

# ***Pomc Cas9-KO Strategy***

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**Reviewer :**

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**Design Date:**

**2019-9-28**

# Project Overview

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**Project Name**

*Pomc*

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**Project type**

Cas9-KO

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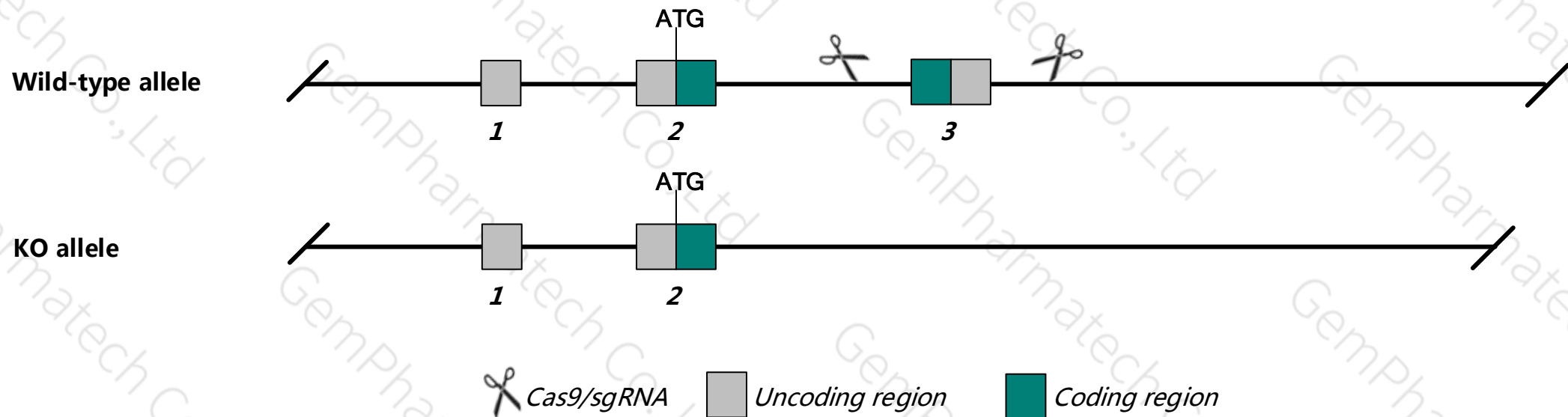
**Animal background**

C57BL/6JGpt

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# Knockout strategy

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



# Technical routes

- The *Pomc* gene has 5 transcripts, According to the structure of *Pomc* gene, exon3 of *Pomc-201* transcript is recommended as the knockout region. The region contains the most of coding sequence. Knock out the region, result in destruction of protein.
- This project uses CRISPR/Cas9 technology to modify *Pomc* gene. The brief process is as follows: sgRNA was transcribed in vitro, Cas9, sgRNA were microinjected into fertilized eggs of C57BL/6JGpt mice and homologous recombination was carried out to obtain F0 mice. A stable and hereditary F1 generation mouse model was obtained by mating F0 generation mice with C57BL/6JGpt mice which were confirmed positive by PCR-sequencing.

- According to the existing MGI data , Homozygotes for a targeted null mutation are obese and exhibit abnormal hormone levels, abnormal pigmentation, increased food intake, and adiposity. Mice homozygous for another knock-out allele exhibit altered reward based behavior and immune response to LPS treatment.
- The *Pomc* gene is located in the Chr12. If the knockout mice are mixed with other mice, two target genes are avoided on the same chromosome as possible, otherwise the offspring of mice with double gene positive and homozygous gene knockout can not be obtained.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of gene transcription and translation processes, all risks cannot be predicted under existing information.

