Hmgb3 Cas9-CKO Strategy

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Reviewer: Ruirui Zhang

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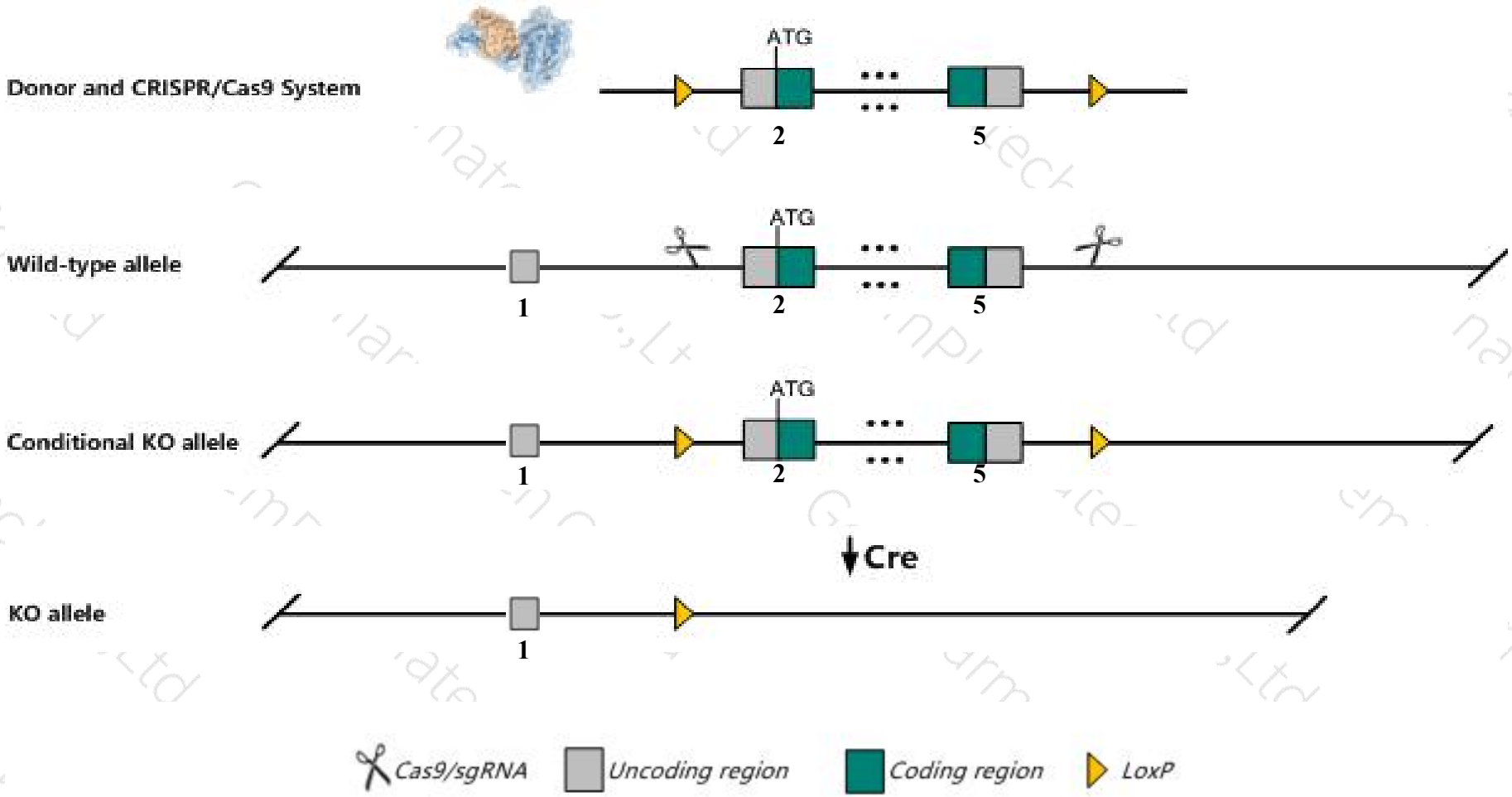
Project Overview

Project Name	***Hmgb3***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Hmgb3* gene. The schematic diagram is as follows:



Technical routes

- The *Hmgb3* gene has 6 transcripts. According to the structure of *Hmgb3* gene, exon2-exon5 of *Hmgb3-204* (ENSMUST00000114582.8) transcript is recommended as the knockout region.The region contains all of the coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Hmgb3* 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 null mice display increased red cell numbers, hematocrit, and hemaglobin content, and decreased mean red cell volume and common myeloid and lymphoid progenitors.

