubiquitous expression in limb E14.5 (RPKM 13.2), liver E14 (RPKM 10.9) and 28 other tissues See more
Orthologs	human all

Genomic context

Location: 10; 10 D2

See Xpot in Genome Data Viewer

Exon count: 26

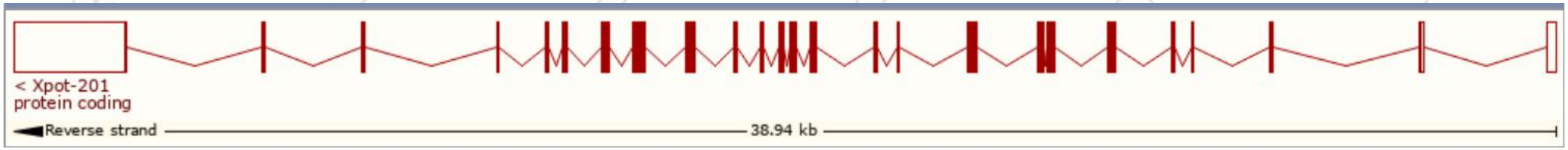
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	10	NC_000076.6 (121587380..121626506, complement)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	10	NC_000076.5 (121024436..121063372, complement)

Transcript information（Ensembl）

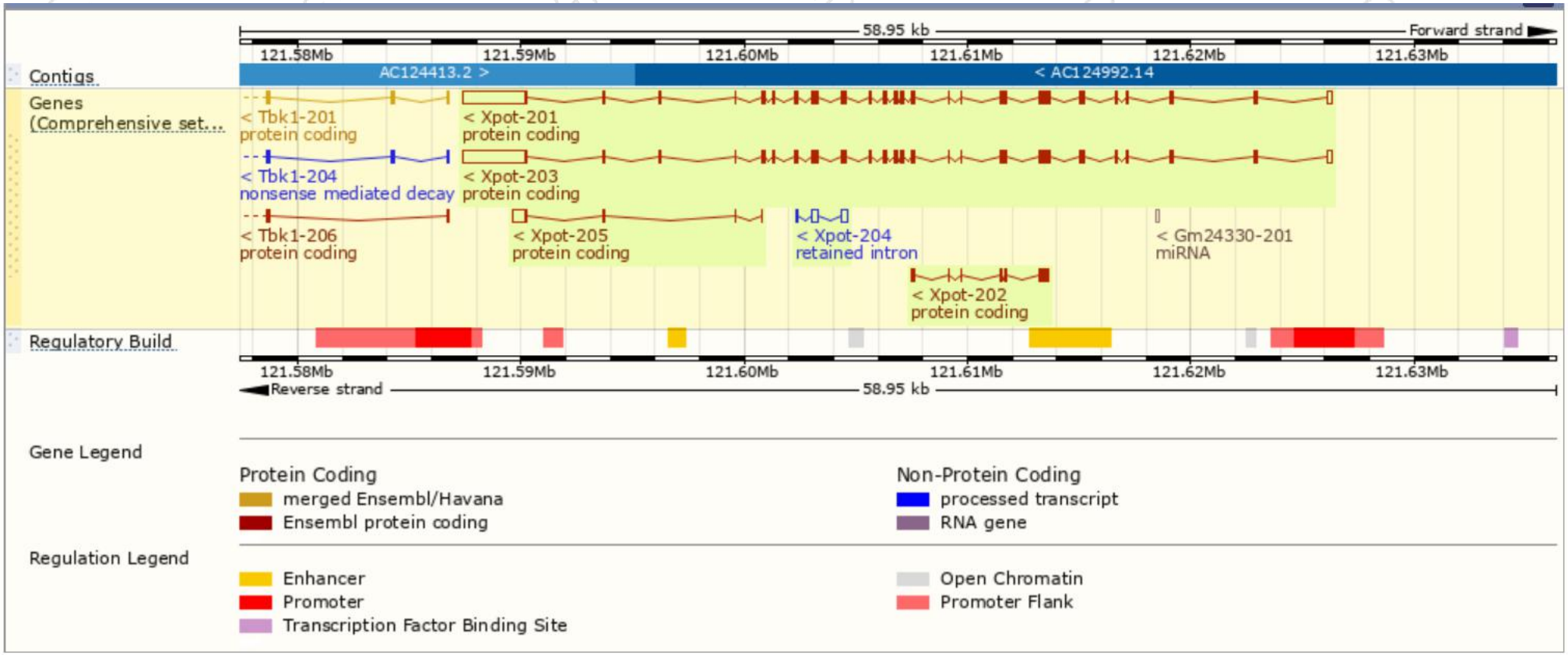
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Xpot-203	ENSMUST00000218004.1	6005	962aa	Protein coding	-	A0A1W2P7Q6	TSL:5 GENCODE basic APPRIS P1
Xpot-201	ENSMUST00000039810.7	5992	963aa	Protein coding	-	Q9CRT8	TSL:5 GENCODE basic
Xpot-205	ENSMUST00000219334.1	729	51aa	Protein coding	-	A0A1W2P6G8	CDS 5' incomplete TSL:3
Xpot-202	ENSMUST00000217865.1	618	206aa	Protein coding	-	A0A1W2P7V1	CDS 5' and 3' incomplete TSL:3
Xpot-204	ENSMUST00000218973.1	658	No protein	Retained intron	-	-	TSL:2

