

# ***Inhba*** Cas9-CKO Strategy

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**Reviewer:**

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# Project Overview

**Project Name**

*Inhba*

**Project type**

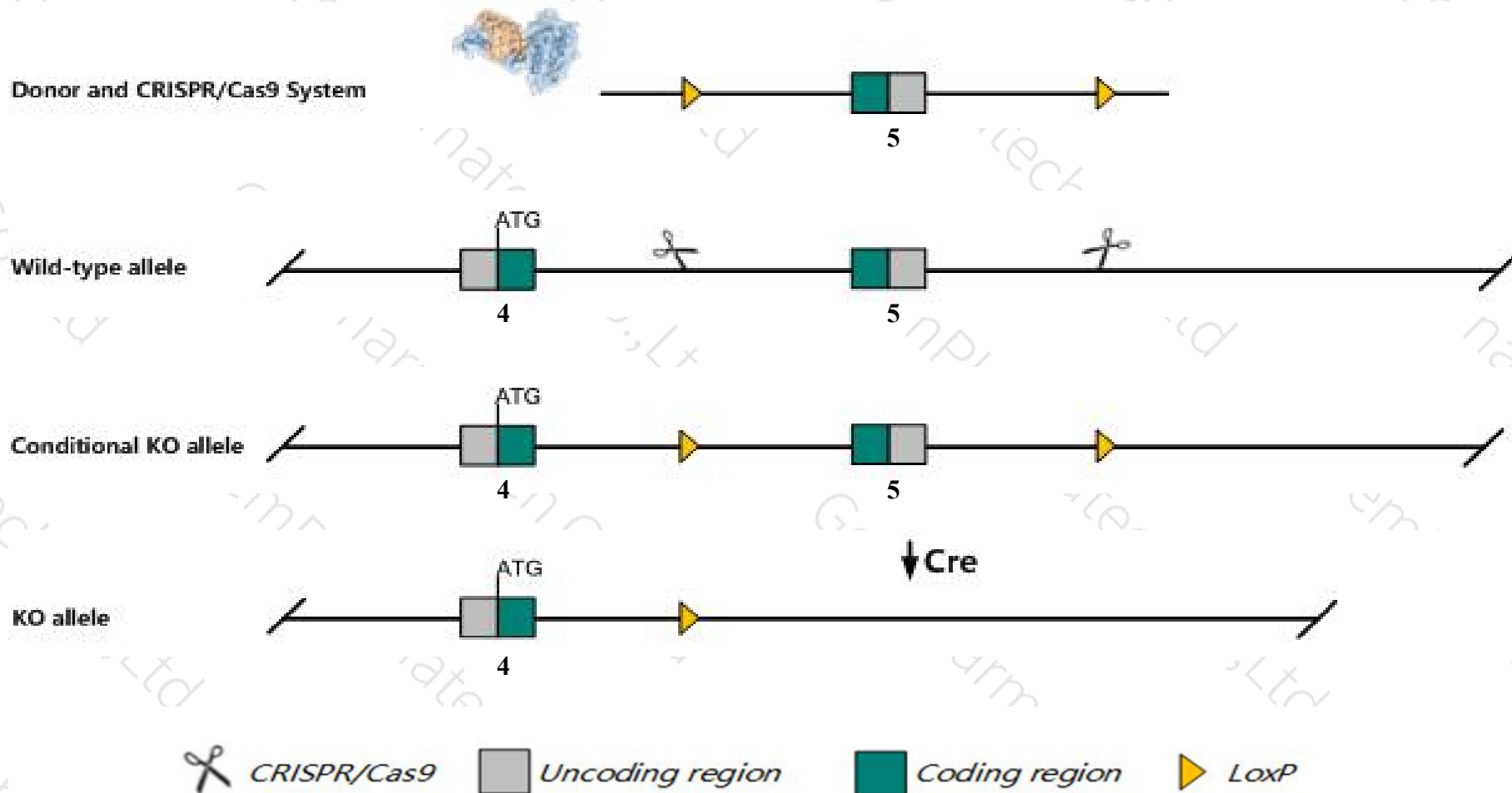
**Cas9-CKO**

**Strain background**

**C57BL/6JGpt**

# Conditional Knockout strategy

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



- The *Inhba* gene has 2 transcripts. According to the structure of *Inhba* gene, exon5 of *Inhba-201* (ENSMUST00000042603.13) transcript is recommended as the knockout region. The region contains 887bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Inhba* 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.

