Bdnf Cas9-KO *Strategy*

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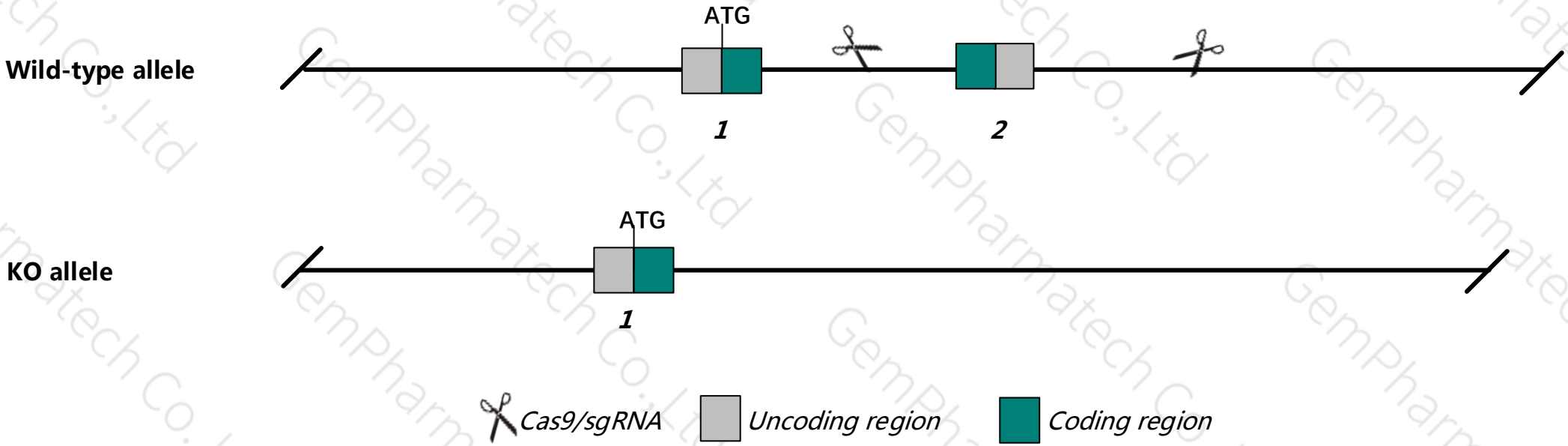
Project Overview

Project Name	*Bdnf*
Project type	Cas9-KO
Animal background	C57BL/6JGpt

Knockout strategy

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



Technical routes

- The *Bdnf* gene has 11 transcripts, According to the structure of *Bdnf* gene, exon2 of *Bdnf-201* transcript is recommended as the knockout region.The region contains the most of coding seqence.Knock out the region,result in destruction of protein.

- This project uses CRISPR/Cas9 technology to modify *Bdnf* 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.

Notice

- According to the existing MGI data，Homozygotes for targeted null alleles exhibit sensory neuron losses affecting coordination, balance, hearing, taste, and breathing, cerebellar abnormalities, increased sympathetic neuron number, and postnatal lethality. Carriers show mild defects.

- The *Bdnf* gene is located in the Chr2. 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）

集萃药康 GemPharmatech

Bdnf brain derived neurotrophic factor [*Mus musculus* (house mouse)]

Gene ID: 12064, updated on 10-Feb-2019

Summary

Official Symbol	Bdnf provided by MGI
Official Full Name	brain derived neurotrophic factor provided by MGI
Primary source	MGI:MGI:88145
See related	Ensembl:ENSMUSG00000048482
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Summary	The protein encoded by this gene is a member of the nerve growth factor family. It is involved in the growth, differentiation and survival of specific types of developing neurons both in the central nervous system (CNS) and the peripheral nervous system. It is also involved in regulating synaptic plasticity in the CNS. Expression of a similar gene in human is reduced in both Alzheimer's and Huntington disease patients. Alternative splicing results in multiple transcript variants encoding different isoforms that may undergo similar processing to generate mature protein. [provided by RefSeq, Oct 2015]
Expression	Biased expression in cortex adult (RPKM 1.7), frontal lobe adult (RPKM 1.4) and 12 other tissues See more
Orthologs	human all

