Selenok Cas9-CKO Strategy

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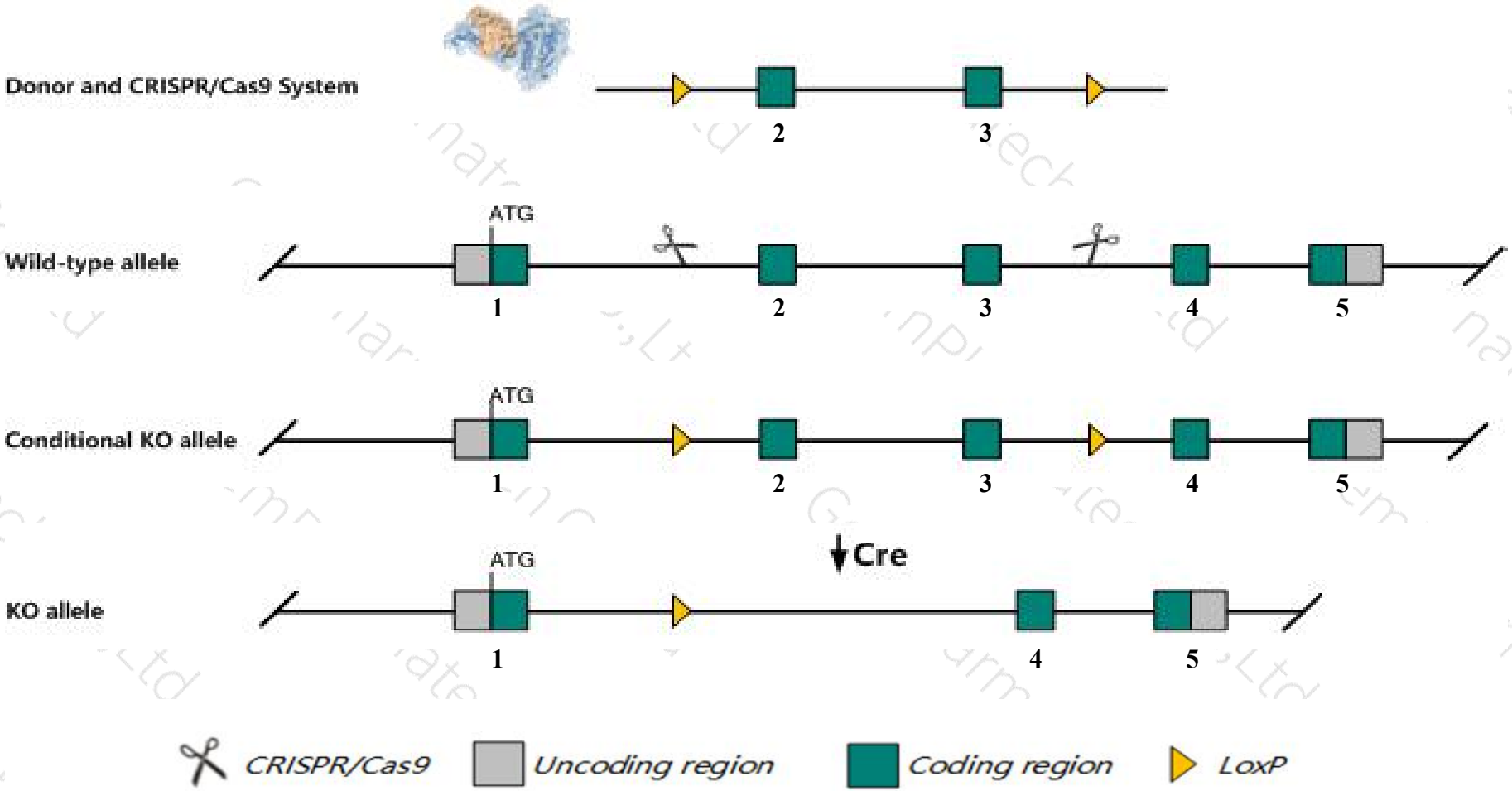
Project Overview

Project Name	***Selenok***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

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



Technical routes

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- The *Selenok* gene has 3 transcripts. According to the structure of *Selenok* gene, exon2-exon3 of *Selenok-201* (ENSMUST00000112268.2) transcript is recommended as the knockout region.The region contains 175bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Selenok* 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

- According to the existing MGI data,Mice homozygous for a knock-out allele exhibit impaired T cell, neutrophil, and macrophage calcium flux and increased susceptibility to viral infection.

