



Nostrin Cas9-CKO Strategy

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Reviewer: Yanhua Shen
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Project Overview

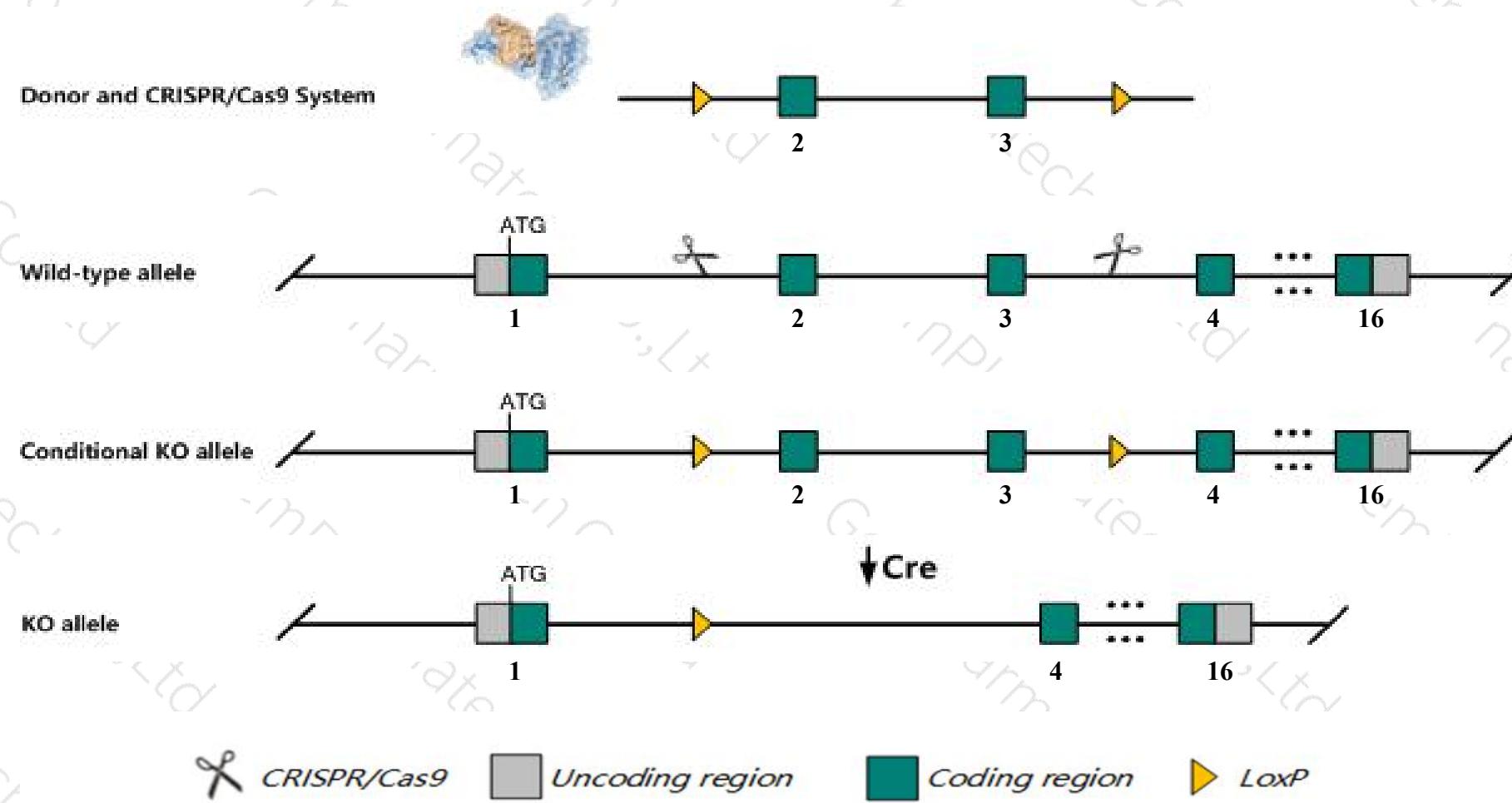
Project Name**Nostrin**

Project type**Cas9-CKO**

Strain background**C57BL/6JGpt**

Conditional Knockout strategy

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



Technical routes

- The *Nostrin* gene has 2 transcripts. According to the structure of *Nostrin* gene, exon2-exon3 of *Nostrin-201* (ENSMUST00000041865.7) transcript is recommended as the knockout region. The region contains 170bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Nostrin* 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.