- According to the existing MGI data, Homozygotes for a targeted null mutation lack vibrissae and lower incisors, have defects in their secondary palates, and die shortly after birth.
- The KO region contains functional region of the *B230303A05Rik* and *Gm48444* gene. Knockout the region may affect the function of *B230303A05Rik* and *Gm48444* gene.
- The *Inhba* gene is located on the Chr13. 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)

## Inhba inhibin beta-A [Mus musculus (house mouse)]

Gene ID: 16323, updated on 31-Jan-2019

### Summary



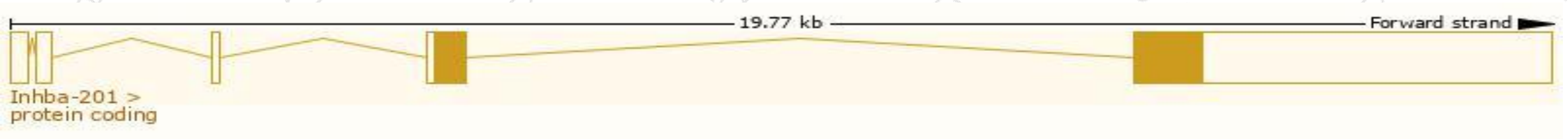
|                           |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                    |
|---------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Official Symbol</b>    | Inhba provided by <a href="#">MGI</a>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              |
| <b>Official Full Name</b> | inhibin beta-A provided by <a href="#">MGI</a>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |
| <b>Primary source</b>     | <a href="#">MGI:MGI:96570</a>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                      |
| <b>See related</b>        | <a href="#">Ensembl:ENSMUSG00000041324</a>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |
| <b>Gene type</b>          | protein coding                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |
| <b>RefSeq status</b>      | REVIEWED                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                           |
| <b>Organism</b>           | <a href="#">Mus musculus</a>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                       |
| <b>Lineage</b>            | Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus                                                                                                                                                                                                                                                                                                                                                                                                                          |
| <b>Summary</b>            | This gene encodes a member of the TGF-beta (transforming growth factor-beta) superfamily of proteins. The encoded preproprotein is proteolytically processed to generate a subunit of the dimeric activin and inhibin protein complexes. These complexes activate and inhibit, respectively, follicle stimulating hormone secretion from the pituitary gland. The encoded protein also plays a role in eye, tooth and testis development. Homozygous knockout mice for this gene lack whiskers and exhibit tooth and palate defects, leading to neonatal lethality. [provided by RefSeq, Aug 2016] |
| <b>Expression</b>         | Biased expression in ovary adult (RPKM 8.8), liver E18 (RPKM 1.2) and 5 other tissues <a href="#">See more</a>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |
| <b>Orthologs</b>          | <a href="#">human</a> <a href="#">all</a>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                          |

