

H19 Cas9-CKO Strategy

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Project Overview



Project Name

H19

Project type

Cas9-CKO

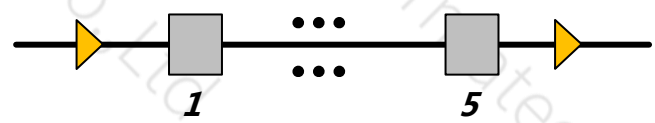
Strain background

C57BL/6JGpt

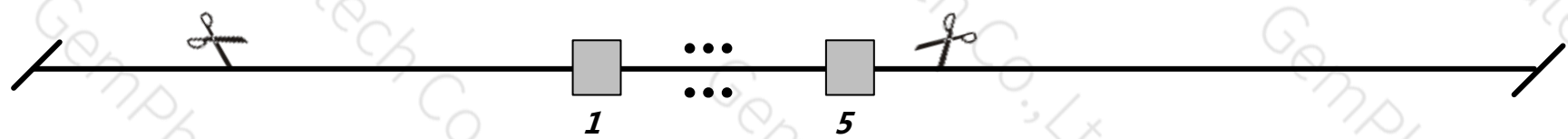
Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *H19* gene. The schematic diagram is as follows:

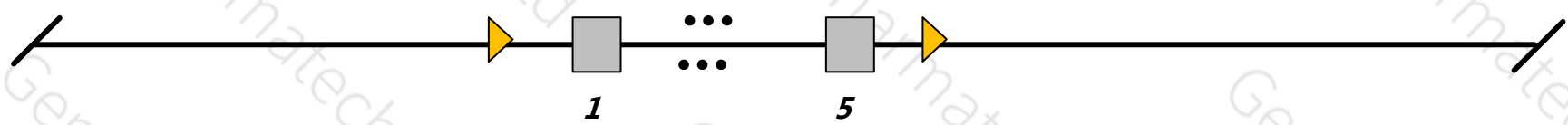
Donor and CRISPR/Cas9 System



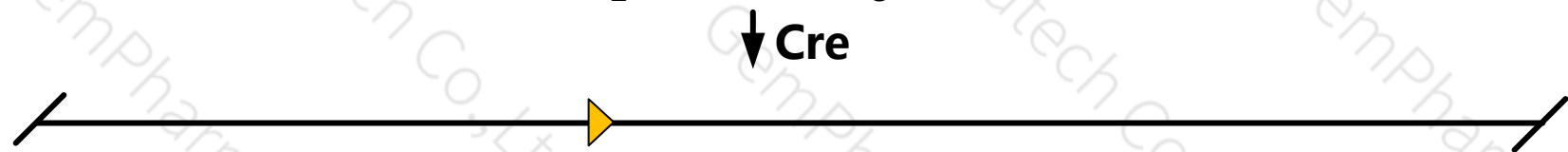
Wild-type allele



Conditional KO allele



KO allele



↓ Cre



- The *H19* gene has 7 transcripts. According to the structure of *H19* gene, exon1-exon5 of *H19*-202 (ENSMUST00000136359.7) transcript is recommended as the knockout region. The region contains all of the sequence. Knock out the region will result in disruption of gene function.
- In this project we use CRISPR/Cas9 technology to modify *H19* gene. The brief process is as follows: sgRNA was transcribed in vitro, donor vector was constructed. Cas9, sgRNA 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 was 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.

- According to the existing MGI data , Mice homozygous for a knock-out allele exhibit increased body weight. Mice heterozygous for a maternally inherited knock-out allele exhibit increased placenta and body weight. Mice heterozygous for a different knock-out allele paternally inherited exhibit decreased fetal and postnatal body weight.
- The target gene overlaps with *Mir675*. After mating with Cre mice, knocking out the target gene will also knock out *Mir675*.
- The target gene overlaps with the predicted gene miscRNA *Gm27483* and miscRNA *Gm27786*. After mating with Cre mice, knocking out the target gene will also knock out the predictive gene, and the impact is unknown.
- The *H19* gene is located on the Chr7. 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)

H19 H19, imprinted maternally expressed transcript [*Mus musculus* (house mouse)]