➢ The *Hmgb3* gene is located on the ChrX. 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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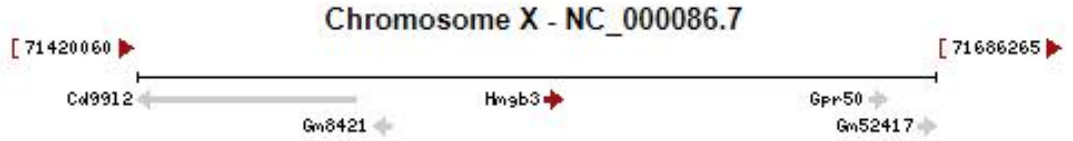
Hmgb3 high mobility group box 3 [*Mus musculus* (house mouse)]

Gene ID: 15354, updated on 10-Oct-2019

Summary

Official Symbol	Hmgb3 provided by MGI
Official Full Name	high mobility group box 3 provided by MGI
Primary source	MGI:MGI:1098219
See related	Ensembl:ENSMUSG00000015217
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	Hmg4; Hmg2a
Expression	Biased expression in CNS E11.5 (RPKM 187.0), CNS E14 (RPKM 108.6) and 11 other tissues See more
Orthologs	human all

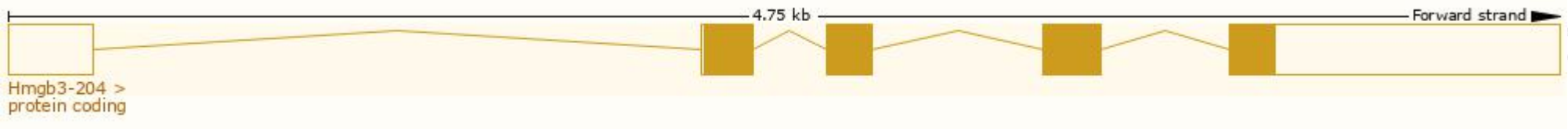


Transcript information（Ensembl）

