

Ambr1 Cas9-KO Strategy

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Design Date: 2020-7-22

Project Overview

Project Name

Ambra1

Project type

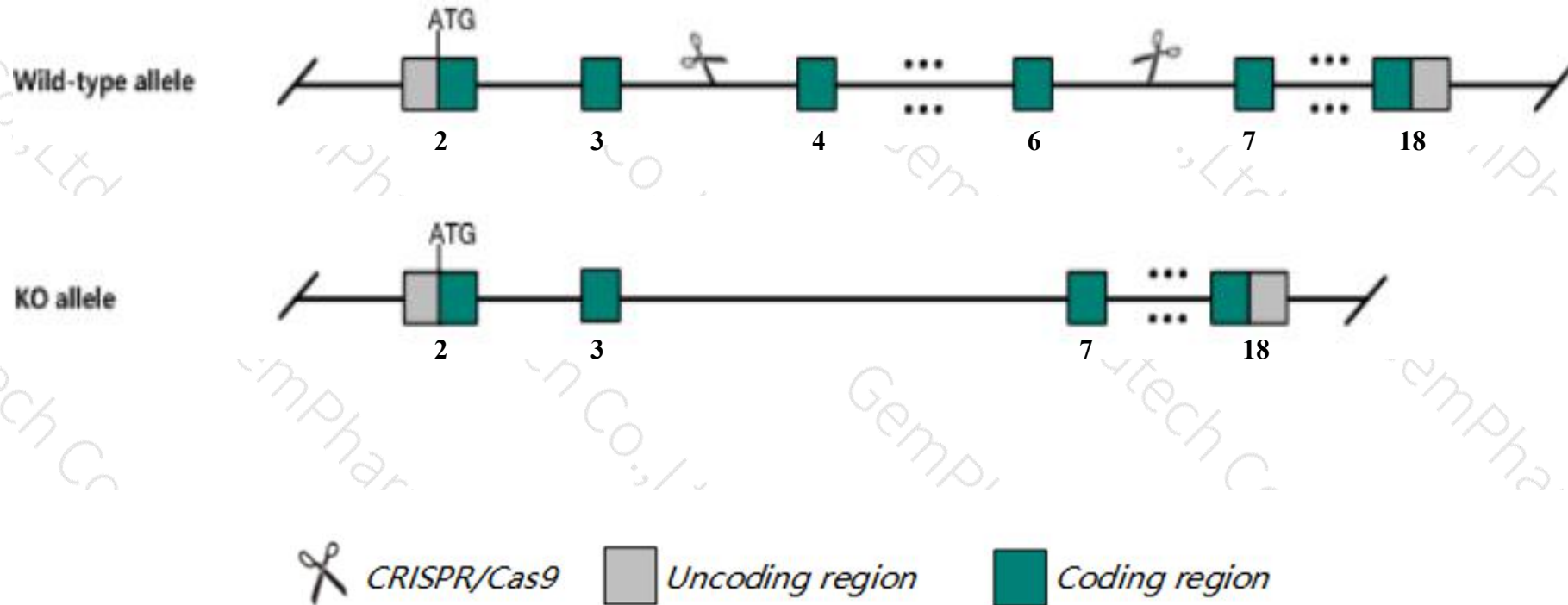
Cas9-KO

Strain background

C57BL/6JGpt

Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Ambra1* gene. The schematic diagram is as follows:



- The *Ambra1* gene has 9 transcripts. According to the structure of *Ambra1* gene, exon4-exon6 of *Ambra1*-202(ENSMUST00000045705.13) transcript is recommended as the knockout region. The region contains 424bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Ambra1* gene. The brief process is as follows: gRNA was transcribed in vitro. Cas9 and gRNA 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.

