

# ***Col25a1 Cas9-KO Strategy***

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**Reviewer :**

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**Design Date:**

**2019-9-28**

# Project Overview

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**Project Name**

*Col25a1*

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**Project type**

Cas9-KO

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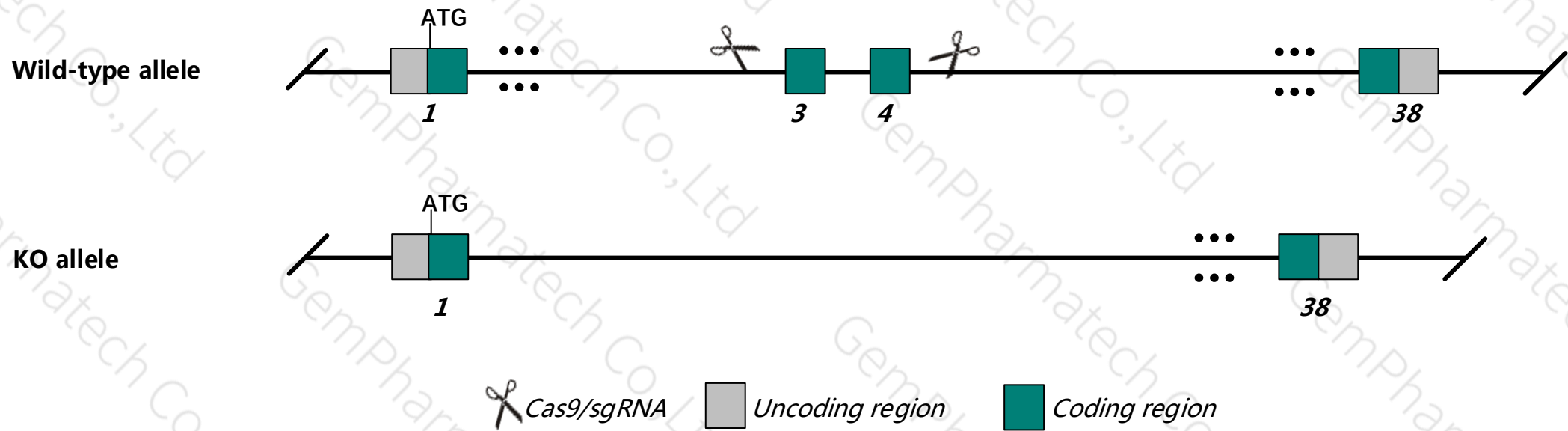
**Animal background**

C57BL/6JGpt

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# Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Col25a1* gene. The schematic diagram is as follows:



# Technical routes

- The *Col25a1* gene has 6 transcripts, According to the structure of *Col25a1* gene, exon3-exon4 of *Col25a1* -201 transcript is recommended as the knockout region. The region contains the 53bp coding sequence. Knock out the region, result in destruction of protein.
- This project uses CRISPR/Cas9 technology to modify *Col25a1* gene. The brief process is as follows: sgRNA was transcribed in vitro, Cas9, sgRNA were microinjected into fertilized eggs of C57BL/6JGpt mice and homologous recombination was carried out to obtain F0 mice. A stable and hereditary F1 generation mouse model was obtained by mating F0 generation mice with C57BL/6JGpt mice which were confirmed positive by PCR-sequencing.

- According to the existing MGI data , Mice homozygous for a knock-out allele exhibit neonatal lethality, cyanosis and abnormal body curvature with apoptosis of phrenic nerve motor neurons and failure of diaphragm innervation.
- The *Col25a1* gene is located in the Chr3. If the knockout mice are mixed with other mice, two target genes are avoided on the same chromosome as possible, otherwise the offspring of mice with double gene positive and homozygous gene knockout can not be obtained.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of gene transcription and translation processes, all risks cannot be predicted under existing information.

