Nckap1 Cas9-CKO Strategy

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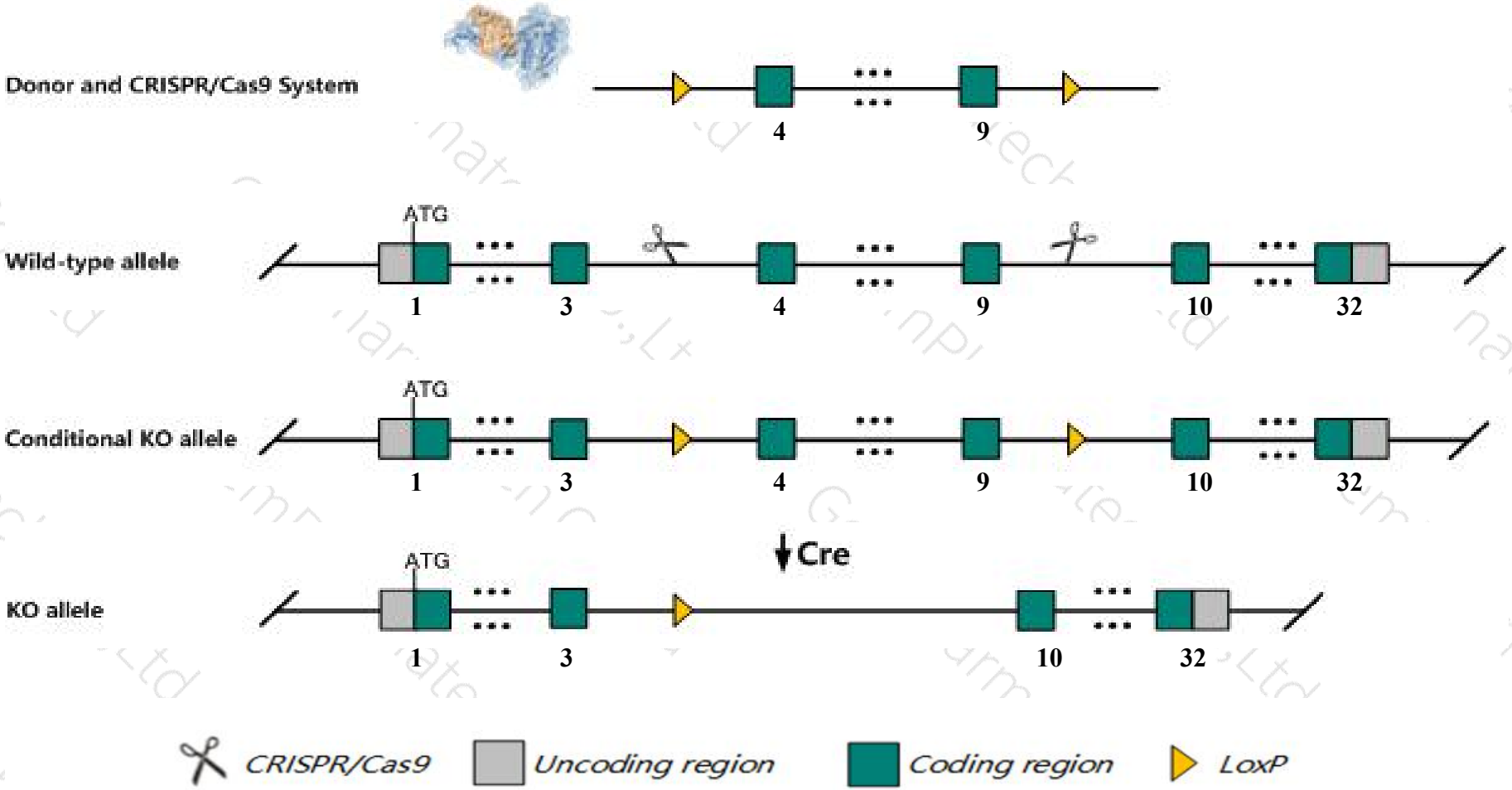
Design Date: 2019-10-18

