

Tmed2 Cas9-CKO Strategy

Designer: Xueting Zhang

Reviewer: Daohua Xu

Design Date: 2020-6-12

Project Overview

Project Name

Tmed2

Project type

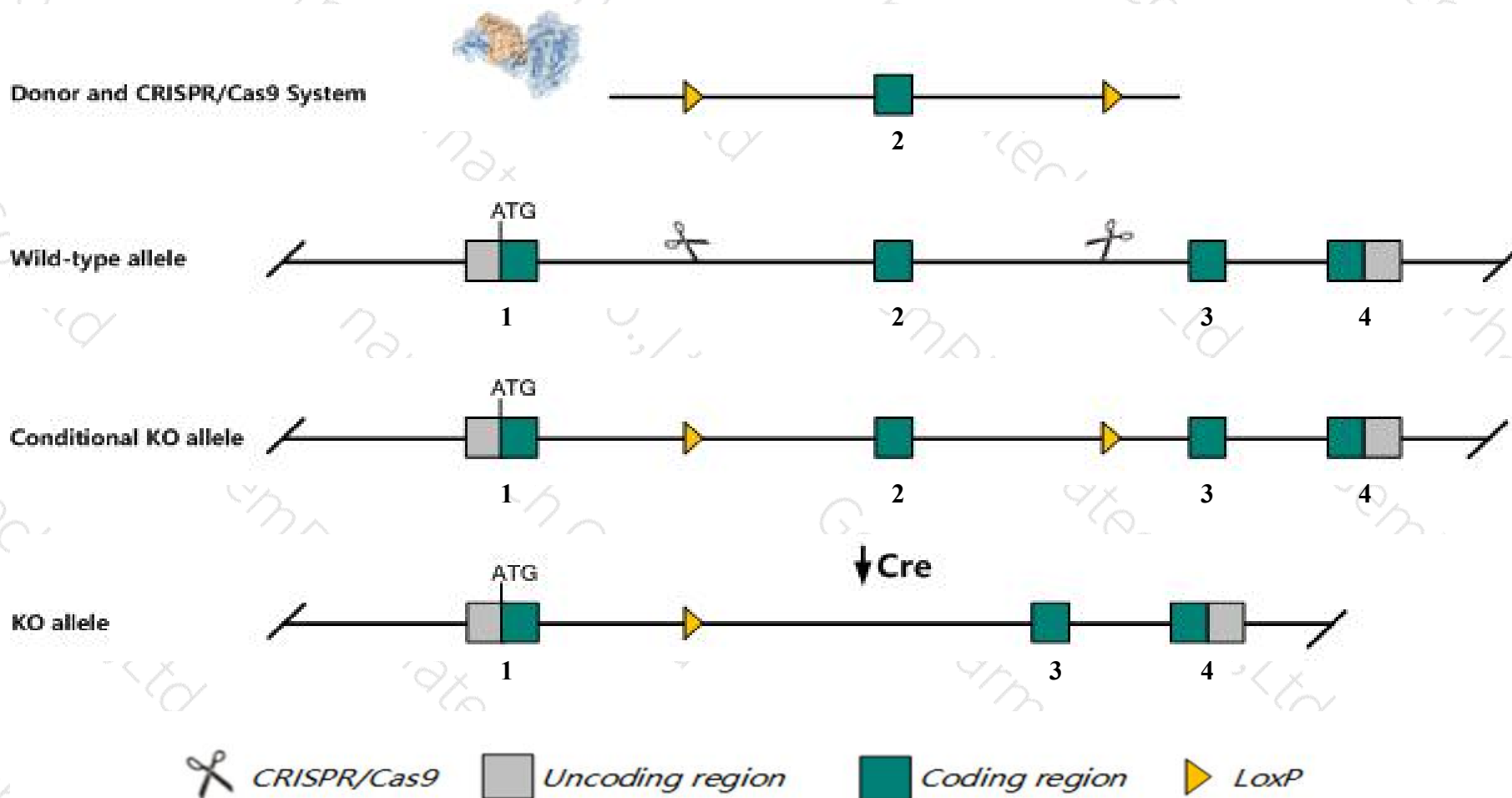
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Tmed2* gene. The schematic diagram is as follows:



- The *Tmed2* gene has 5 transcripts. According to the structure of *Tmed2* gene, exon2 of *Tmed2-201* (ENSMUST00000060226.10) transcript is recommended as the knockout region. The region contains 193bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Tmed2* 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, mice homozygous for an enu-induced allele exhibit embryonic lethality, embryonic growth retardation, caudal body truncation, and abnormal heart development.
- The effect on transcript *Tmed2-203* is unknown.
- The transcripts of *Tmed2-204* may not be affected.
- The *Tmed2* gene is located on the Chr5. 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)

Tmed2 transmembrane p24 trafficking protein 2 [Mus musculus (house mouse)]

Gene ID: 56334, updated on 13-Mar-2020

Summary



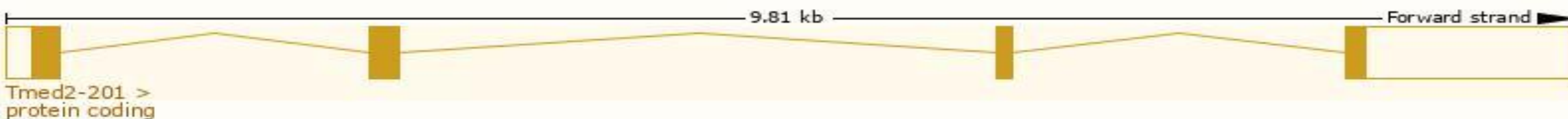
Official Symbol	Tmed2 provided by MGI
Official Full Name	transmembrane p24 trafficking protein 2 provided by MGI
Primary source	MGI:MGI:1929269
See related	Ensembl:ENSMUSG00000029390
Gene type	protein coding
RefSeq status	PROVISIONAL
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1110032D12Rik, 1810020N21Rik, Rnp24, Sid394, p24beta1
Expression	Ubiquitous expression in placenta adult (RPKM 136.3), ovary adult (RPKM 134.4) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

