Atp2b2 Cas9-CKO Strategy

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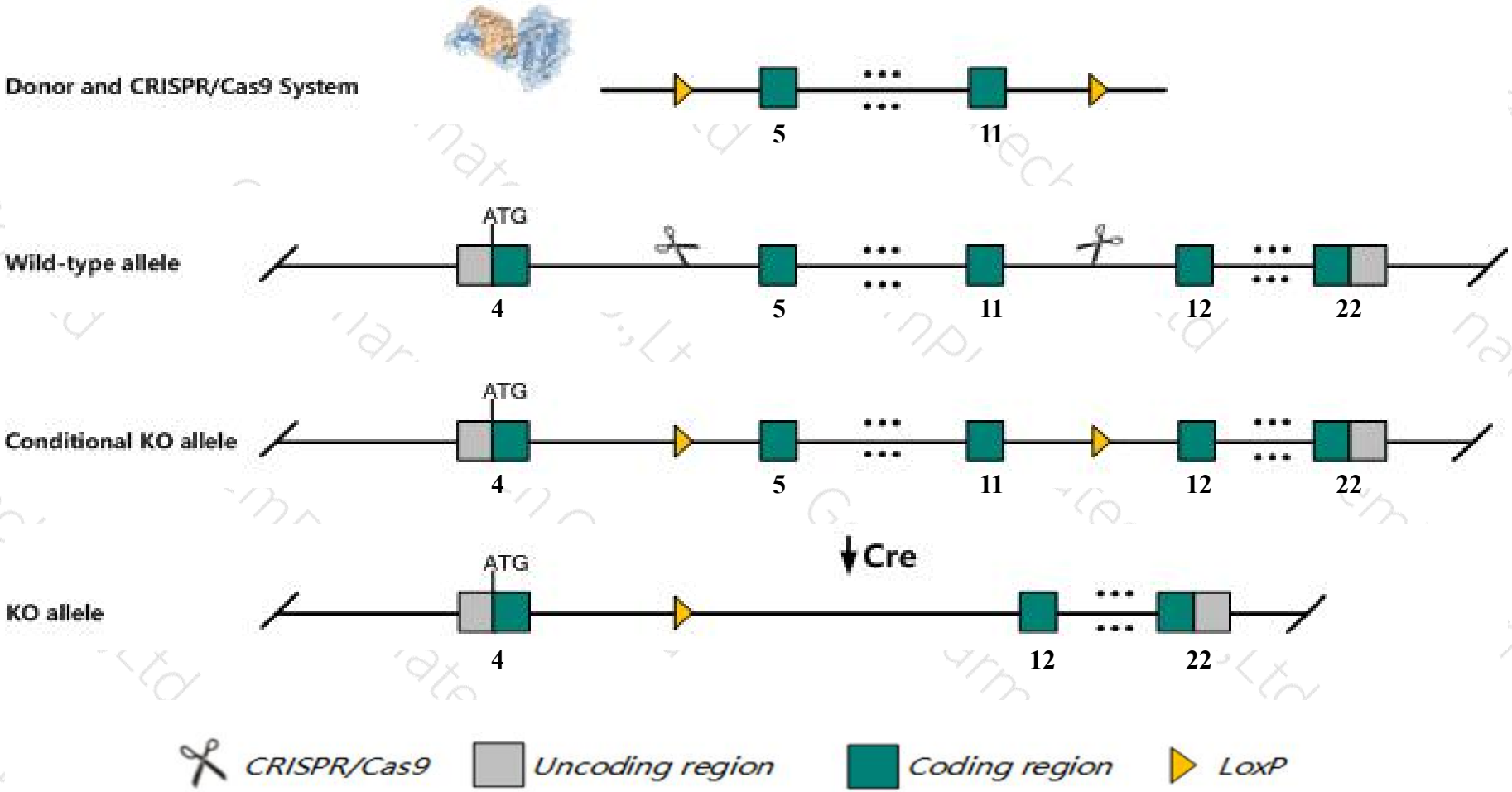
Project Overview

Project Name	***Atp2b2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Atp2b2* gene. The schematic diagram is as follows:



Technical routes

- The *Atp2b2* gene has 8 transcripts. According to the structure of *Atp2b2* gene, exon5-exon11 of *Atp2b2-203* (ENSMUST00000101045.9) transcript is recommended as the knockout region.The region contains 1325bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Atp2b2* 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,Homozygous mutants exhibit slower growth, balance problems, and deafness, associated with cerebellar abnormalities, an absence of otoconia, and abnormalities of the organ of Corti. Heterozygotes exhibit appreciable age-dependent hearing loss.
- The non-coding transcripts 205 and 207 are unaffected.
- *Gm44167*-201 will be deleted together.
- The *Atp2b2* 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）