Project Overview

Project Name	***Nckap1***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Nckap1* gene. The schematic diagram is as follows:



Technical routes

- The *Nckap1* gene has 5 transcripts. According to the structure of *Nckap1* gene, exon4-exon9 of *Nckap1-202* (ENSMUST00000111760.2) transcript is recommended as the knockout region.The region contains 571bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Nckap1* 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 disruptions in this gene exhibit growth arrest at midgestation, an open neural tube, cardia bifida, defective foregut development, defects in endoderm and mesoderm migration and sometimes duplication of the anteroposterior body axis.
- The KO region contains functional region of the *Gm13689* gene.Knockout the region may affect the function of *Gm13689* gene.
- The *Nckap1* gene is located on the Chr2. 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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Nckap1 NCK-associated protein 1 [Mus musculus (house mouse)]

Gene ID: 50884, updated on 7-Apr-2019

Summary

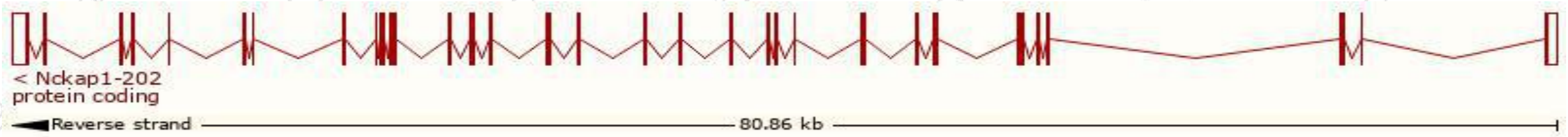
Official Symbol	Nckap1 provided by MGI
Official Full Name	NCK-associated protein 1 provided byMGI
Primary source	MGI:MGI:1355333
See related	Ensembl:ENSMUSG00000027002
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	C79304, H19, Hem-2, Hem2, Nap1, mKIAA0587, mh19, p125Nap1
Expression	Ubiquitous expression in cortex adult (RPKM 59.6), frontal lobe adult (RPKM 57.9) and 26 other tissuesSee more
Orthologs	human all