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Notice

- According to the existing MGI data, Mice homozygous for a knock-out allele exhibit impaired retinal vascular angiogenesis, endothelial cell proliferation, endothelial cell migration and induced neovascularization.
- Transcript *Nostrin*-202 may not be affected.
- The *Nostrin* 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)

Nostrin nitric oxide synthase trafficker [*Mus musculus* (house mouse)]

Gene ID: 329416, updated on 10-Oct-2019

Summary



Official Symbol Nostrin provided by MGI

Official Full Name nitric oxide synthase trafficker provided by MGI

Primary source MGI:MGI:3606242

See related Ensembl:ENSMUSG0000034738

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 Daip2; mDaIP2

Expression Biased expression in placenta adult (RPKM 11.0), large intestine adult (RPKM 4.4) and 11 other tissues [See more](#)

Orthologs [human](#) [all](#)

Genomic context



Location: 2; 2 C2

[See Nostrin in Genome Data Viewer](#)

Exon count: 16

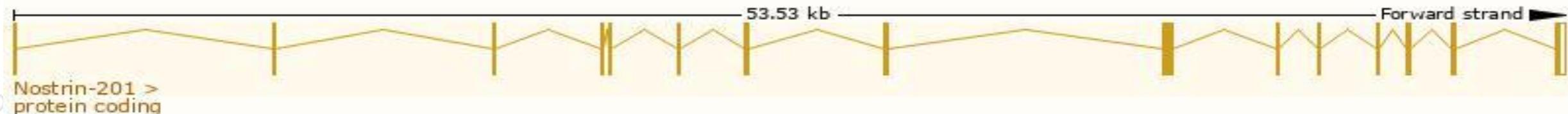
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	2	NC_000068.7 (69135800..69189330)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	2	NC_000068.6 (68973857..69027387)

Transcript information (Ensembl)

The gene has 2 transcripts, all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Nostrin-201	ENSMUST00000041865.7	1808	506aa	Protein coding	CCDS16087	Q6WKZ7	TSL:1 GENCODE basic APPRIS P1
Nostrin-202	ENSMUST00000141276.1	609	No protein	lncRNA	-	-	TSL:3