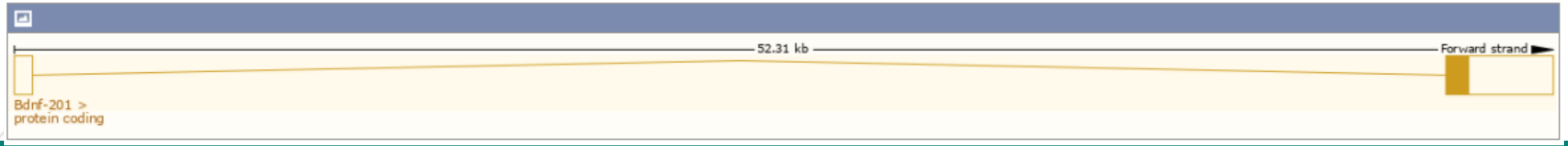
Transcript information （Ensembl）

The gene has 11 transcripts, and all transcripts are shown below ：

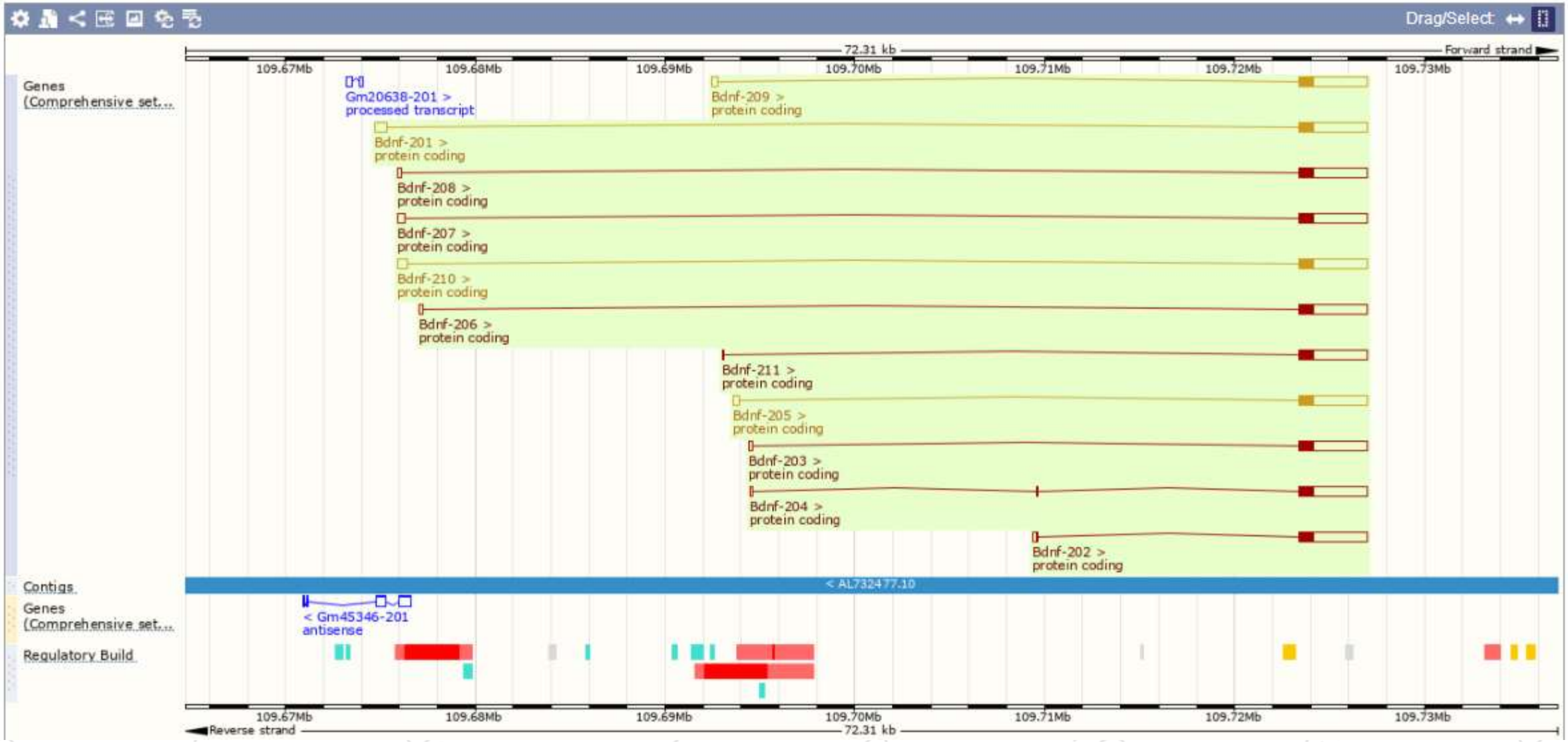
Show/hide columns (1 hidden) Filter

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	RefSeq	Flags
Bdnf-201	ENSMUST00000053317.11	4266	257aa	Protein coding	CCDS38193	A2AII2	NM_007540 NP_031566	TSL:1 GENCODE basic APPRIS P4
Bdnf-210	ENSMUST00000111051.9	4125	249aa	Protein coding	CCDS38194	P21237 Q541P3	NM_001048139 NP_001041604	TSL:1 GENCODE basic APPRIS ALT1
Bdnf-207	ENSMUST00000111047.8	4042	249aa	Protein coding	CCDS38194	P21237 Q541P3	NM_001285416 NP_001272345	TSL:1 GENCODE basic APPRIS ALT1
Bdnf-205	ENSMUST00000111045.8	3973	249aa	Protein coding	CCDS38194	P21237 Q541P3	NM_001048142 NP_001041607	TSL:1 GENCODE basic APPRIS ALT1
Bdnf-209	ENSMUST00000111050.9	3965	249aa	Protein coding	CCDS38194	P21237 Q541P3	NM_001048141 NP_001041606	TSL:1 GENCODE basic APPRIS ALT1