- The *Selenok* gene is located on the Chr14. 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）

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Selenok selenoprotein K [Mus musculus (house mouse)]

Gene ID: 80795, updated on 31-Jan-2019

Summary

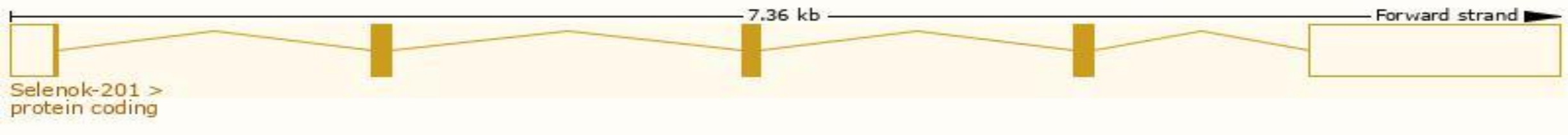
Official Symbol	Selenok provided by MGI
Official Full Name	selenoprotein K provided by MGI
Primary source	MGI:MGI:1931466
See related	Ensembl:ENSMUSG00000042682
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1110001C03Rik, AA673253, HSPC030, Hsp30, Selk
Summary	The protein encoded by this gene belongs to the selenoprotein K family. It is a transmembrane protein that is localized in the endoplasmic reticulum (ER), and is involved in ER-associated degradation (ERAD) of misfolded, glycosylated proteins. It also has a role in the protection of cells from ER stress-induced apoptosis. Knockout studies in mice show the importance of this gene in promoting Ca(2+) flux in immune cells and mounting effective immune response. This protein is a selenoprotein, containing the rare amino acid selenocysteine (Sec). Sec is encoded by the UGA codon, which normally signals translation termination. The 3' UTRs of selenoprotein mRNAs contain a conserved stem-loop structure, designated the Sec insertion sequence (SECIS) element, that is necessary for the recognition of UGA as a Sec codon, rather than as a stop signal. Multiple pseudogenes of this locus have been identified. [provided by RefSeq, Sep 2017]
Expression	Broad expression in liver E18 (RPKM 94.4), CNS E18 (RPKM 45.8) and 23 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