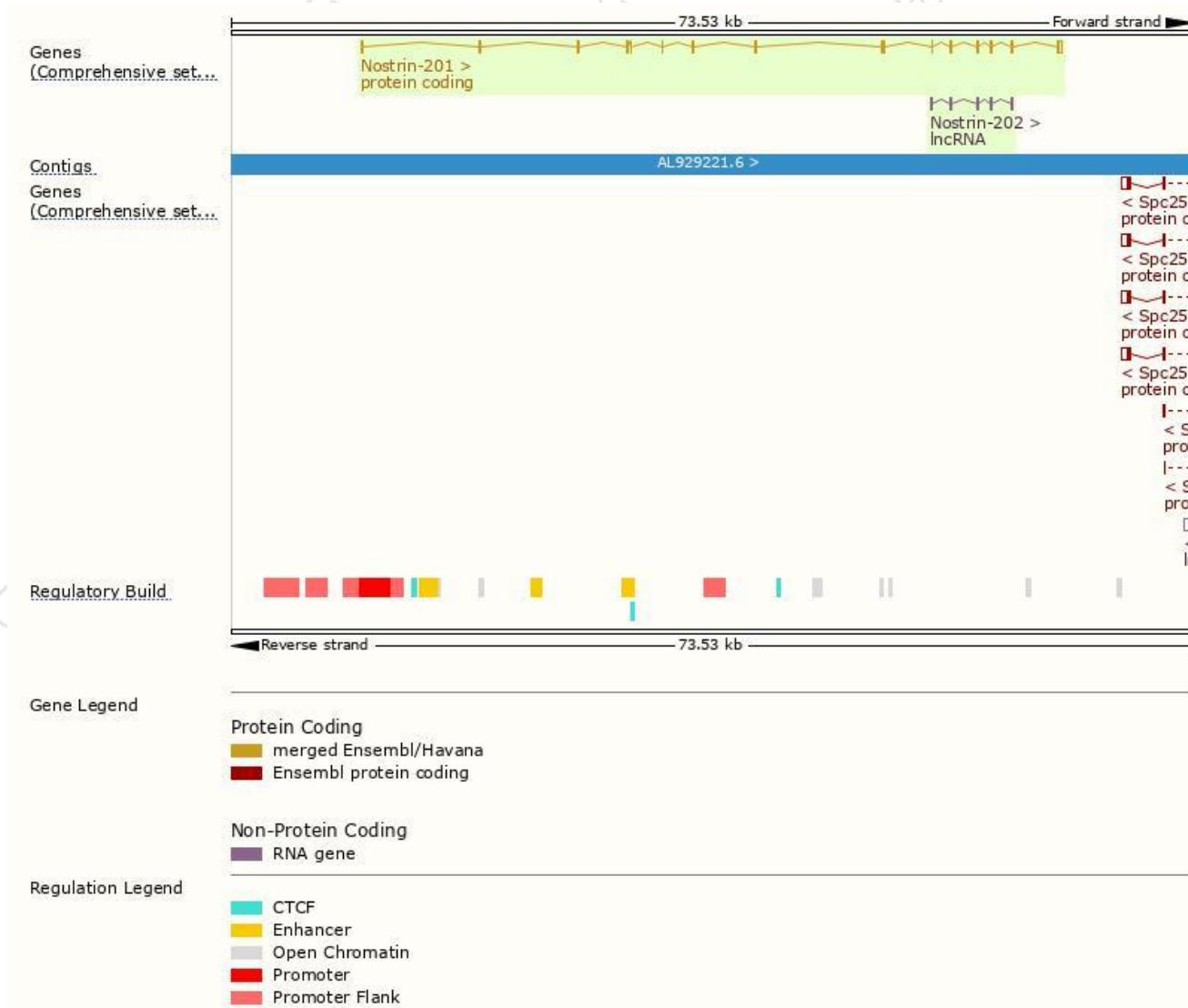
The strategy is based on the design of *Nostrin-201* transcript, The transcription is shown below