# Transcript information (Ensembl)

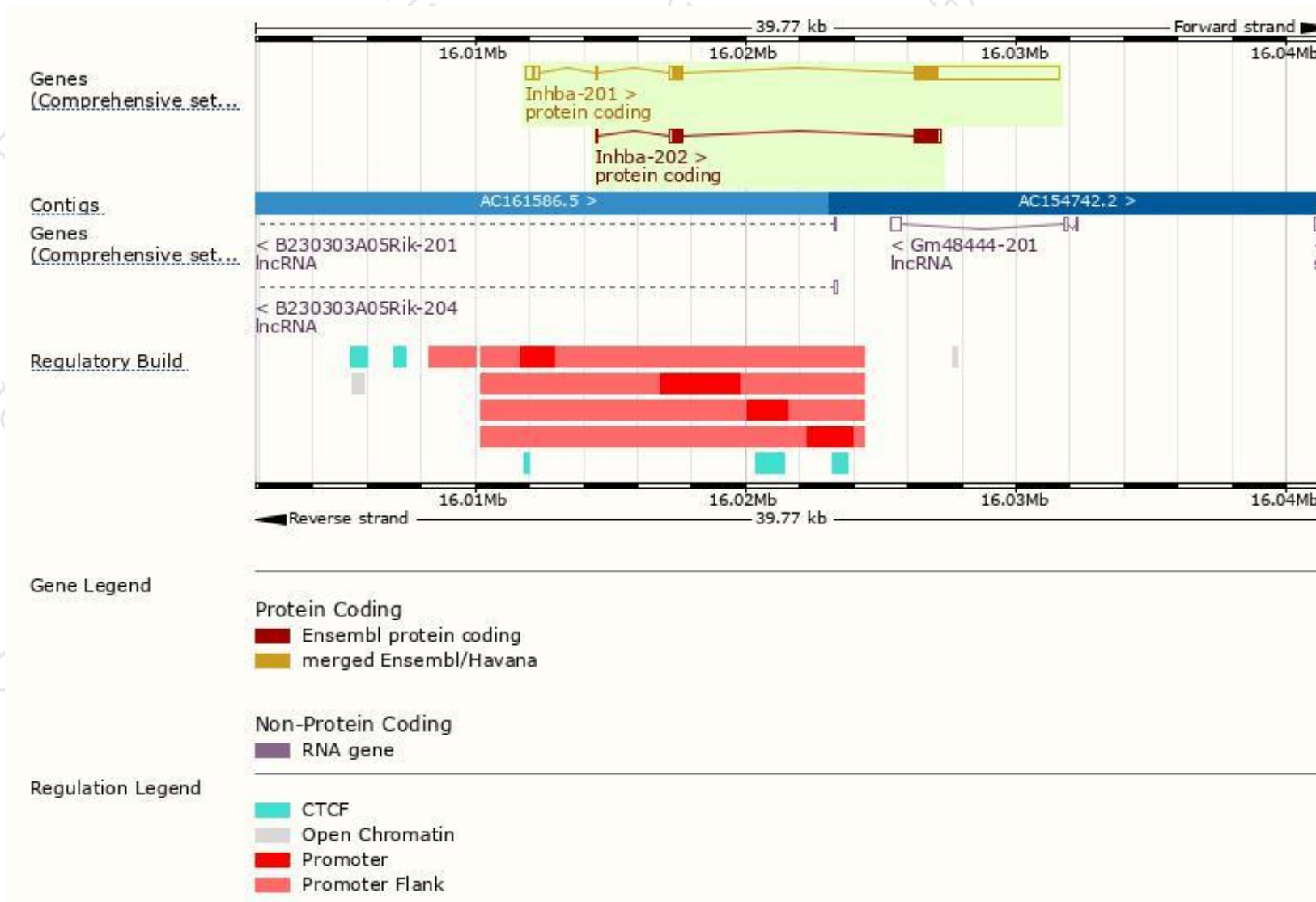
The gene has 2 transcripts,all transcripts are shown below:

| Name      | Transcript ID                         | bp   | Protein               | Biotype        | CCDS                      | UniProt                                       | Flags                         |
|-----------|---------------------------------------|------|-----------------------|----------------|---------------------------|-----------------------------------------------|-------------------------------|
| Inhba-201 | <a href="#">ENSMUST00000042603.13</a> | 6390 | <a href="#">424aa</a> | Protein coding | <a href="#">CCDS26251</a> | <a href="#">Q04998</a> <a href="#">Q3UY39</a> | TSL:1 GENCODE basic APPRIS P1 |
| Inhba-202 | <a href="#">ENSMUST00000164993.1</a>  | 1529 | <a href="#">424aa</a> | Protein coding | <a href="#">CCDS26251</a> | <a href="#">Q04998</a> <a href="#">Q3UY39</a> | TSL:1 GENCODE basic APPRIS P1 |