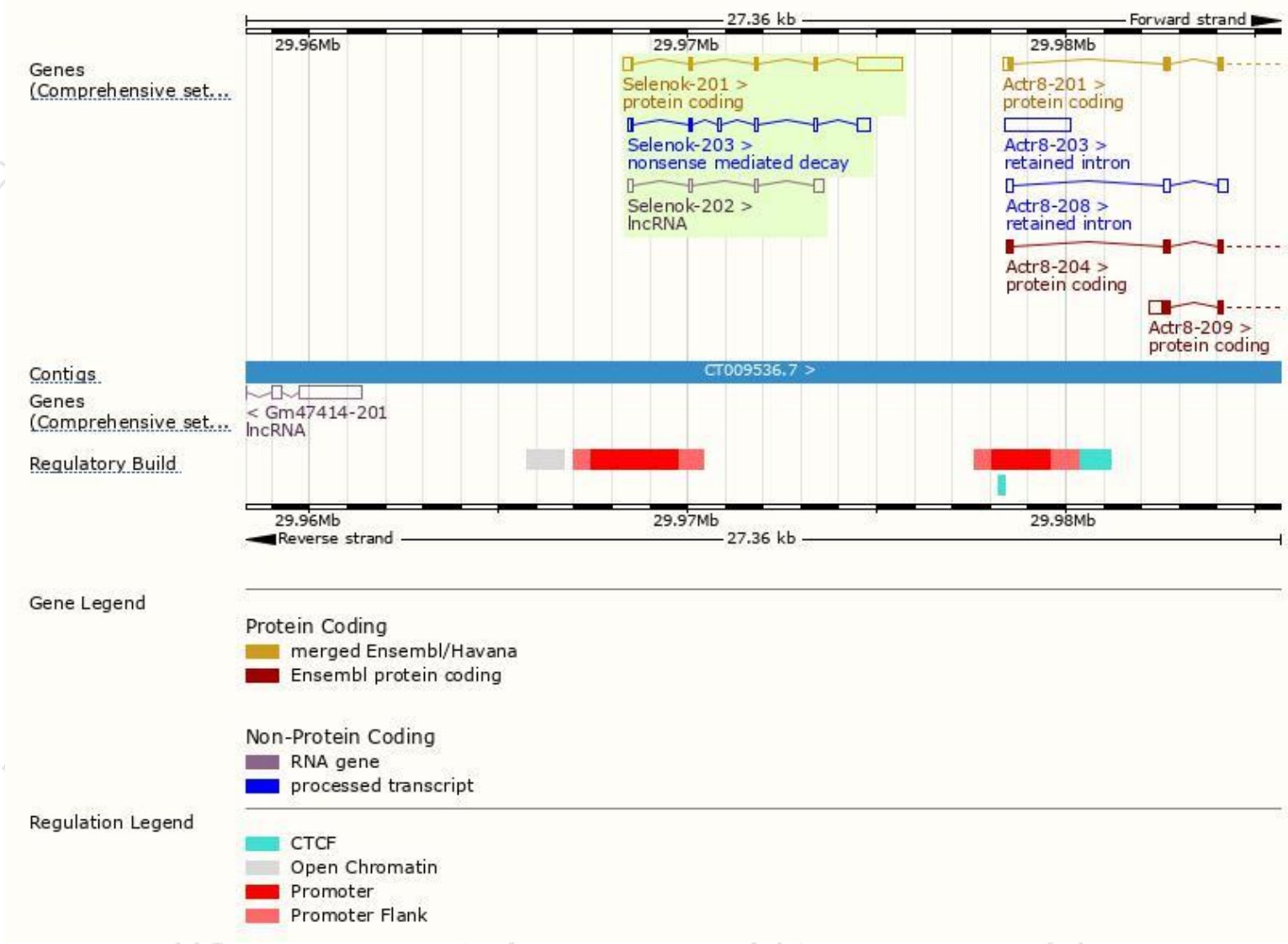
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Selenok-201	ENSMUST00000112268.2	1678	94aa	Protein coding	CCDS36844	Q9JLJ1	TSL:1 GENCODE basic APPRIS P1
Selenok-203	ENSMUST00000223998.1	799	40aa	Nonsense mediated decay	-	A0A286YD19	
Selenok-202	ENSMUST00000133229.1	513	No protein	lncRNA	-	-	TSL:2