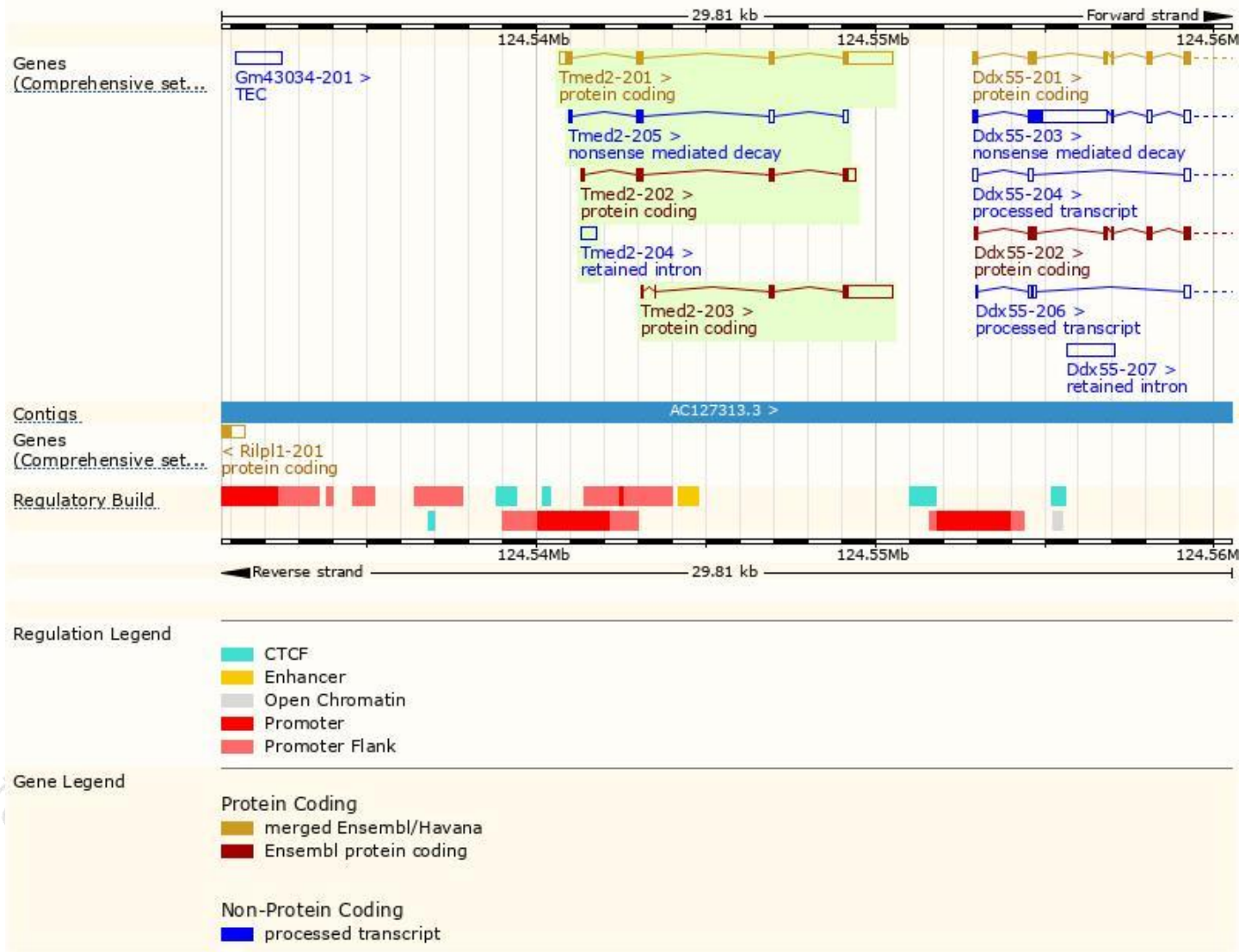
The gene has 5 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Tmed2-201	ENSMUST00000060226.10	2074	201aa	Protein coding	CCDS19679	A2RS53 Q9R0Q3	TSL:1 GENCODE basic APPRIS P1
Tmed2-203	ENSMUST00000135464.1	1630	112aa	Protein coding	-	Q8BPI2	CDS 5' incomplete TSL:1
Tmed2-202	ENSMUST00000124529.7	735	176aa	Protein coding	-	F6V6T4	CDS 5' incomplete TSL:2
Tmed2-205	ENSMUST00000200031.1	455	55aa	Nonsense mediated decay	-	A0A0G2JEB1	CDS 5' incomplete TSL:5
Tmed2-204	ENSMUST00000197112.1	425	No protein	Retained intron	-	-	TSL:NA