# Gene information ( NCBI )

**Col25a1 collagen, type XXV, alpha 1 [**

**(house mouse) ]**

Gene ID: 77018, updated on 8-Dec-2018

## Summary

**Official Symbol** Col25a1 provided by [MGI](#)

**Official Full Name** collagen, type XXV, alpha 1 provided by [MGI](#)

**Primary source** [MGI:MGI:1924268](#)

**See related** [Ensembl:ENSMUSG00000058897](#)

**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** CLAC-P; 2700062B08Rik

**Expression** Biased expression in bladder adult (RPKM 7.3), limb E14.5 (RPKM 4.6) and 10 other tissues [See more](#)

**Orthologs** [human](#) [all](#)



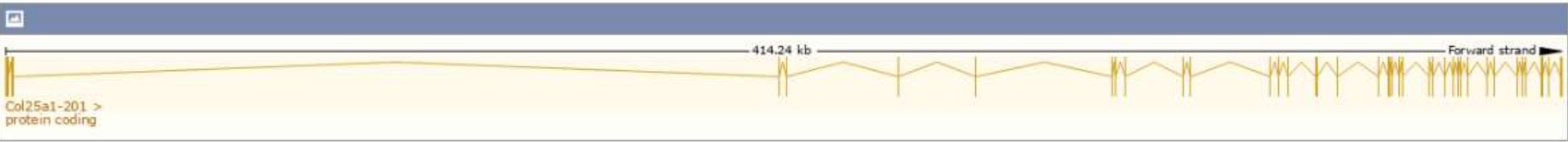
# Transcript information ( Ensembl )

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

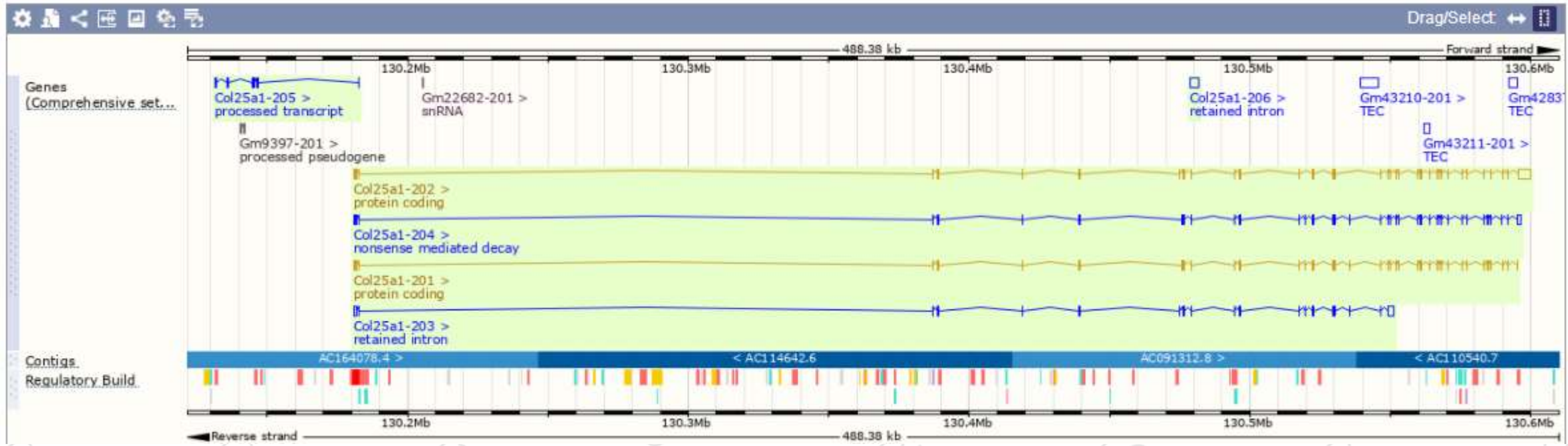
Show/hide columns (1 hidden) Filter

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags
Col25a1-202	<a href="#">ENSMUST00000106353.1</a>	6701	<a href="#">640aa</a>	Protein coding	<a href="#">CCDS38636</a>	<a href="#">E9Q5L6</a>	<a href="#">NM_198711</a> <a href="#">NP_942004</a>	TSL:5 GENCODE basic APPRIS ALT2
Col25a1-201	<a href="#">ENSMUST00000080335.10</a>	2624	<a href="#">666aa</a>	Protein coding	<a href="#">CCDS38637</a>	<a href="#">Q99MQ5</a>	<a href="#">NM_001244952</a> <a href="#">NM_029838</a> <a href="#">NP_001231881</a> <a href="#">NP_084114</a>	TSL:5 GENCODE basic APPRIS P4
Col25a1-204	<a href="#">ENSMUST00000183368.7</a>	3778	<a href="#">593aa</a>	Nonsense mediated decay	-	<a href="#">V9GWX5</a>	-	TSL:5
Col25a1-205	<a href="#">ENSMUST00000197950.1</a>	1078	No protein	Processed transcript	-	-	-	TSL:1
Col25a1-203	<a href="#">ENSMUST00000143830.2</a>	3514	No protein	Retained intron	-	-	-	TSL:5
Col25a1-206	<a href="#">ENSMUST00000198205.1</a>	2723	No protein	Retained intron	-	-	-	TSL:NA

