

Txnrd3 Cas9-CKO Strategy

Designer: Daohua Xu

Reviewer: Huimin Su

Design Date: 2020-4-8

Project Overview

Project Name

Txnrd3

Project type

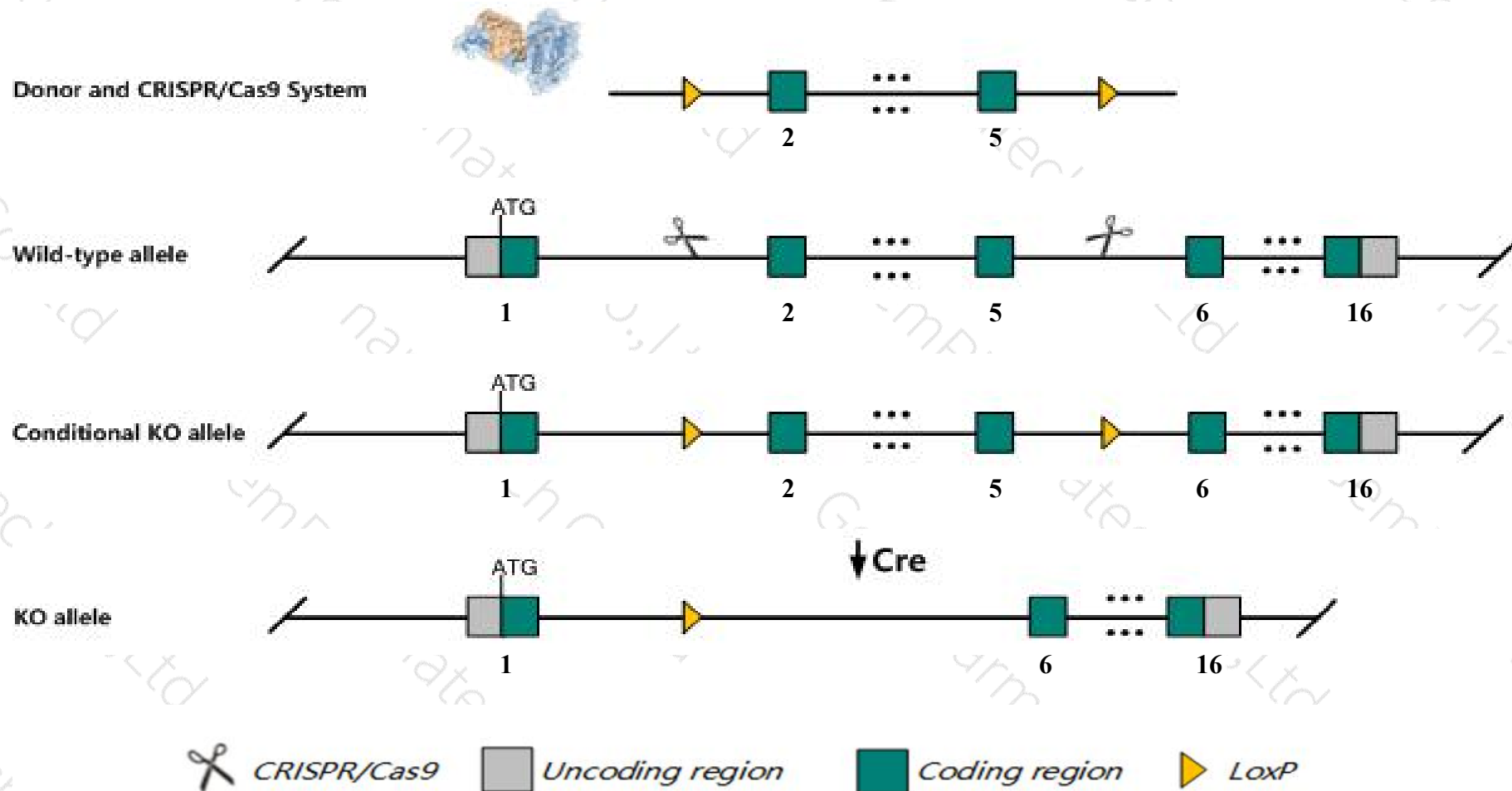
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Txnrd3* gene. The schematic diagram is as follows:



- The *Txnrd3* gene has 2 transcripts. According to the structure of *Txnrd3* gene, exon2-exon5 of *Txnrd3-201* (ENSMUST00000000828.12) transcript is recommended as the knockout region. The region contains 349bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Txnrd3* 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

- The *Txnrd3* gene is located on the Chr6. 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)

Txnrd3 thioredoxin reductase 3 [Mus musculus (house mouse)]