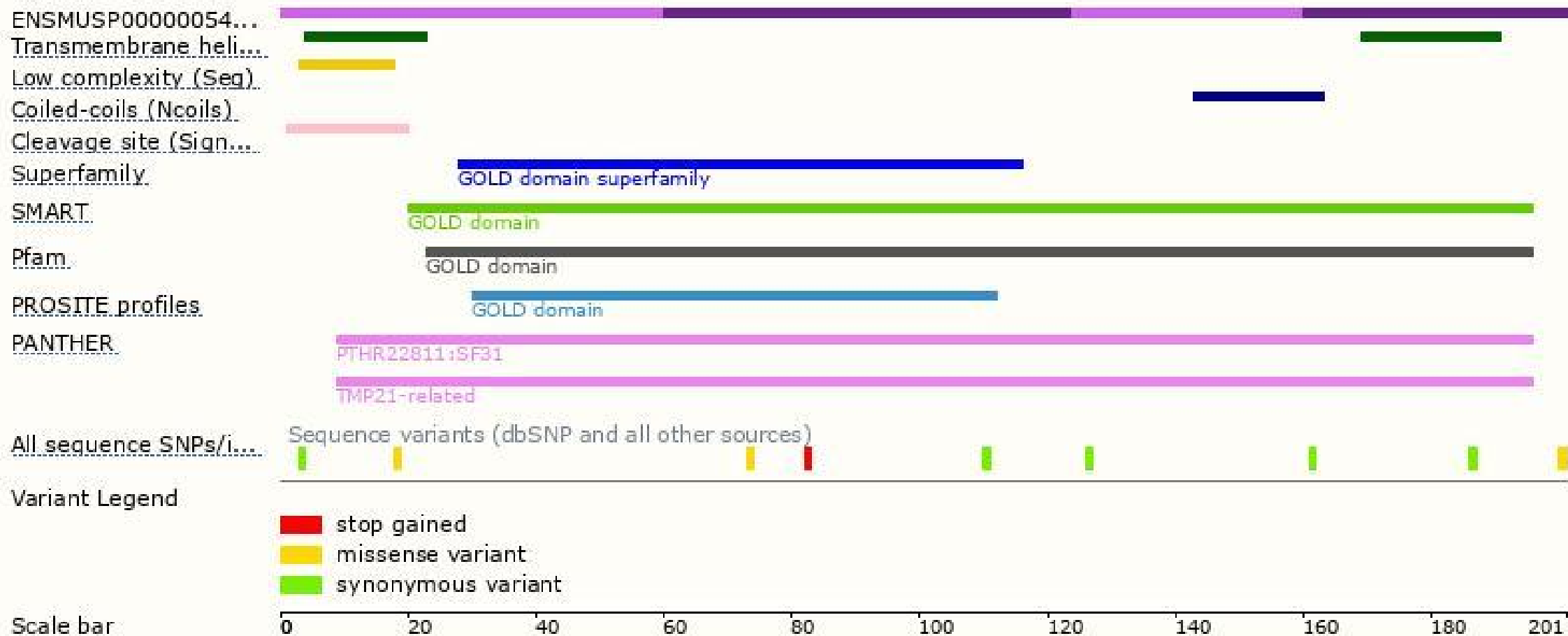
The strategy is based on the design of *Tmed2-201* transcript, the transcription is shown below:



Genomic location distribution

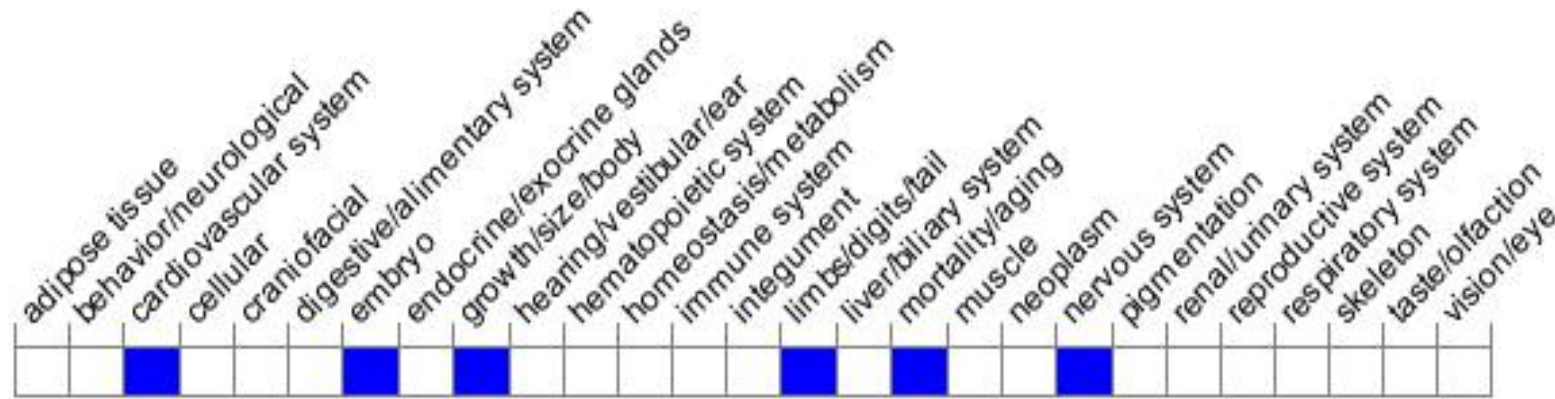


Protein domain



Mouse phenotype description(MGI)

Phenotype Overview



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 an ENU-induced allele exhibit embryonic lethality, embryonic growth retardation, caudal body truncation, and abnormal heart development.

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

Tel: 400-9660890