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Atp2b2 ATPase, Ca++ transporting, plasma membrane 2 [Mus musculus (house mouse)]

Gene ID: 11941, updated on 7-Apr-2019

Summary

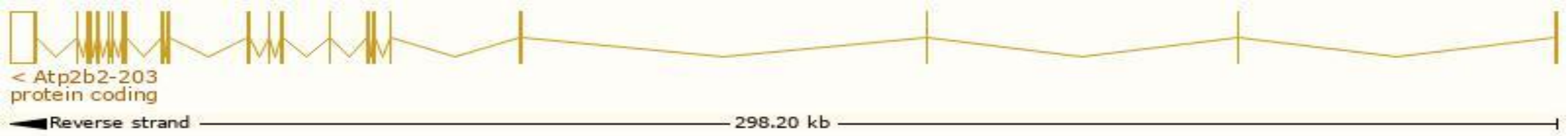
Official Symbol	Atp2b2 provided by MGI
Official Full Name	ATPase, Ca++ transporting, plasma membrane 2 provided by MGI
Primary source	MGI:MGI:105368
See related	Ensembl:ENSMUSG00000030302
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	D6Abb2e, Gena300, PMCA2, Tmy, dfw, jog, wms, wri
Expression	Biased expression in cortex adult (RPKM 49.9), cerebellum adult (RPKM 49.7) and 4 other tissues See more
Orthologs	human all

Transcript information（Ensembl）

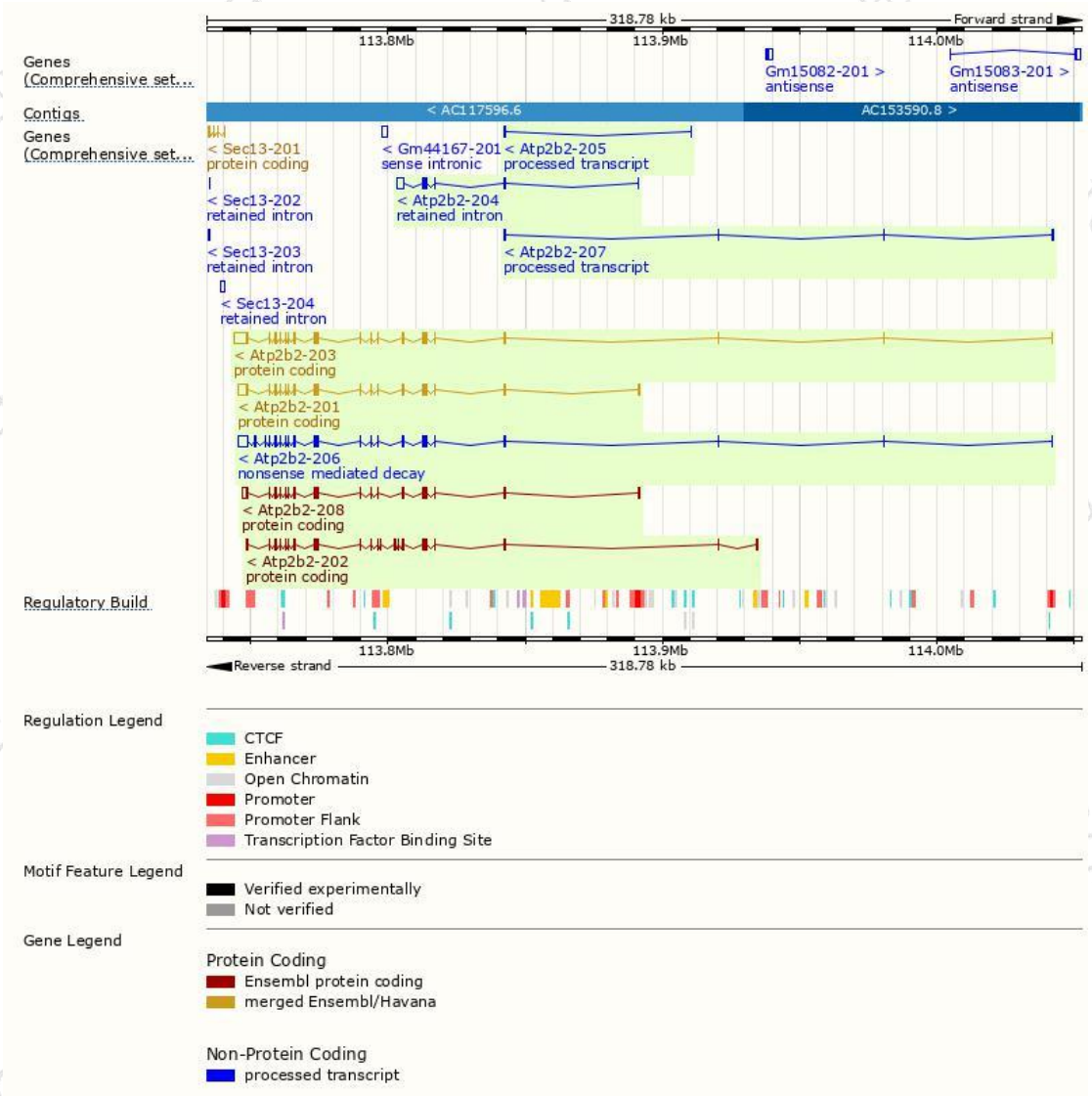
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Atp2b2-203	ENSMUST00000101045.9	8877	1198aa	Protein coding	CCDS20433	Q3UHJ3 Q9R0K7	TSL:1 GENCODE basic APPRIS P3
Atp2b2-201	ENSMUST00000089003.11	7067	1198aa	Protein coding	CCDS20433	Q3UHJ3 Q9R0K7	TSL:1 GENCODE basic APPRIS P3
Atp2b2-202	ENSMUST00000101044.8	4586	1243aa	Protein coding	CCDS85130	F8WHB1	TSL:1 GENCODE basic APPRIS ALT1
Atp2b2-208	ENSMUST00000205052.2	5658	1194aa	Protein coding	-	Q3UHH0	TSL:1 GENCODE basic APPRIS ALT1
Atp2b2-206	ENSMUST00000152831.7	7326	1154aa	Nonsense mediated decay	-	S4R1C4	TSL:1
Atp2b2-207	ENSMUST00000154738.1	659	No protein	Processed transcript	-	-	TSL:5
Atp2b2-205	ENSMUST00000144507.1	617	No protein	Processed transcript	-	-	TSL:1
Atp2b2-204	ENSMUST00000135199.1	3463	No protein	Retained intron	-	-	TSL:1