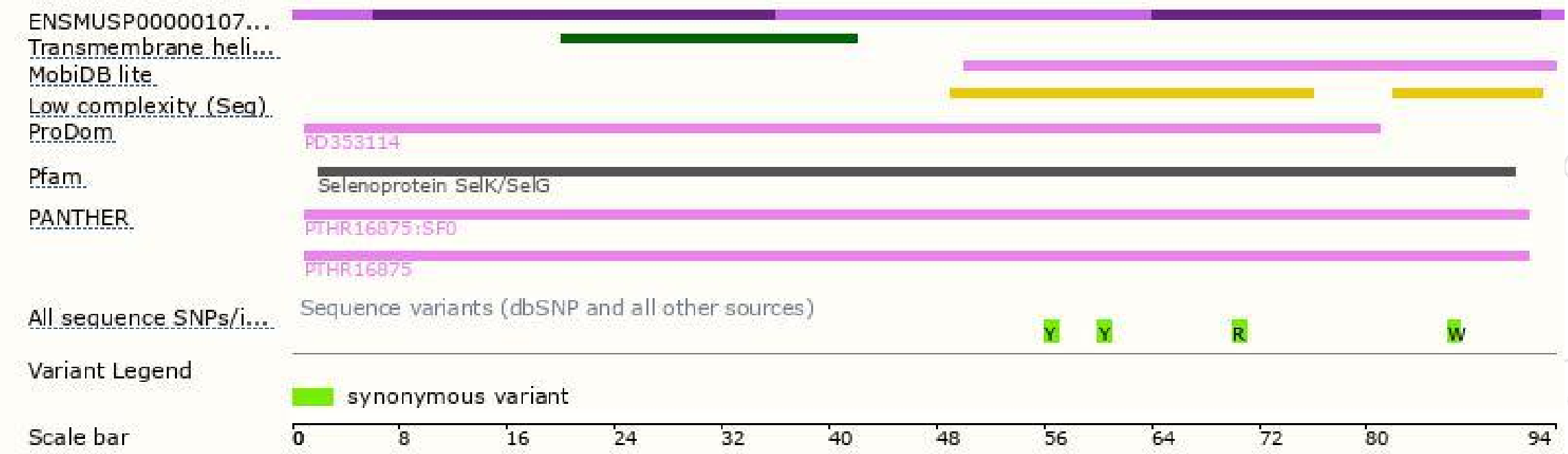
The strategy is based on the design of *Selenok-201* transcript,The transcription is shown below



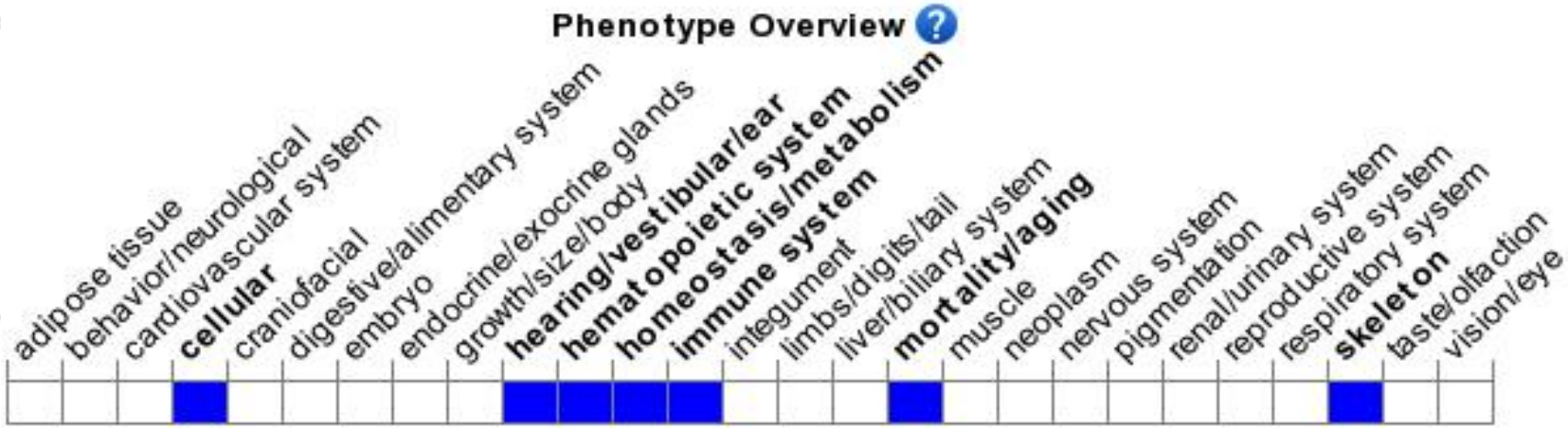
Genomic location distribution



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 impaired T cell, neutrophil, and macrophage calcium flux and increased susceptibility to viral infection.

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

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