Transcript information（Ensembl）

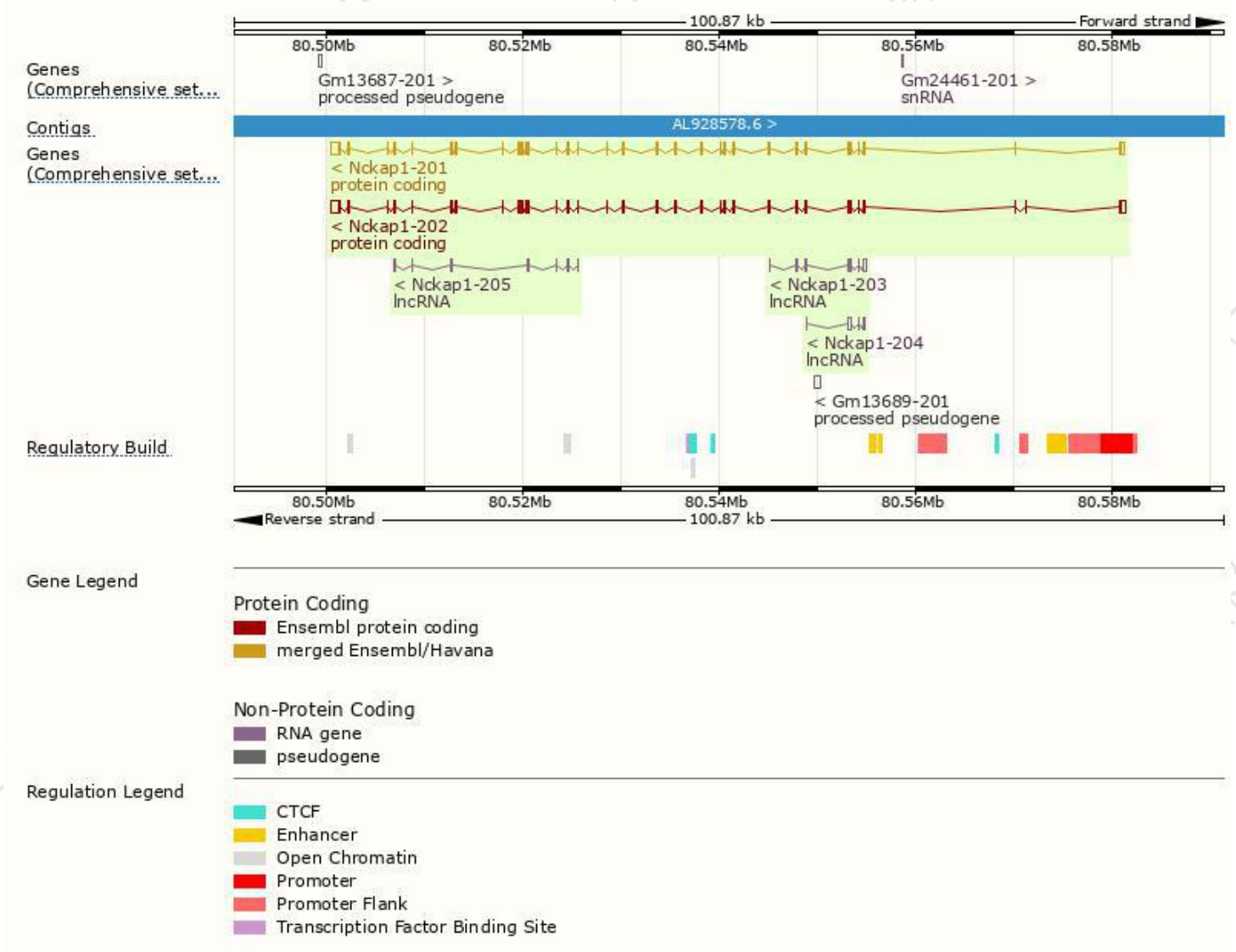
The gene has 5 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nckap1-202	ENSMUST00000111760.2	4597	1134aa	Protein coding	CCDS71082	A2AS98	TSL:1 GENCODE basic APPRIS ALT1
Nckap1-201	ENSMUST00000028386.11	4469	1128aa	Protein coding	CCDS16177	P28660	TSL:1 GENCODE basic APPRIS P3
Nckap1-203	ENSMUST00000131872.7	885	No protein	lncRNA	-	-	TSL:2
Nckap1-205	ENSMUST00000154793.1	687	No protein	lncRNA	-	-	TSL:5
Nckap1-204	ENSMUST00000134587.1	498	No protein	lncRNA	-	-	TSL:5