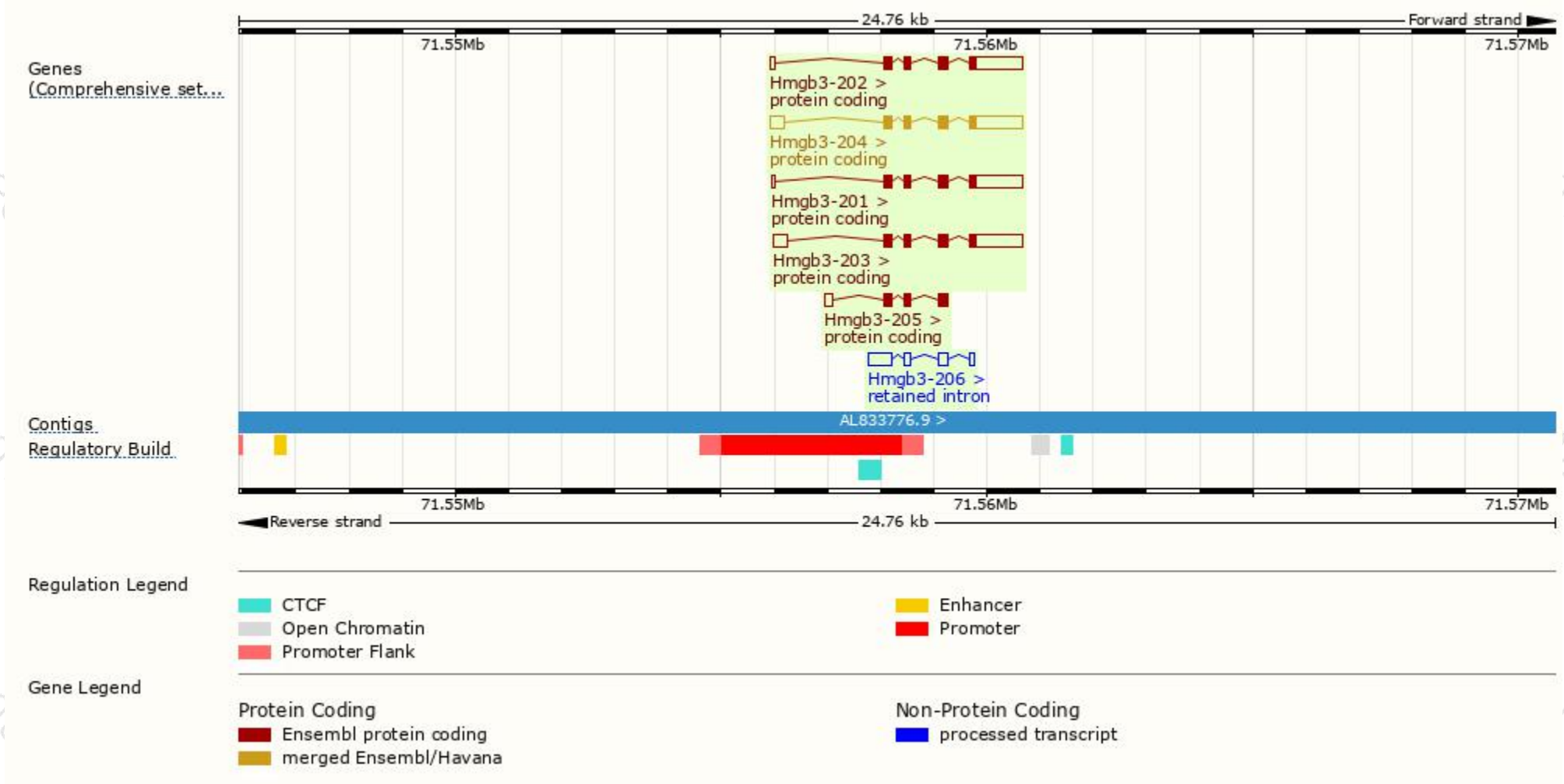
The gene has 6 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Hmgb3-204	ENSMUST00000114582.8	1743	200aa	Protein coding	CCDS30180	O54879 Q544R9	TSL:1 GENCODE basic APPRIS P1
Hmgb3-203	ENSMUST00000088874.9	1728	200aa	Protein coding	CCDS30180	O54879 Q544R9	TSL:2 GENCODE basic APPRIS P1
Hmgb3-202	ENSMUST00000072699.12	1571	200aa	Protein coding	CCDS30180	O54879 Q544R9	TSL:1 GENCODE basic APPRIS P1
Hmgb3-201	ENSMUST00000015361.10	1544	200aa	Protein coding	CCDS30180	O54879 Q544R9	TSL:1 GENCODE basic APPRIS P1
Hmgb3-205	ENSMUST00000123100.1	634	159aa	Protein coding	-	A2AP78	CDS 3' incomplete TSL:5
Hmgb3-206	ENSMUST00000132145.1	847	No protein	Retained intron	-	-	TSL:1

