

# Rspo1 Cas9-KO Strategy

**Designer:** 

**Huan Wang** 

**Reviewer:** 

**Huan Fan** 

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# **Project Overview**



**Project Name** 

Rspo1

**Project type** 

Cas9-KO

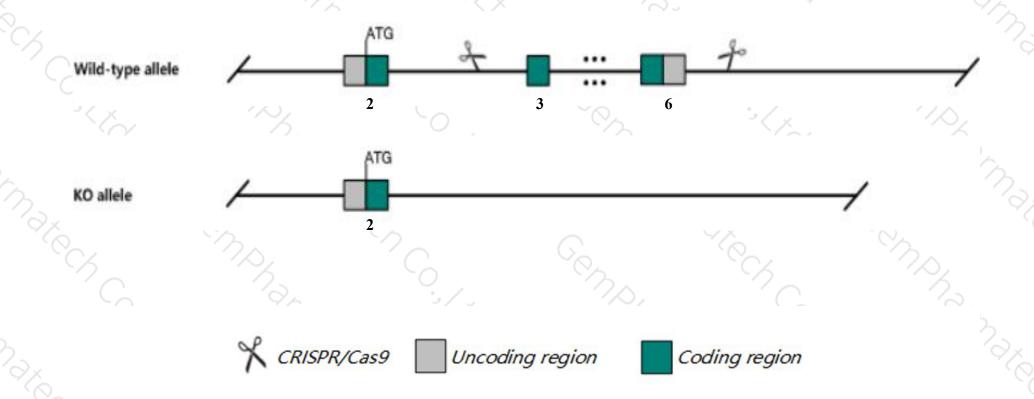
Strain background

C57BL/6JGpt

# **Knockout strategy**



This model will use CRISPR/Cas9 technology to edit the *Rspo1* gene. The schematic diagram is as follows:



### **Technical routes**



- ➤ The *Rspo1* gene has 2 transcripts. According to the structure of *Rspo1* gene, exon3-exon6 of *Rspo1-201* (ENSMUST00000030687.7) transcript is recommended as the knockout region. The region contains most coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Rspo1* gene. The brief process is as follows: gRNA was transcribed in vitro.Cas9 and gRNA 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.

### **Notice**



- ➤ According to the existing MGI data, homozygous inactivation of this gene leads to abnormal ovarian development and masculinized features including pseudohermaphroditism in genital ducts, depletion of fetal oocytes, male-like vascularization, and ectopic testosterone production in the ovaries.
- > The *Rspo1* gene is located on the Chr4. 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 gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

### Gene information (NCBI)



#### Rspo1 R-spondin 1 [Mus musculus (house mouse)]

Gene ID: 192199, updated on 13-Mar-2020

#### Summary

☆ ?

Official Symbol Rspo1 provided by MGI

Official Full Name R-spondin 1 provided by MGI

Primary source MGI:MGI:2183426

See related Ensembl: ENSMUSG00000028871

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 R-spondin, Rspondin

Expression Broad expression in ovary adult (RPKM 13.8), CNS E11.5 (RPKM 10.5) and 19 other tissuesSee more

Orthologs <u>human</u> all

# Transcript information (Ensembl)



The gene has 2 transcripts, all transcripts are shown below:

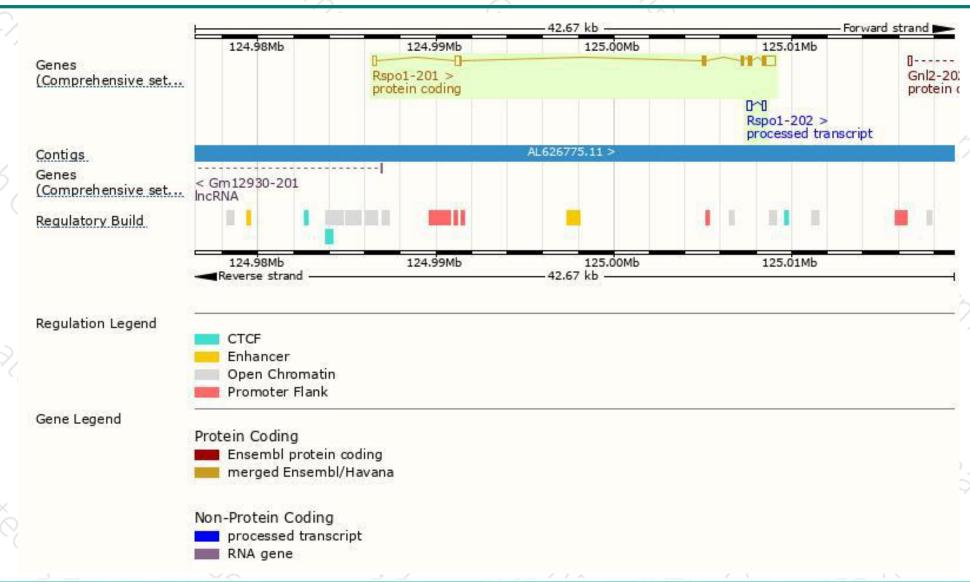
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Rspo1-201	ENSMUST00000030687.7	1834	265aa	Protein coding	CCDS18633	B1ASC1	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Rspo1-202	ENSMUST00000140698.1	446	No protein	Processed transcript		-	TSL:3