Gene ID: 14955, updated on 28-Oct-2018

Summary

Official Symbol H19 provided by [MGI](#)

Official Full Name H19, imprinted maternally expressed transcript provided by [MGI](#)

Primary source [MGI:MGI:95891](#)

See related [Ensembl:ENSMUSG00000000031](#)

Gene type ncRNA

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 AI747191; EyeLinc6

Expression Biased expression in liver E18 (RPKM 5536.3), placenta adult (RPKM 4196.2) and 5 other tissues [See more](#)

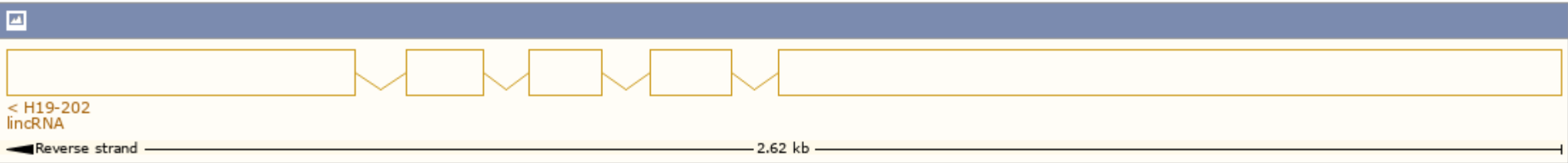
Orthologs [human](#) [all](#)

Transcript information (Ensembl)

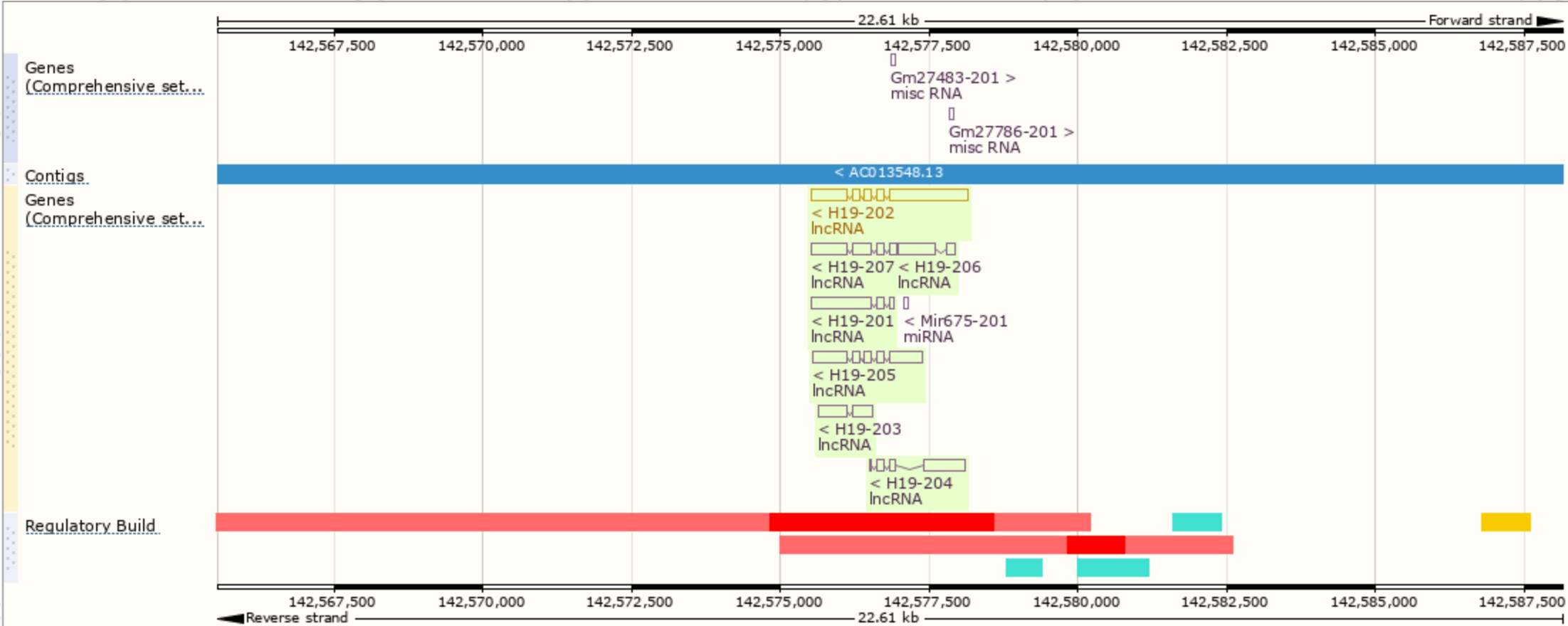
The gene has 7 transcripts, and all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	Flags
H19-202	ENSMUST00000136359.7	2286	No protein	lncRNA	-	TSL:1 Gencode basic
H19-205	ENSMUST00000152754.8	1521	No protein	lncRNA	-	TSL:5
H19-201	ENSMUST00000132294.8	1217	No protein	lncRNA	-	TSL:2
H19-207	ENSMUST00000228514.1	1178	No protein	lncRNA	-	-
H19-204	ENSMUST00000149974.1	935	No protein	lncRNA	-	TSL:3
H19-203	ENSMUST00000140716.1	817	No protein	lncRNA	-	TSL:2
H19-206	ENSMUST00000228259.1	761	No protein	lncRNA	-	-