Gene ID: 232223, updated on 13-Mar-2020

Summary

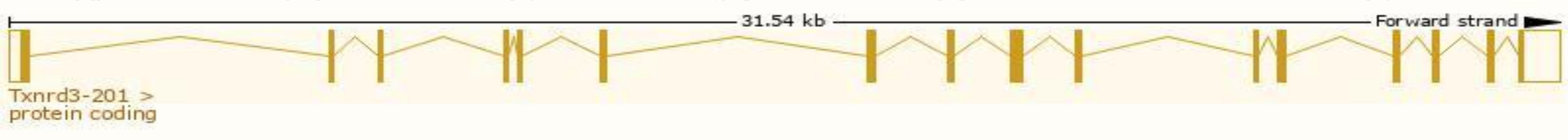
Official Symbol	Txnrd3 provided by MGI
Official Full Name	thioredoxin reductase 3 provided by MGI
Primary source	MGI:MGI:2386711
See related	Ensembl:ENSMUSG000000000811
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AI196535, TGR, TR2
Summary	The protein encoded by this gene belongs to the pyridine nucleotide-disulfide oxidoreductase family, and is a member of the thioredoxin (Trx) system. Three thioredoxin reductase (TrxR) isozymes are found in mammals. TrxRs are selenocysteine-containing flavoenzymes, which reduce thioredoxins, as well as other substrates, and play a key role in redox homeostasis. This gene encodes the third TrxR, which unlike the other two isozymes, contains an additional N-terminal glutaredoxin (Grx) domain, and shows highest expression in testis. The Grx domain allows this isozyme to participate in both Trx and glutathione systems. It functions as a homodimer containing FAD, and selenocysteine (Sec) at the active site. Sec is encoded by UGA codon that normally signals translation termination. The 3' UTRs of selenoprotein mRNAs contain a conserved stem-loop structure, the Sec insertion sequence (SECIS) element, which is necessary for the recognition of UGA as a Sec codon rather than as a stop signal. Alternatively spliced transcript variants encoding different isoforms have been found for this gene. There is evidence for additional isoforms resulting from the use of a non-AUG (CUG), and an in-frame downstream AUG as translation initiation codons (PMID:20018845). [provided by RefSeq, Aug 2017]
Expression	Biased expression in testis adult (RPKM 82.0), kidney adult (RPKM 6.4) and 4 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

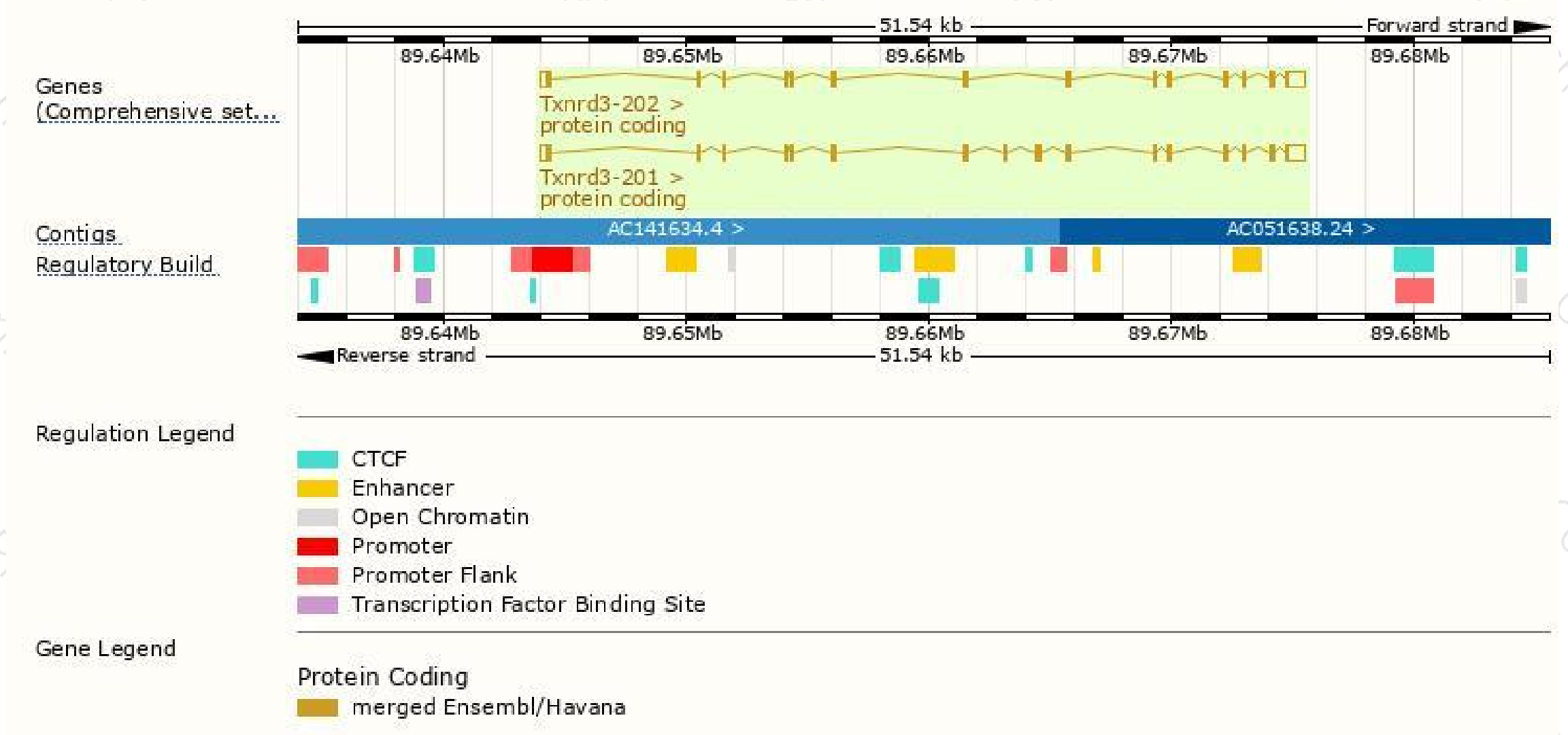
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Txnrd3-201	ENSMUST00000000828.12	2836	615aa	Protein coding	CCDS57436	G3X8P6	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
Txnrd3-202	ENSMUST00000101171.1	2494	501aa	Protein coding	CCDS57437	D3Z0H7	TSL:1 GENCODE basic

