

Dazl Cas9-CKO Strategy

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Project Overview

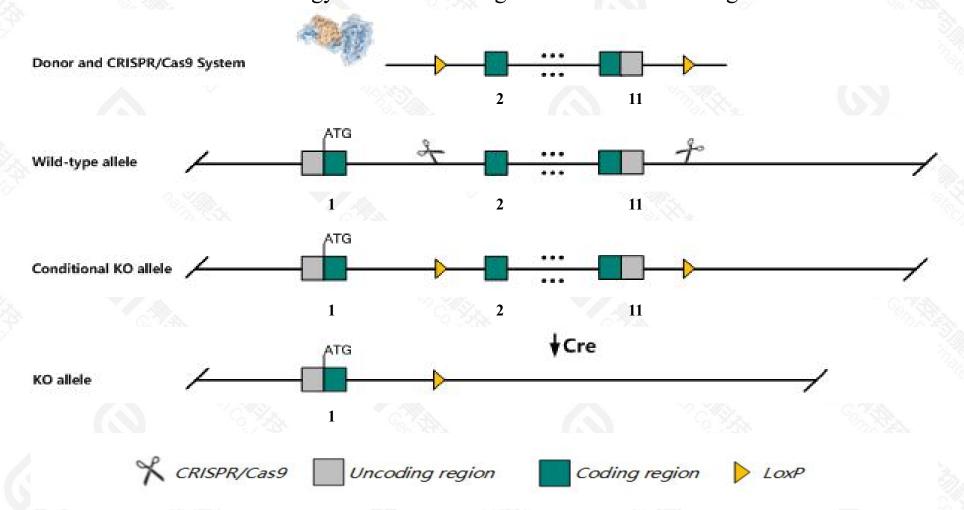


Project Name	Dazl		
Project type	Cas9-CKO		
Strain background	C57BL/6JGpt		

Conditional Knockout strategy



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



Technical routes



- > The *Dazl* gene has 6 transcripts. According to the structure of *Dazl* gene, exon2-exon11 of *Dazl-*201(ENSMUST00000010736.9) transcript is recommended as the knockout region. The region contains most of the coding sequence. Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Dazl* 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, males homozygous for a targeted null mutation exhibit sterility with a complete absence of mature germ cells. In mutants, few spermatogonia enter meiosis, and those that do fail to proceed beyond pachytene.
- The *Dazl* 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)



Dazl deleted in azoospermia-like [Mus musculus (house mouse)]

Gene ID: 13164, updated on 13-Mar-2020

Summary

☆ ?

Official Symbol Dazl provided by MGI

Official Full Name deleted in azoospermia-like provided by MGI

Primary source MGI:MGI:1342328

See related Ensembl: ENSMUSG00000010592

RefSeq status REVIEWED
Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;

Muroidea; Muridae; Murinae; Mus; Mus

Also known as Daz-like, Dazh, Dazl1, Dazla, Tpx-2, Tpx2

Summary This gene encodes a member of the depleted in azoospermia-like (DAZL) protein family. Members of this family contain an RNA recognition

motif, interact with poly A binding proteins, and may be involved in the initiation of translation. The encoded protein is expressed in the

cytoplasm of pluripotent stem cells, and in both male and female germ cells, where it is essential for gametogenesis. Disruption of this gene is

associated with infertility. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Apr 2013]

Expression Biased expression in testis adult (RPKM 15.0) and ovary adult (RPKM 0.8)See more

Orthologs human all

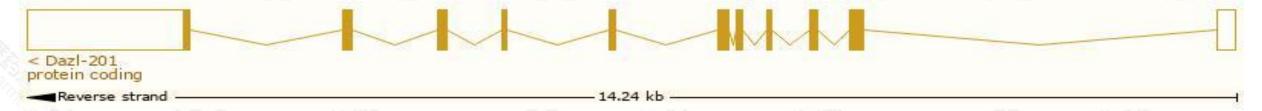
Transcript information (Ensembl)



The gene has 6 transcripts, all transcripts are shown below:

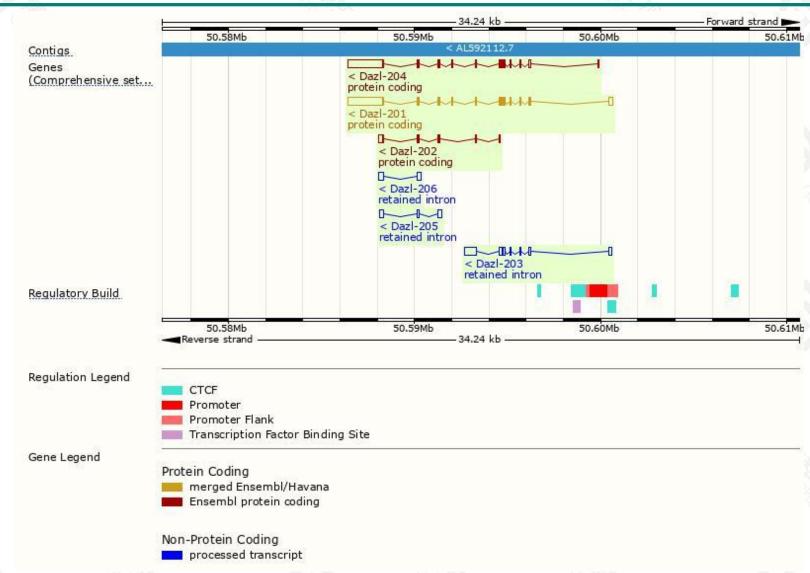
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Dazl-201	ENSMUST00000010736.8	2976	298aa	Protein coding	CCDS37649	Q64368	TSL:1 GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P
Dazl-204	ENSMUST00000233248.1	2820	<u>261aa</u>	Protein coding	-	Q3TUC3	GENCODE basic
Dazl-202	ENSMUST00000232796.1	600	<u>133aa</u>	Protein coding	12	A0A3B2WD26	CDS 5' incomplete
Dazl-203	ENSMUST00000232875.1	1342	No protein	Retained intron	2	2	
Dazl-205	ENSMUST00000233620.1	512	No protein	Retained intron	-	-	
Dazl-206	ENSMUST00000233756.1	457	No protein	Retained intron	-	-	

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



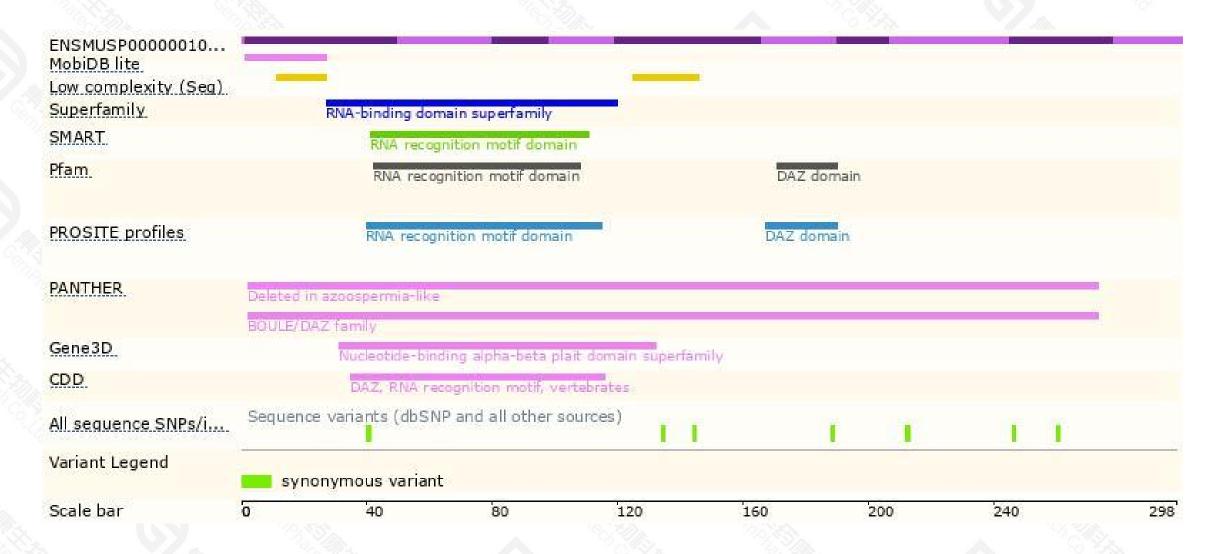
Genomic location distribution





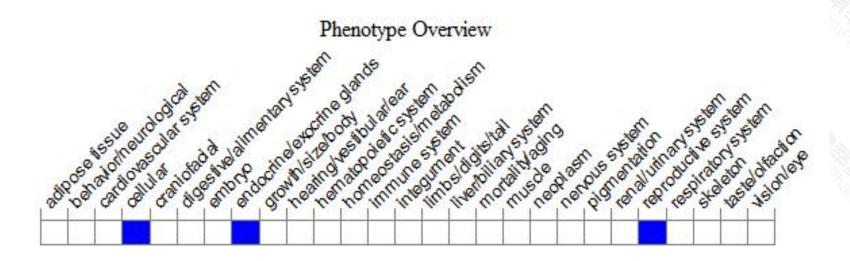
Protein domain





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,males homozygous for a targeted null mutation exhibit sterility with a complete absence of mature germ cells. In mutants, few spermatogonia enter meiosis, and those that do fail to proceed beyond pachytene.



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

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