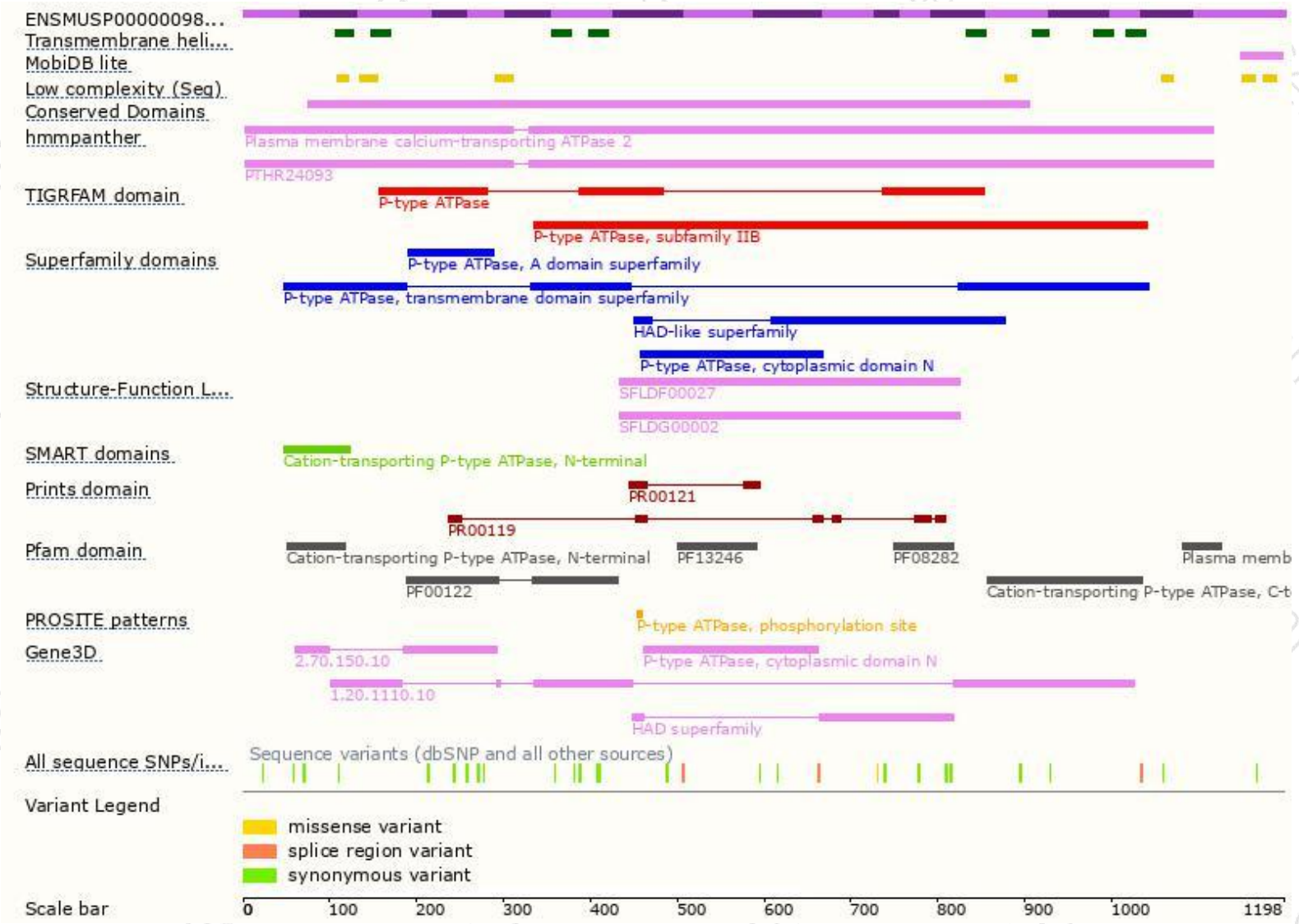
The strategy is based on the design of *Atp2b2-203* transcript,The transcription is shown below



