Foxo1 Cas9-CKO *Strategy*

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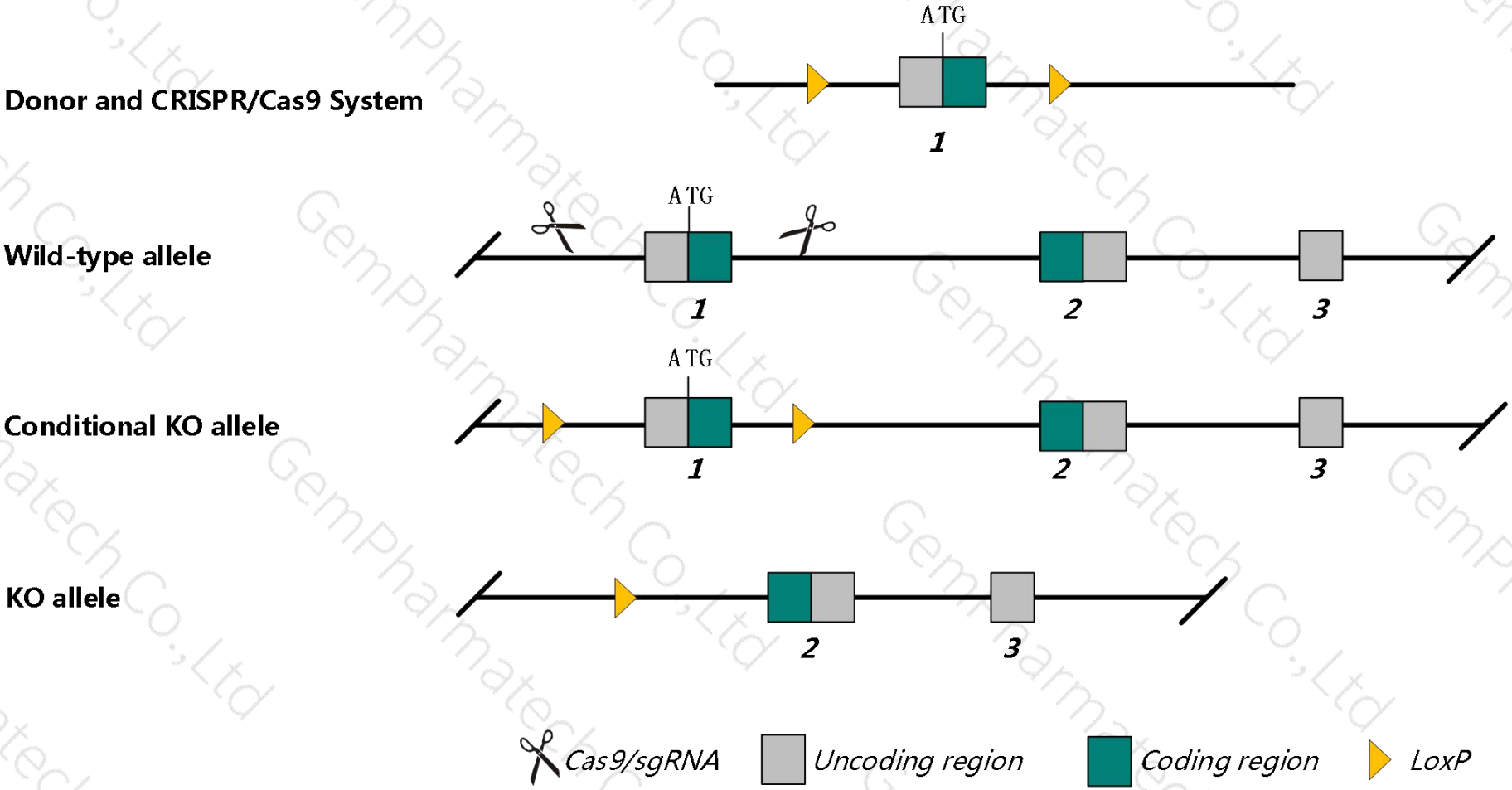
Project Overview

Project Name	***Foxo1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Foxo1* gene. The schematic diagram is as follows:



Technical routes

- The *Foxo1* gene has 1 transcripts. According to the structure of *Foxo1* gene, the predicted promoter region and exon1 of *Foxo1*-201 (ENSMUST00000053764.6) transcript is recommended as the knockout region.The region contains the predicted promoter sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Foxo1* gene. The brief process is as follows: gRNA was transcribed in vitro, donor was constructed.Cas9, gRNA 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 or cell types.