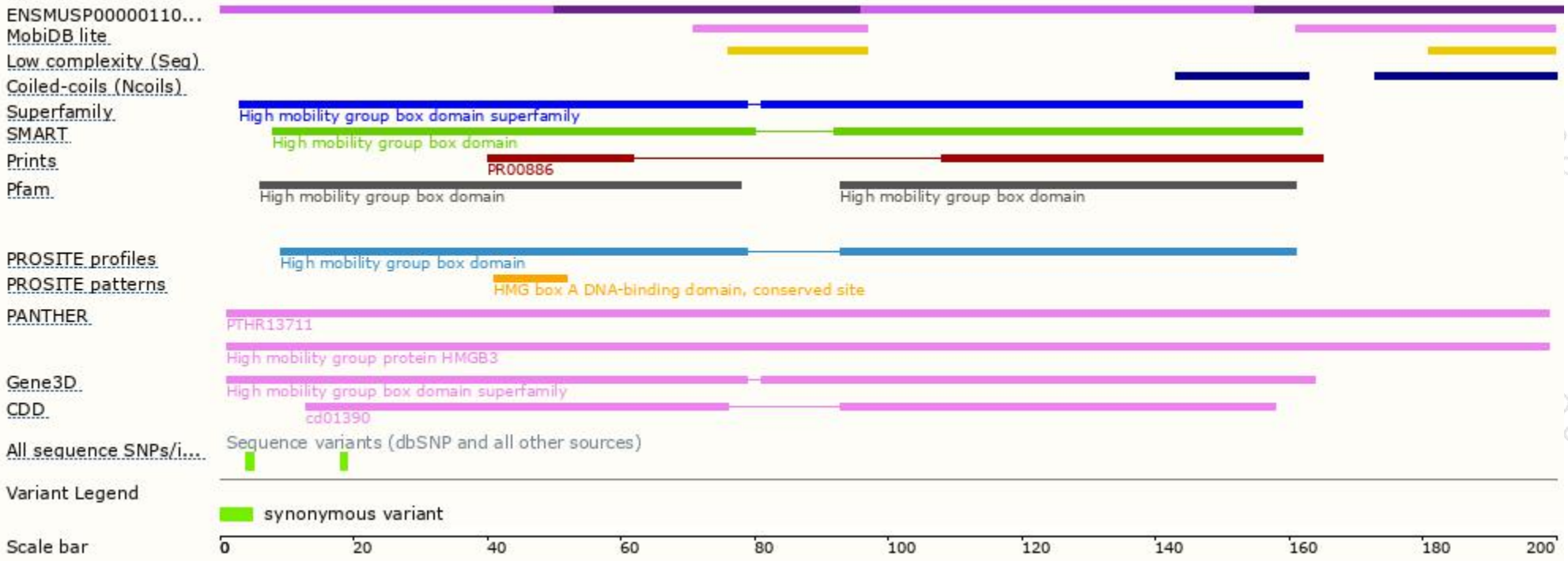
The strategy is based on the design of *Hmgb3-204* transcript,The transcription is shown below



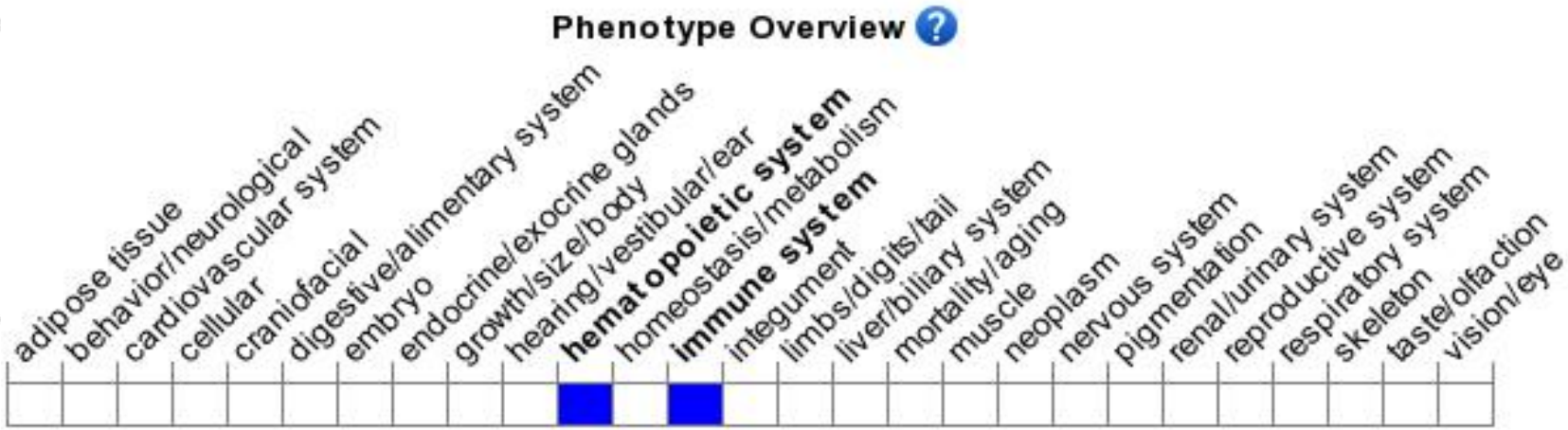
Genomic location distribution



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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