The strategy is based on the design of *Rspo1-201* transcript, the transcription is shown below

Rspo1-201 > protein coding

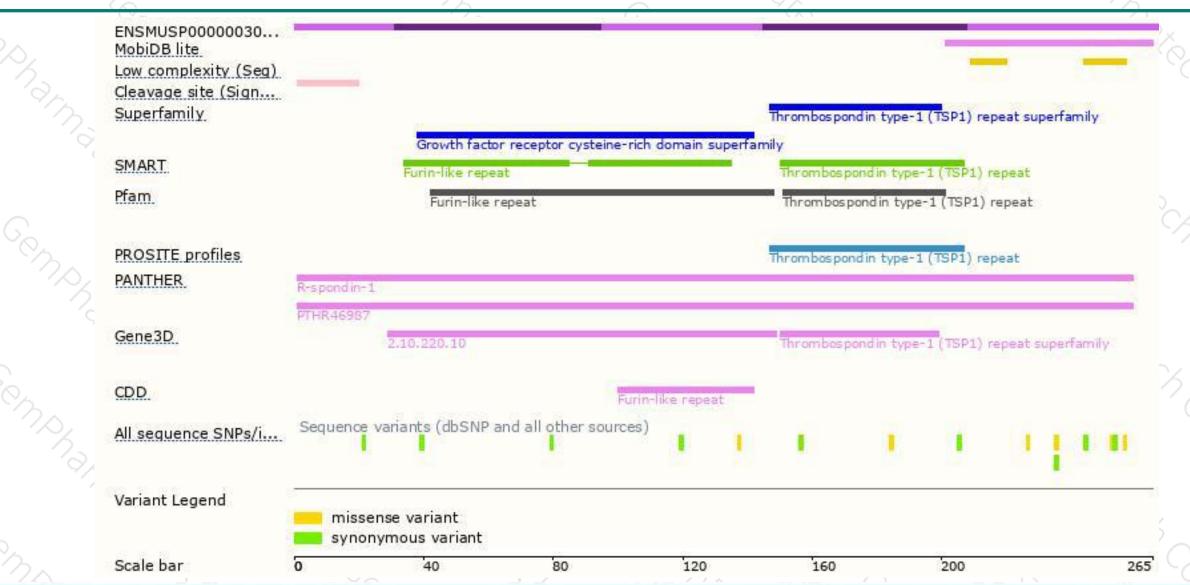
### Genomic location distribution





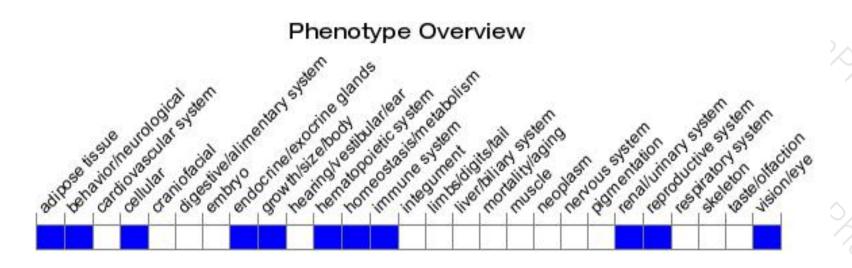
### Protein domain





## 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, homozygous inactivation of this gene leads to abnormal ovarian development and masculinized features including pseudohermaphroditism in genital ducts, depletion of fetal oocytes, male-like vascularization, and ectopic testosterone production in the ovaries.



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





