

# Phex Cas9-KO Strategy

Designer: Daohua Xu

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# **Project Overview**



Project Name Phex

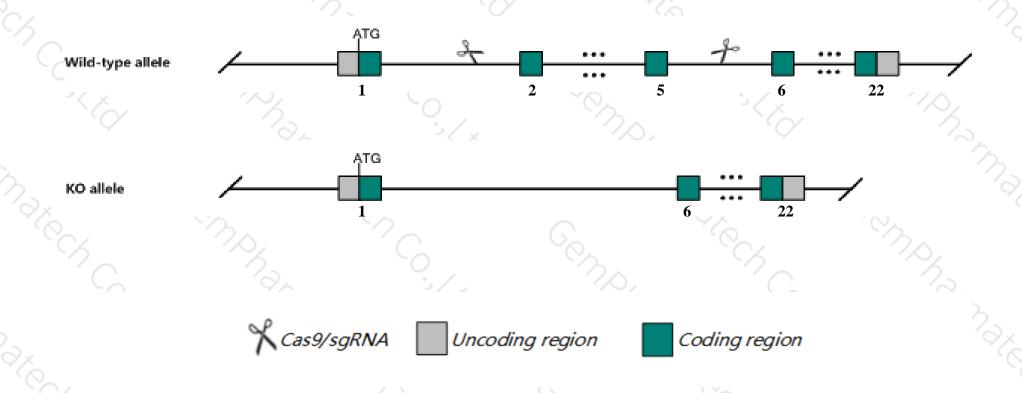
Project type Cas9-KO

Strain background C57BL/6JGpt

# **Knockout strategy**



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



### **Technical routes**



- ➤ The *Phex* gene has 3 transcripts. According to the structure of *Phex* gene, exon2-exon5 of *Phex-201*(ENSMUST00000079945.10) transcript is recommended as the knockout region. The region contains 545bp coding sequence Knock out the region will result in disruption of protein function.
- ➤ In this project we use CRISPR/Cas9 technology to modify *Phex* gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA 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.

### **Notice**



- ➤ According to the existing MGI data, Males hemizygous for a null mutation exhibit reduced body size, shortened hindlimbs and tail, osteomalacia, and markedly reduced plasma phosphate levels due to impaired kidney reabsorption. Female heterozygotes exhibit milder symptoms.
- > The *Phex* 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 the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

# Gene information (NCBI)



#### Phex phosphate regulating endopeptidase homolog, X-linked [Mus musculus (house mouse)]

Gene ID: 18675, updated on 3-Feb-2019

#### Summary



Official Symbol Phex provided by MGI

Official Full Name phosphate regulating endopeptidase homolog, X-linked provided by MGI

Primary source MGI:MGI:107489

See related Ensembl:ENSMUSG00000057457

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 Gy, HPDR, HPDR1, Hyp, PEX

Expression Biased expression in limb E14.5 (RPKM 1.5), CNS E18 (RPKM 0.2) and 7 other tissuesSee more

Orthologs <u>human</u> all

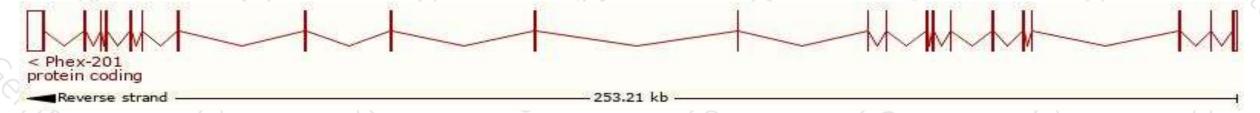
## Transcript information (Ensembl)



The gene has 3 transcripts, all transcripts are shown below:

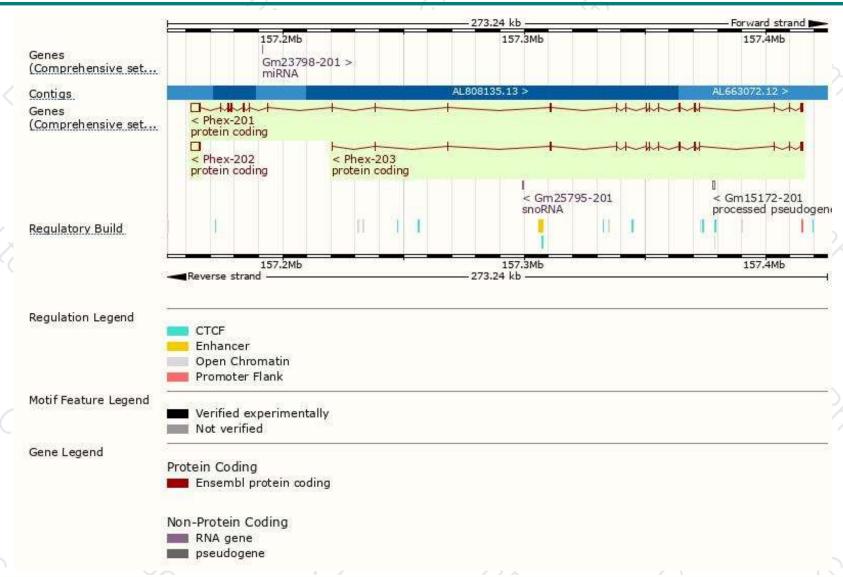
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Phex-201	ENSMUST00000079945.10	6265	<u>749aa</u>	Protein coding	CCDS30497	P70669 Q3TYM9	TSL:1 GENCODE basic APPRIS P1
Phex-202	ENSMUST00000135713.7	3588	<u>34aa</u>	Protein coding	-	<u>F6S0V7</u>	CDS 5' incomplete TSL:NA
Phex-203	ENSMUST00000138396.2	2201	<u>548aa</u>	Protein coding	-	A2AC80	CDS 3' incomplete TSL:5

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



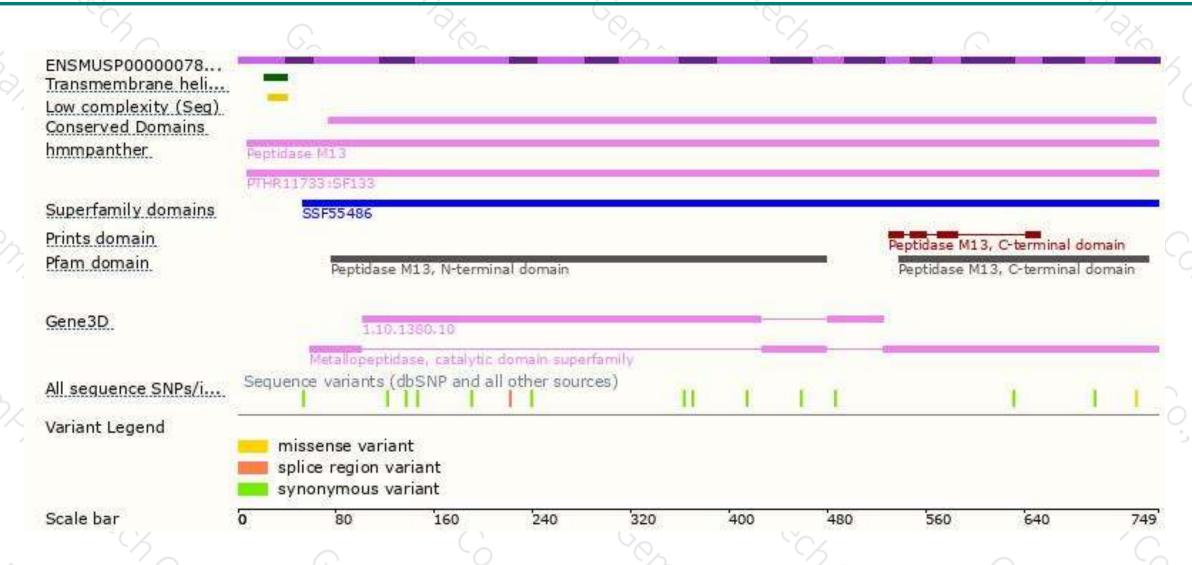
### Genomic location distribution





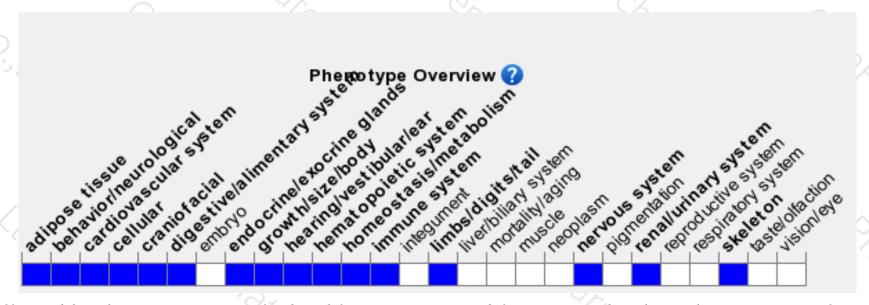
### Protein domain





### 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, Males hemizygous for a null mutation exhibit reduced body size, shortened hindlimbs and tail, osteomalacia, and markedly reduced plasma phosphate levels due to impaired kidney reabsorption. Female heterozygotes exhibit milder symptoms.



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

Tel: 025-5864 1534