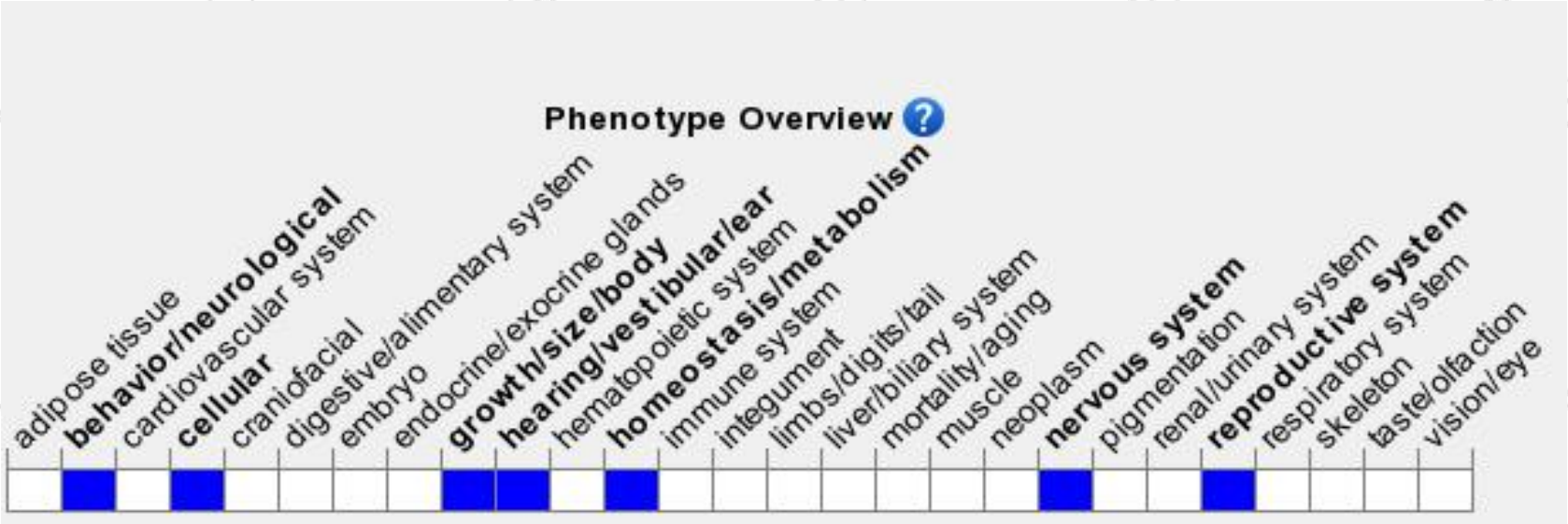
Genomic location distribution



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/).

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

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