- According to the existing MGI data, most mice homozygous for a gene trap mutation die at E10-E14.5 with severe neural tube defects manifest as midbrain/hindbrain exencephaly and/or spina bifida and associated with impaired autophagy, accumulation of ubiquitinated proteins, abnormal cell proliferation and excessive apoptosis.
- The *Ambra1* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

Ambra1 autophagy/beclin 1 regulator 1 [*Mus musculus* (house mouse)]

Gene ID: 228361, updated on 7-Jul-2020

Summary

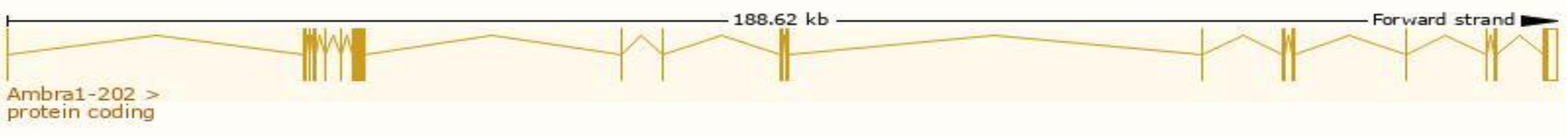
Official Symbol	Ambra1 provided by MGI
Official Full Name	autophagy/beclin 1 regulator 1 provided by MGI
Primary source	MGI:MGI:2443564
See related	Ensembl:ENSMUSG00000040506
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	AA474864; AV021921; mKIAA1736; A130023A14; 2310079H06Rik; D030051N19Rik
Expression	Ubiquitous expression in testis adult (RPKM 13.4), adrenal adult (RPKM 12.3) and 28 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

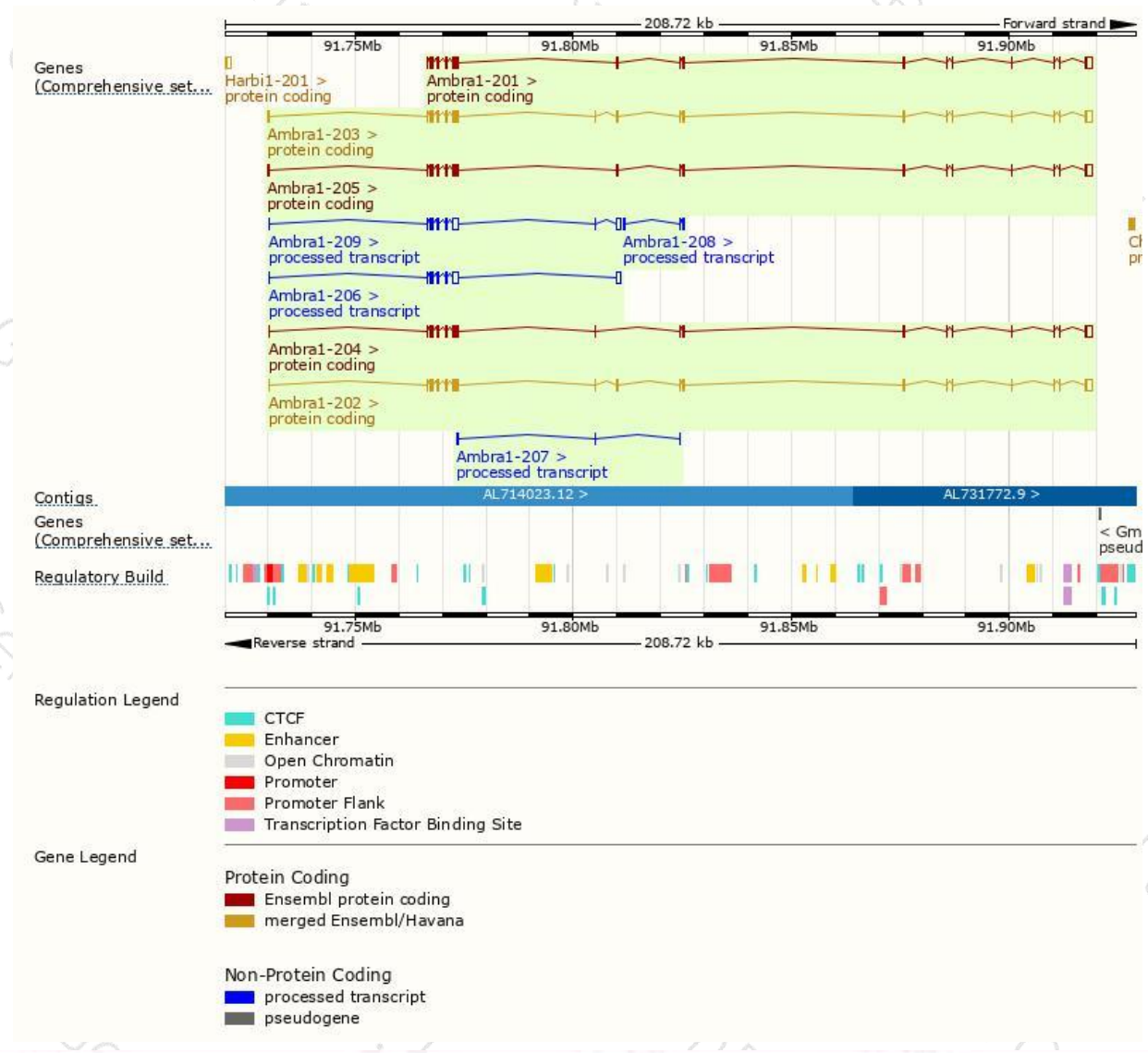
The gene has 9 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Ambra1-202	ENSMUST00000045705.13	5203	1300aa	Protein coding	CCDS38179	A2AH22	TSL:1 GENCODE basic APPRIS P4
Ambra1-203	ENSMUST00000099712.9	5021	1209aa	Protein coding	CCDS38180	A2AH22	TSL:1 GENCODE basic APPRIS ALT1
Ambra1-204	ENSMUST00000111316.8	5063	1240aa	Protein coding	-	A2AH22	TSL:5 GENCODE basic APPRIS ALT2
Ambra1-205	ENSMUST00000111317.8	4929	1180aa	Protein coding	-	A2AH22	TSL:5 GENCODE basic APPRIS ALT1
Ambra1-201	ENSMUST00000045699.7	4690	1180aa	Protein coding	-	A2AH22	TSL:5 GENCODE basic APPRIS ALT1
Ambra1-209	ENSMUST00000156496.7	3419	No protein	Processed transcript	-	-	TSL:1
Ambra1-206	ENSMUST00000124132.7	3332	No protein	Processed transcript	-	-	TSL:1
Ambra1-207	ENSMUST00000133490.1	663	No protein	Processed transcript	-	-	TSL:5
Ambra1-208	ENSMUST00000142224.1	248	No protein	Processed transcript	-	-	TSL:5