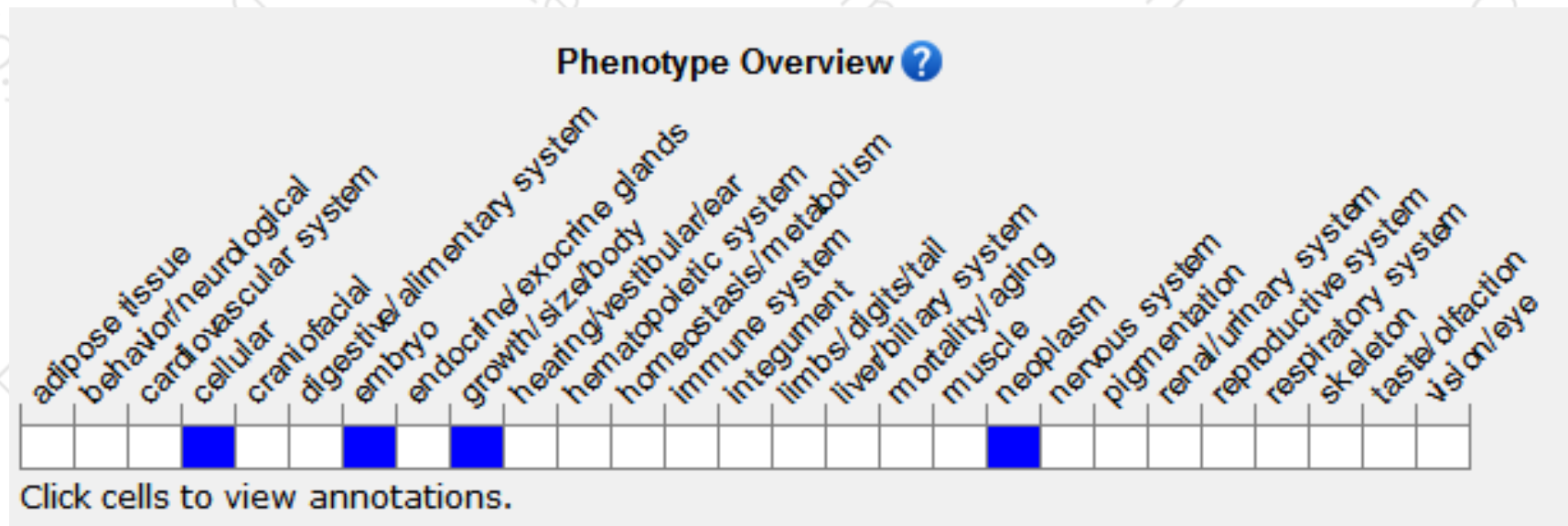
The strategy is based on the design of *H19-202* transcript, The transcription is shown below



Genomic location distribution



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 increased body weight. Mice heterozygous for a maternally inherited knock-out allele exhibit increased placenta and body weight. Mice heterozygous for a different knock-out allele paternally inherited exhibit decreased fetal and postnatal body weight.

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