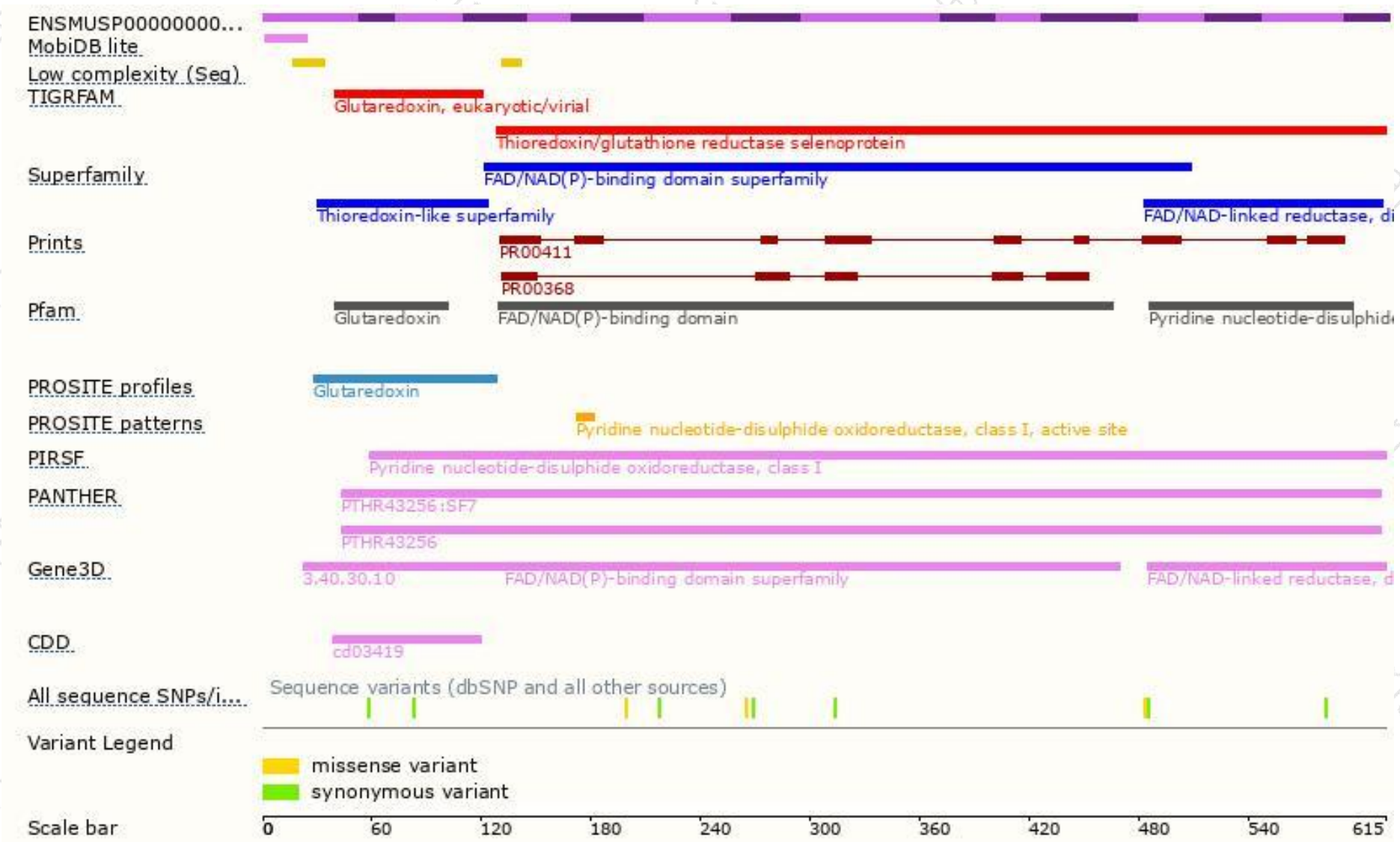
The strategy is based on the design of *Txnrd3-201* transcript,the transcription is shown below:



Genomic location distribution



Protein domain



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

Tel: 400-9660890