# Gene information ( NCBI )

## Pomc pro-opiomelanocortin-alpha [ *Mus musculus* (house mouse) ]

Gene ID: 18976, updated on 23-Dec-2018

### Summary

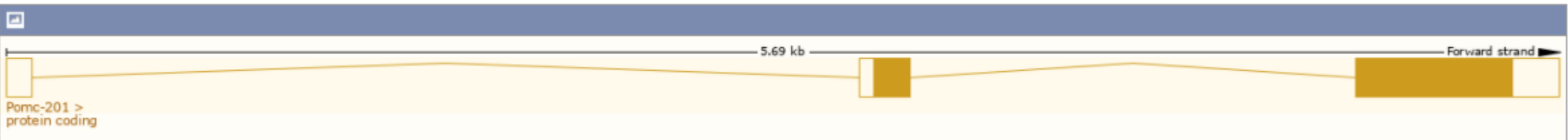
Official Symbol	Pomc provided by <a href="#">MGI</a>
Official Full Name	pro-opiomelanocortin-alpha provided by <a href="#">MGI</a>
Primary source	<a href="#">MGI:MGI:97742</a>
See related	<a href="#">Ensembl:ENSMUSG00000020660</a>
Gene type	protein coding
RefSeq status	REVIEWED
Organism	<a href="#">Mus musculus</a>
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	BE; Npp; ACTH; Clip; Pomc1; Pomc-1; Beta-LPH; alphaMSH; beta-MSH; Gamma-LPH; alpha-MSH; gamma-MSH
Summary	This gene encodes a polypeptide hormone precursor that undergoes extensive, tissue-specific, post-translational processing. Processing yields several biologically active peptides, which are involved in diverse cellular functions, such as energy homeostasis, steroidogenesis, and increased melanin production in melanocytes. In mouse deficiency of this gene is associated with obesity, defects in adrenal development, and altered pigmentation. A pseudogene of this gene is located on chromosome 19. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Jun 2013]
Expression	Biased expression in testis adult (RPKM 4.3), CNS E14 (RPKM 2.4) and 11 other tissues <a href="#">See more</a>
Orthologs	<a href="#">human</a> <a href="#">all</a>

# Transcript information ( Ensembl )

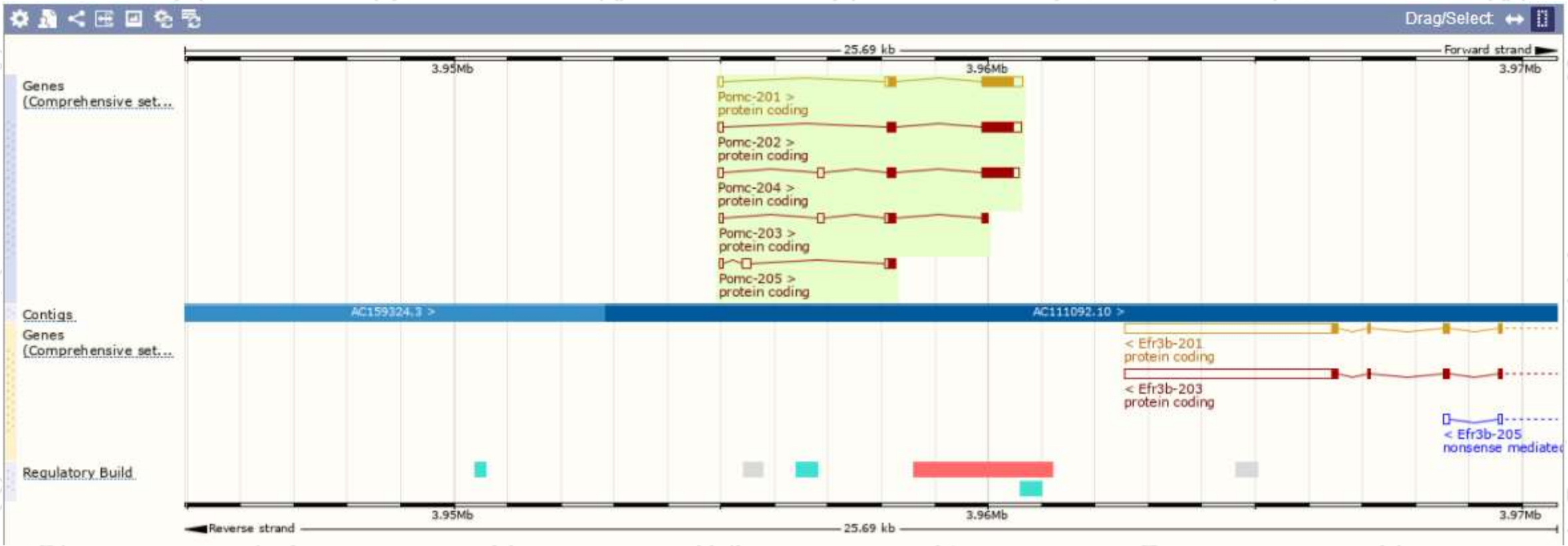
The gene has 5 transcripts, and all transcripts are shown below :