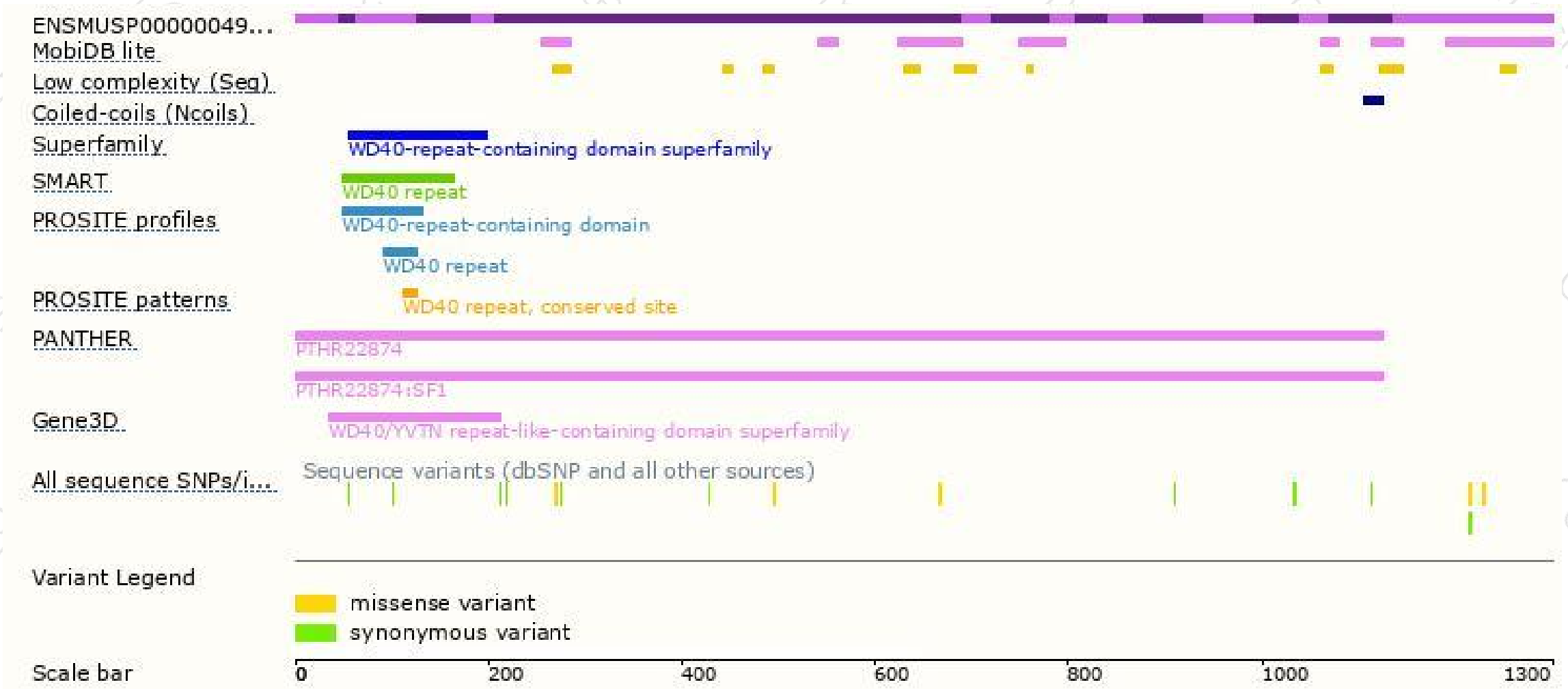
The strategy is based on the design of *Ambra1-202* transcript,the transcription is shown below:



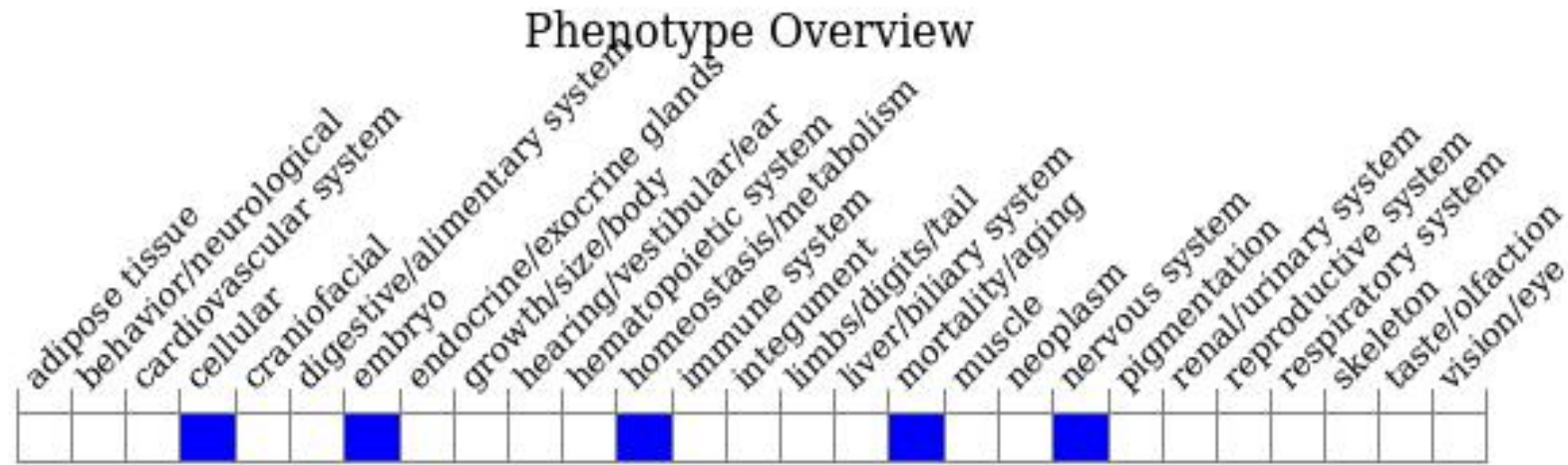
Genomic location distribution



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, most mice homozygous for a gene trap mutation die at E10-E14.5 with severe neural tube defects manifest as midbrain/hindbrain exencephaly and/or spina bifida and associated with impaired autophagy, accumulation of ubiquitinated proteins, abnormal cell proliferation and excessive apoptosis.

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

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