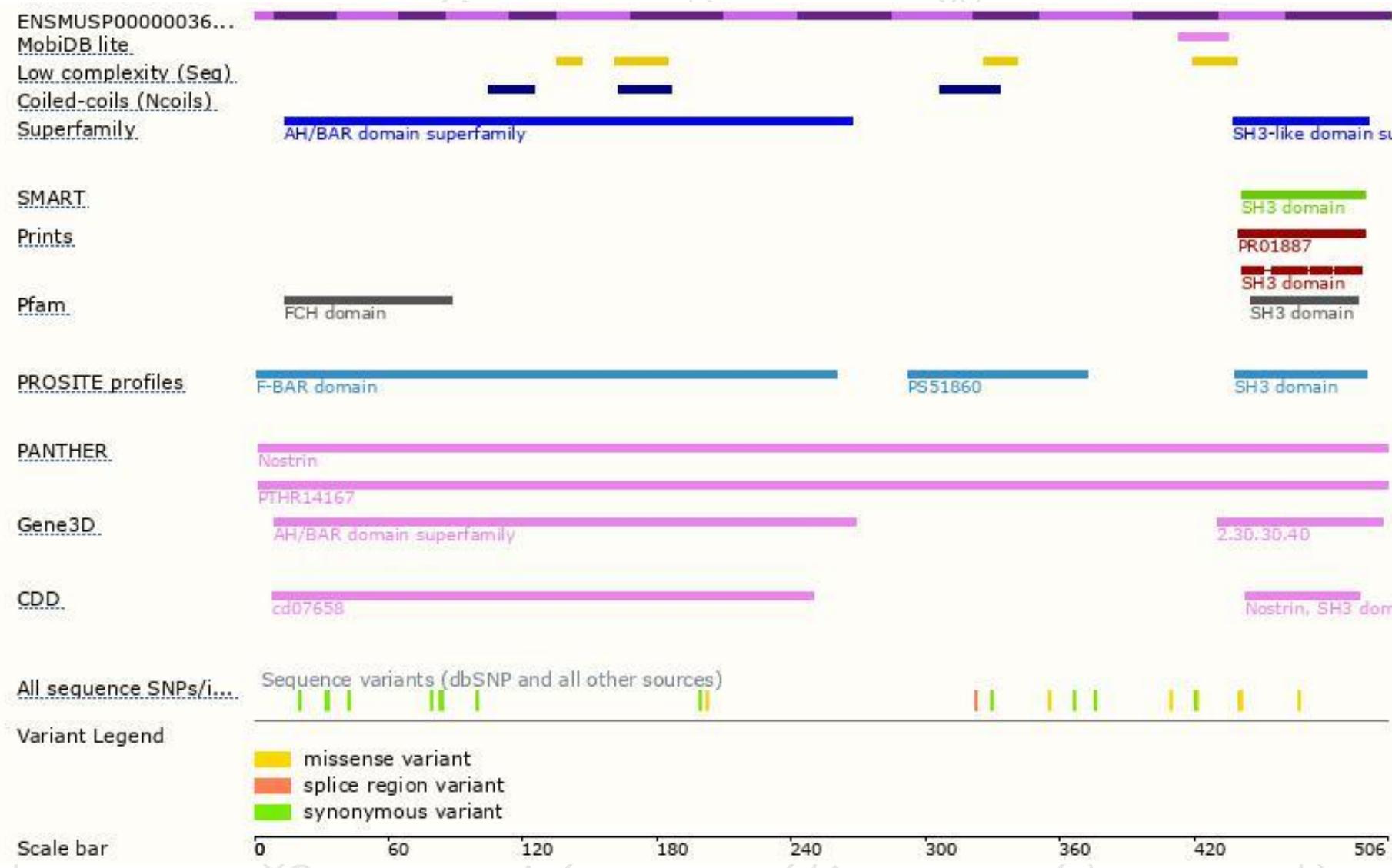


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Genomic location distribution



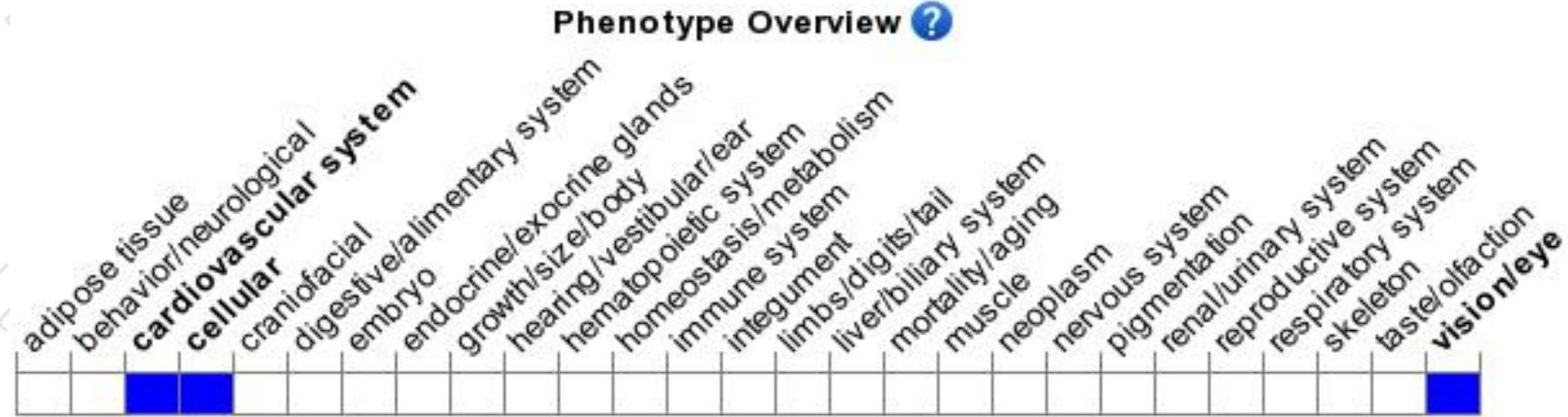
Protein domain





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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 retinal vascular angiogenesis, endothelial cell proliferation, endothelial cell migration and induced neovascularization.



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

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