Show/hide columns (1 hidden)								Filter	
Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags	
Pomc-204	<a href="#">ENSMUST00000219543.1</a>	1043	<a href="#">235aa</a>	Protein coding	<a href="#">CCDS25785</a>	<a href="#">P01193</a>	<a href="#">NM_001278582</a> <a href="#">NM_001278583</a> <a href="#">NP_001265511</a> <a href="#">NP_001265512</a>	TSL:1	GENCODE basic APPRIS P1
Pomc-201	<a href="#">ENSMUST00000020990.6</a>	1031	<a href="#">235aa</a>	Protein coding	<a href="#">CCDS25785</a>	<a href="#">P01193</a>	<a href="#">NM_001278581</a> <a href="#">NM_008895</a> <a href="#">NP_001265510</a> <a href="#">NP_032921</a>	TSL:1	GENCODE basic APPRIS P1
Pomc-202	<a href="#">ENSMUST00000218089.1</a>	977	<a href="#">235aa</a>	Protein coding	<a href="#">CCDS25785</a>	<a href="#">P01193</a>	<a href="#">NM_001278584</a> <a href="#">NP_001265513</a>	TSL:1	GENCODE basic APPRIS P1
Pomc-203	<a href="#">ENSMUST00000218169.1</a>	480	<a href="#">79aa</a>	Protein coding	-	<a href="#">A0A1W2P7R2</a>	-	CDS 3' incomplete	TSL:2
Pomc-205	<a href="#">ENSMUST00000220006.1</a>	438	<a href="#">44aa</a>	Protein coding	-	<a href="#">A0A1W2P724</a>	-	CDS 3' incomplete	TSL:3

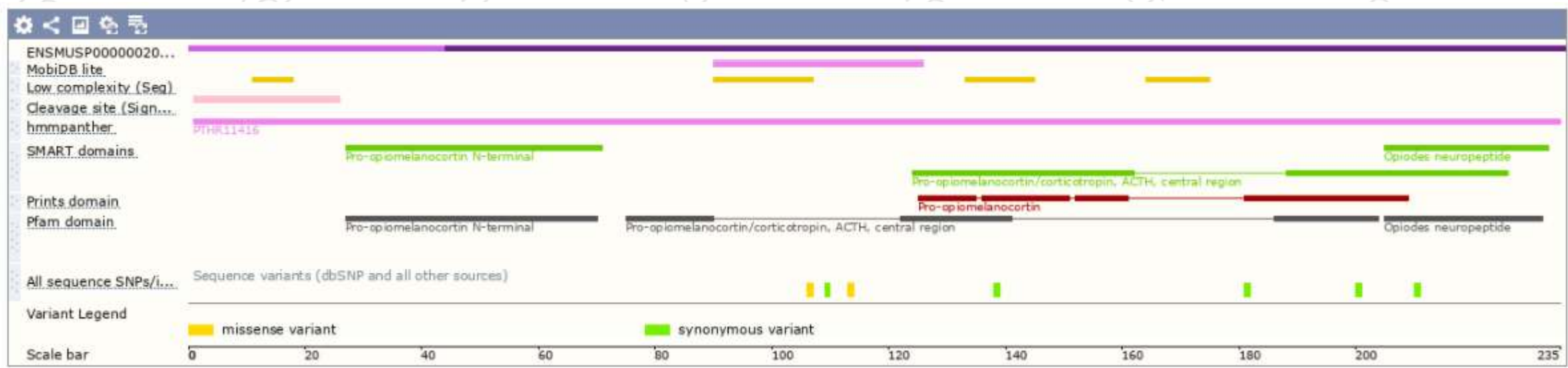
The strategy is based on the design of *Pomc-201* transcript,The transcription is shown below :