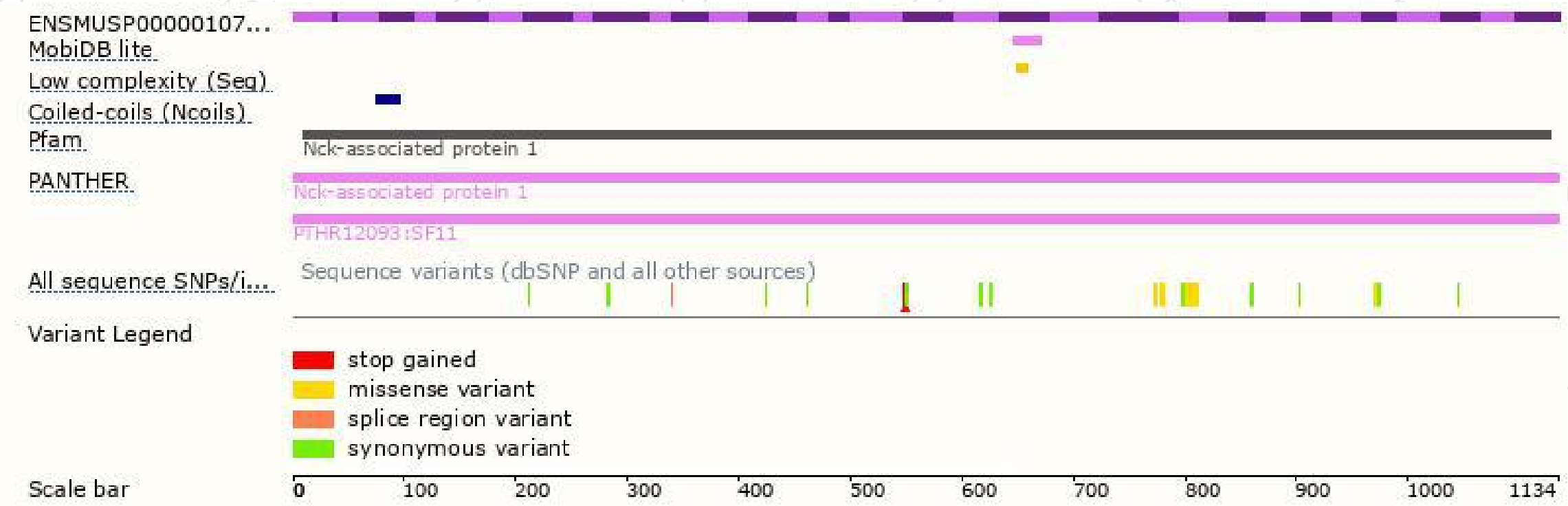
The strategy is based on the design of *Nckap1-202* transcript,The transcription is shown below



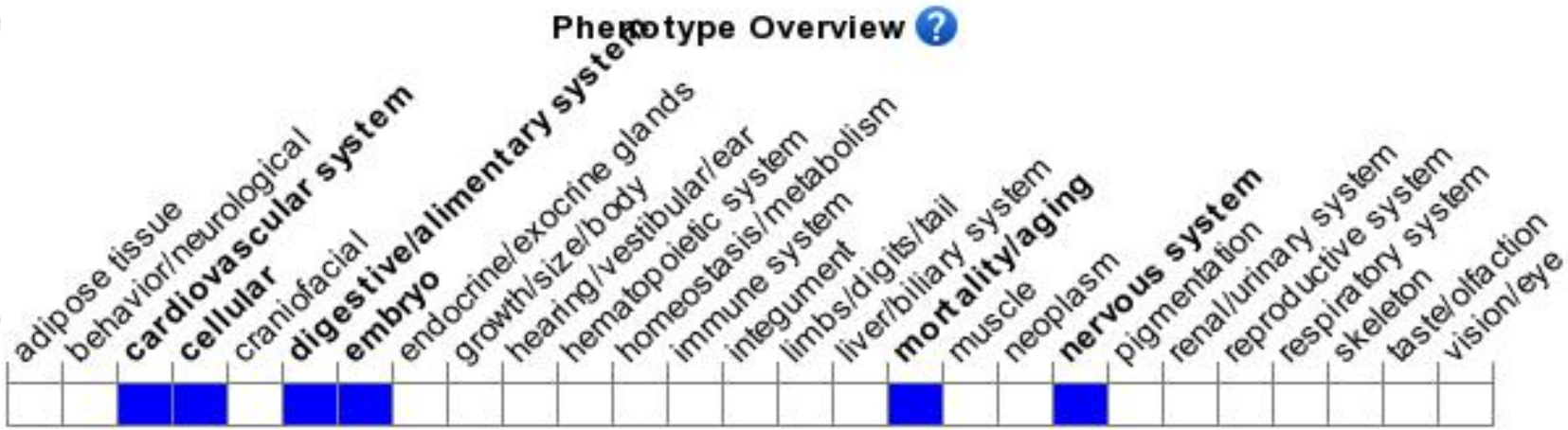
Genomic location distribution



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 disruptions in this gene exhibit growth arrest at midgestation, an open neural tube, cardia bifida, defective foregut development, defects in endoderm and mesoderm migration sometimes duplication of the anteroposterior body axis.

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

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