Notice

- According to the existing MGI data: Homozygous null embryos die at E10.5-E11.5 from vasculature defects. Heterozygote null mice have slightly elevated glycogen levels. Conditionally targeted homozygotes display hemangiomas or defects in nave T cell homeostasis depending on the targeted cell type.
- The *Foxo1* gene is located on the Chr3. 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 the loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information（NCBI）

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Foxo1 forkhead box O1 [*Mus musculus* (house mouse)]

Gene ID: 56458, updated on 23-Oct-2018

Summary

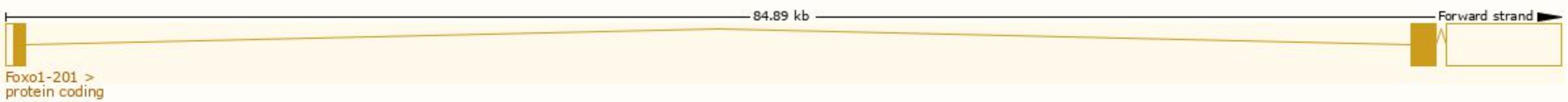
Official Symbol	Foxo1 provided by MGI
Official Full Name	forkhead box O1 provided by MGI
Primary source	MGI:MGI:1890077
See related	Ensembl:ENSMUSG00000044167 Vega:OTTMUSG00000021250
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Afxh; FKHR; Fkhr1; Foxo1a; AI876417
Expression	Broad expression in ovary adult (RPKM 82.6), spleen adult (RPKM 21.1) and 16 other tissues See more
Orthologs	human all

Transcript information （Ensembl

The gene has 1 transcripts, and all transcripts are shown below ：

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags
Foxo1-201	ENSMUST00000053764.6	8665	652aa	Protein coding	CCDS17343	Q9R1E0	NM_019739 NP_062713	TSL:1 GENCODE basic APPRIS P1

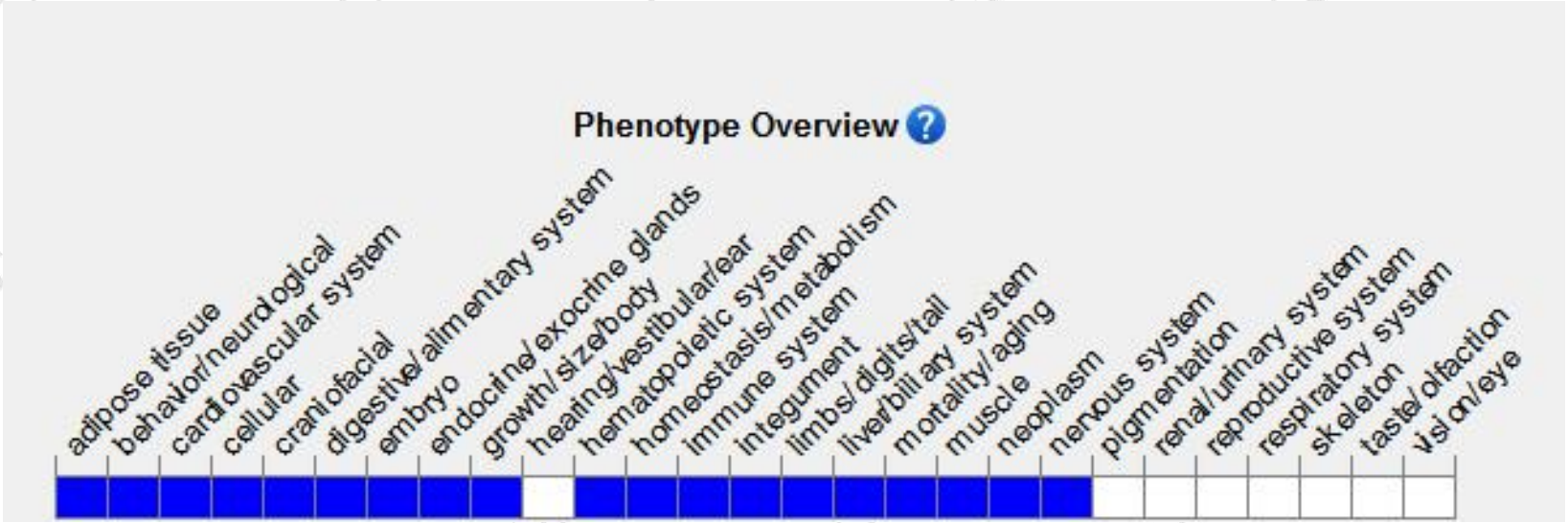
The strategy is based on the design of Foxo1-201 transcript,The transcription is shown below



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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