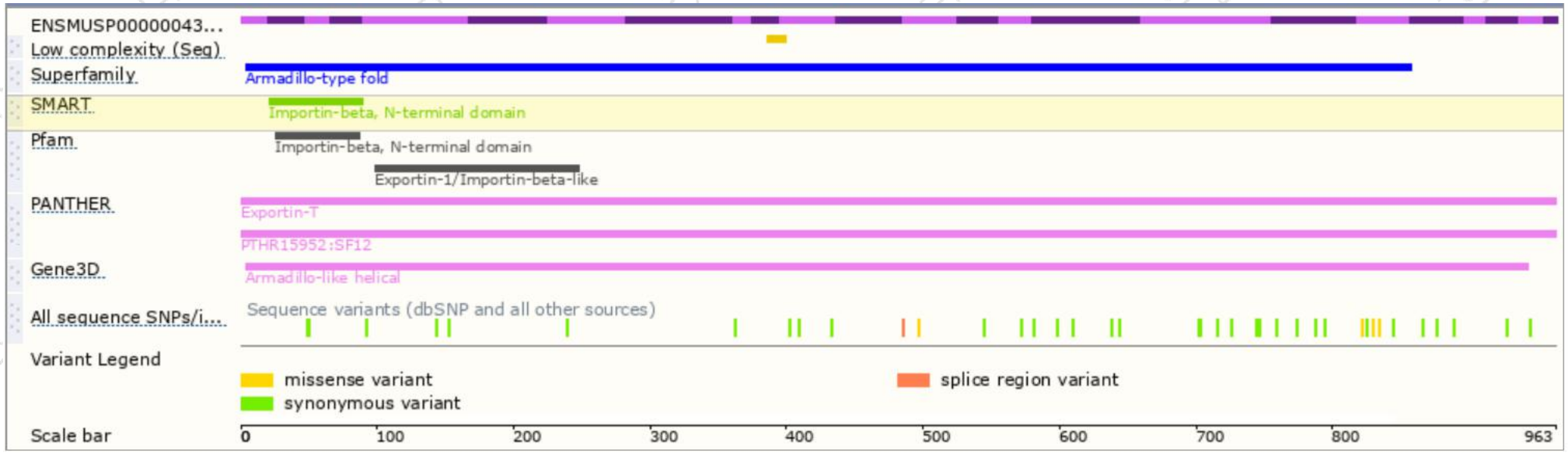
The strategy is based on the design of *Xpot-201* transcript,the transcription is shown below:



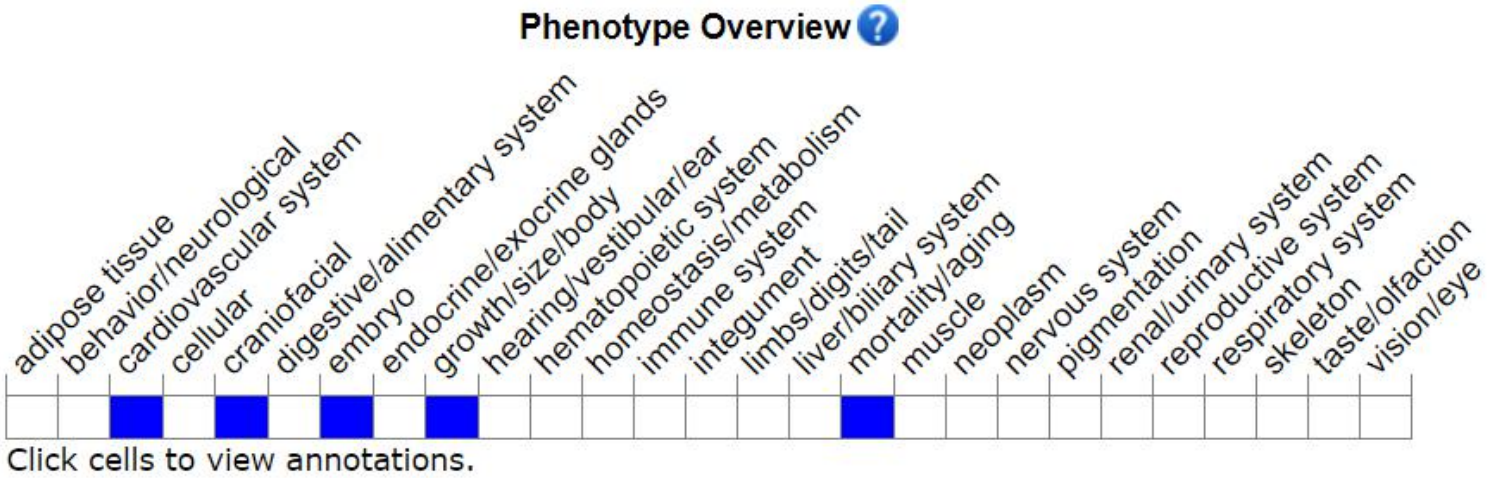
Genomic location distribution



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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