The strategy is based on the design of *Col25a1* -201 transcript,The transcription is shown below :

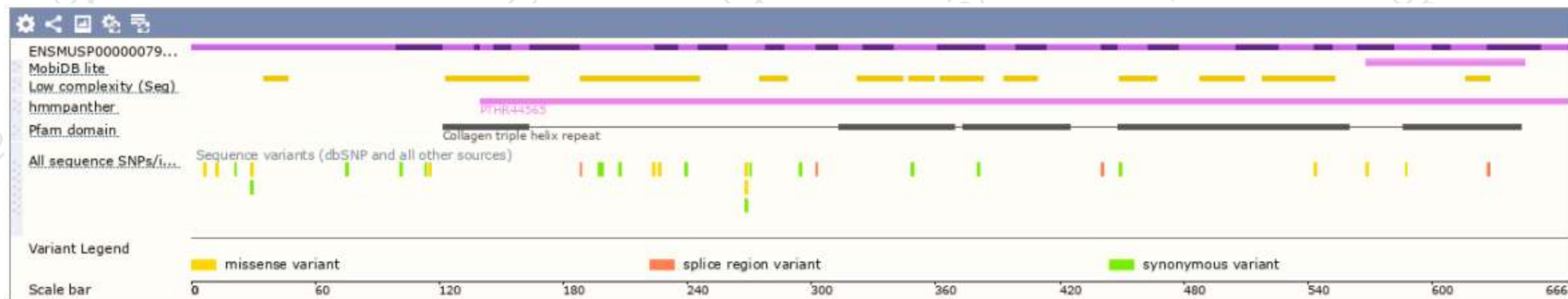


# Genomic location distribution

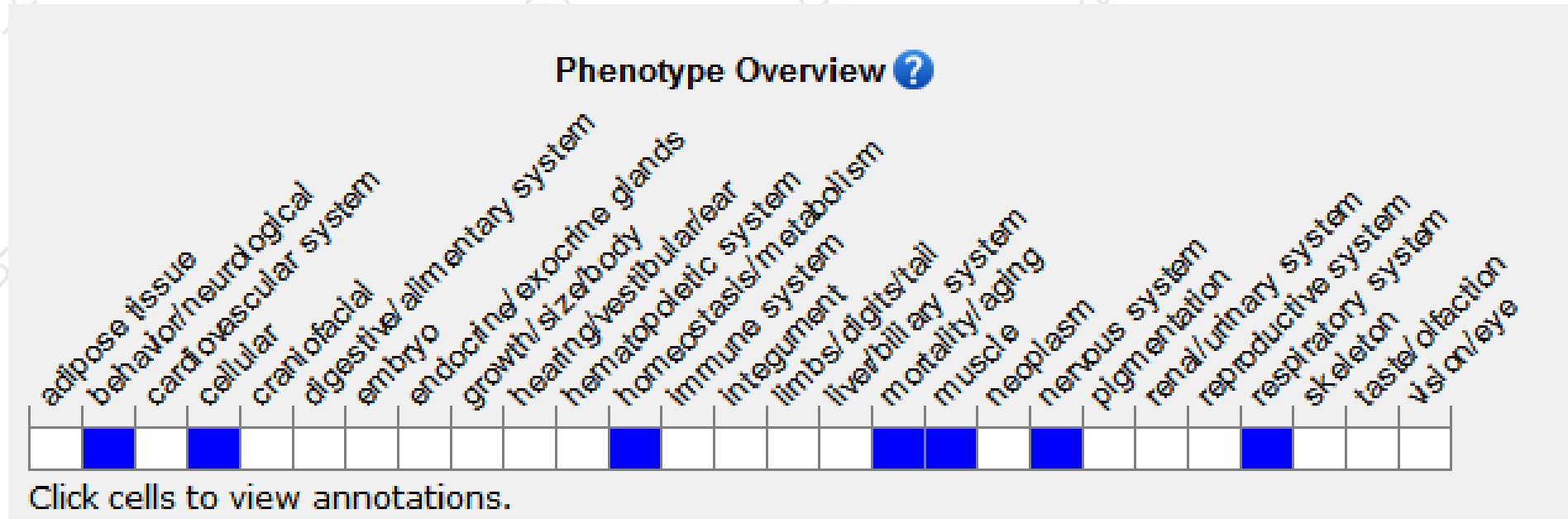




# Protein domain



# 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, Mice homozygous for a knock-out allele exhibit neonatal lethality, cyanosis and abnormal body curvature with apoptosis of phrenic nerve motor neurons and failure of diaphragm innervation.

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