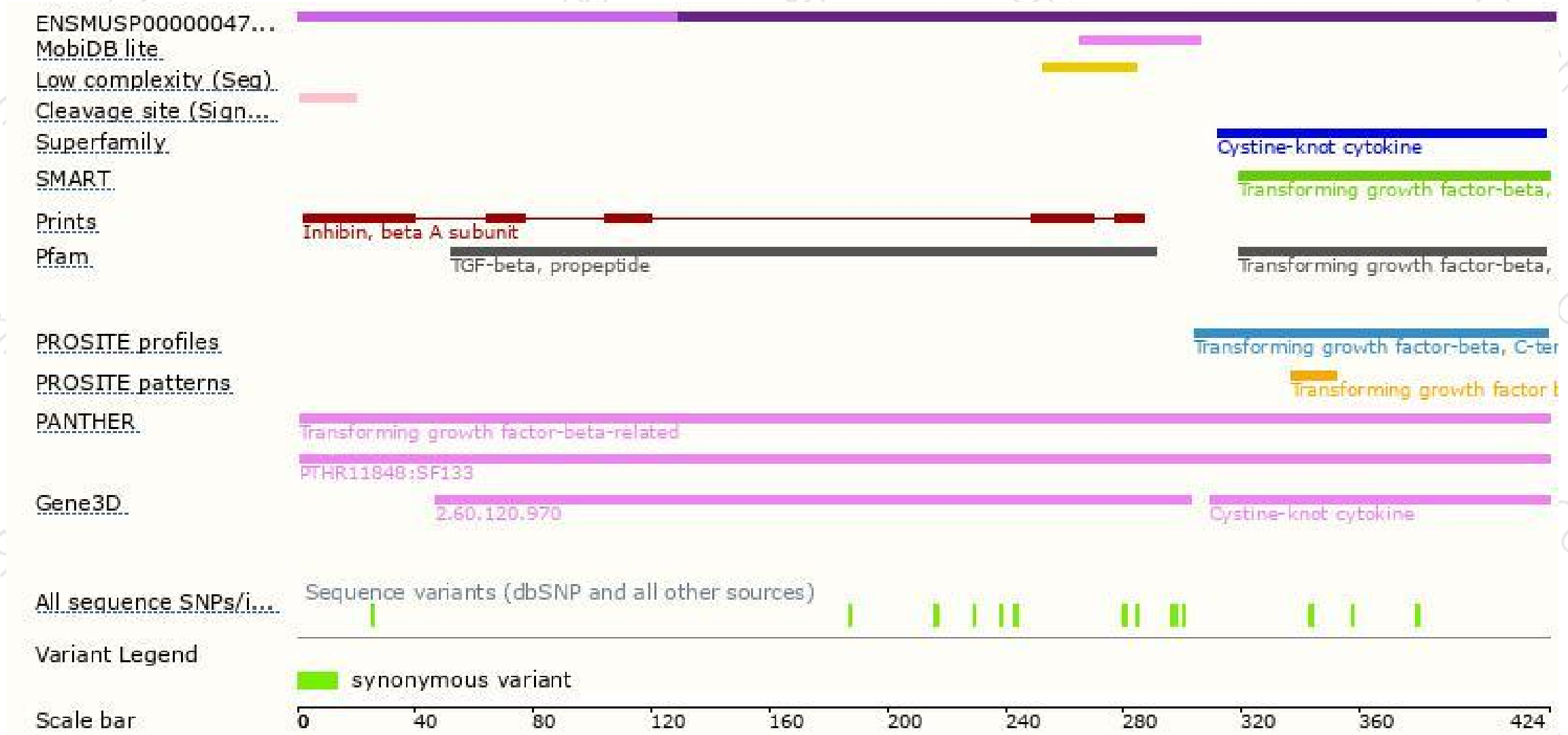
The strategy is based on the design of *Inhba-201* transcript,The transcription is shown below



# 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, Homozygotes for a targeted null mutation lack vibrissae and lower incisors, have defects in their secondary palates, and die shortly after birth.

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

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