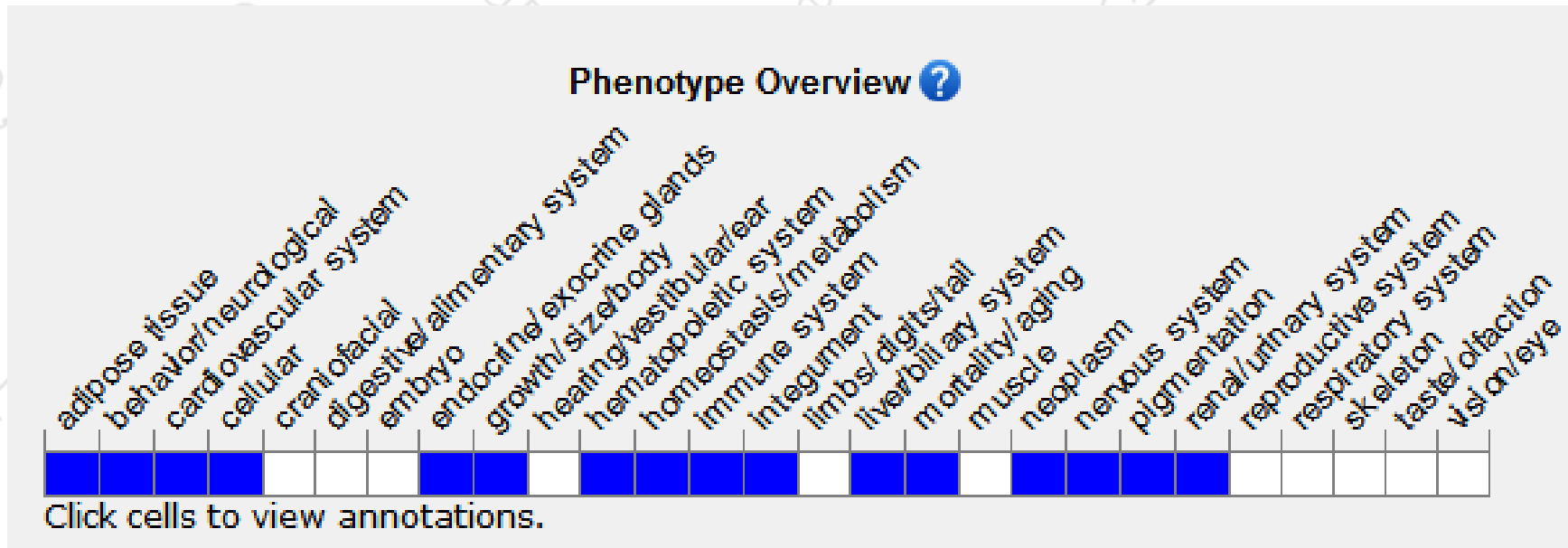
# Genomic location distribution



# 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, Homozygotes for a targeted null mutation are obese and exhibit abnormal hormone levels, abnormal pigmentation, increased food intake, and adiposity. Mice homozygous for another knock-out allele exhibit altered reward based behavior and immune response to LPS treatment.

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