Bdnf-204	ENSMUST00000111044.2	3854	249aa	Protein coding	CCDS38194	P21237 Q541P3	NM_001285421 NM_001316310 NP_001272350 NP_001303239	TSL:1 GENCODE basic APPRIS ALT1
Bdnf-203	ENSMUST00000111043.8	3839	249aa	Protein coding	CCDS38194	P21237 Q541P3	NM_001285417 NP_001272346	TSL:1 GENCODE basic APPRIS ALT1
Bdnf-208	ENSMUST00000111049.8	3830	249aa	Protein coding	CCDS38194	P21237 Q541P3	NM_001285418 NP_001272347	TSL:1 GENCODE basic APPRIS ALT1
Bdnf-206	ENSMUST00000111046.8	3819	249aa	Protein coding	CCDS38194	P21237 Q541P3	NM_001285419 NP_001272348	TSL:1 GENCODE basic APPRIS ALT1
Bdnf-211	ENSMUST00000176893.7	3707	249aa	Protein coding	CCDS38194	P21237 Q541P3	NM_001285420 NM_001285422 NP_001272349 NP_001272351	TSL:1 GENCODE basic APPRIS ALT1
Bdnf-202	ENSMUST00000111042.2	3905	289aa	Protein coding	-	H9H9S8	-	TSL:1 GENCODE basic

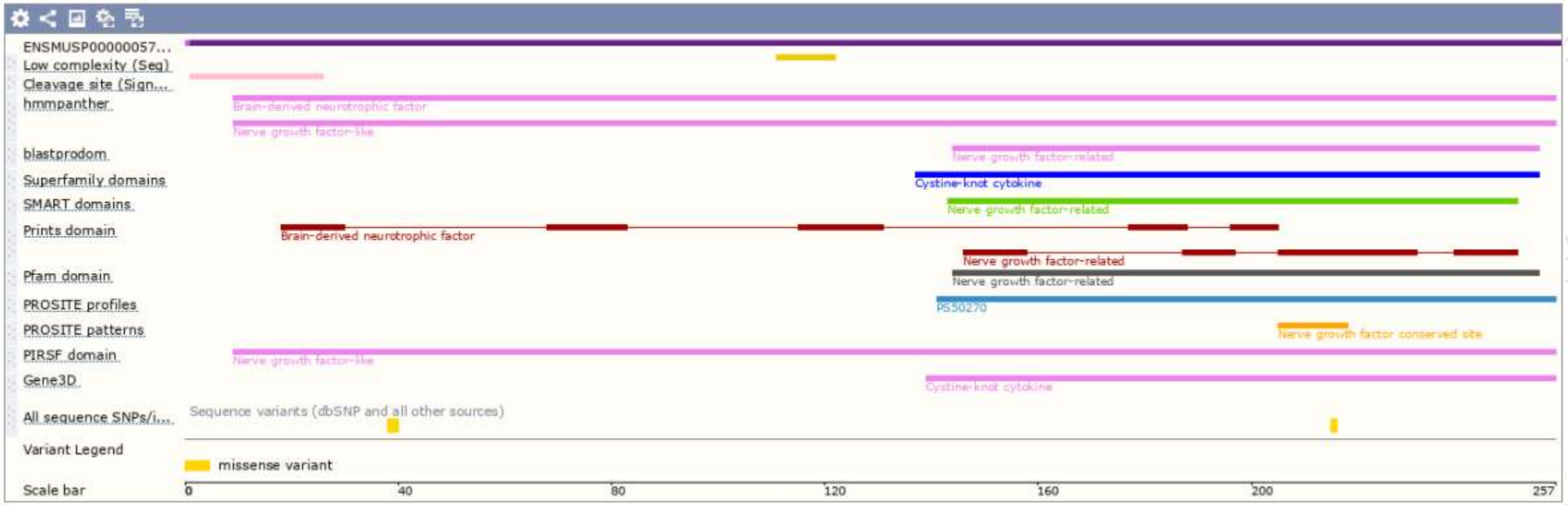
The strategy is based on the design of *Bdnf-201* transcript,The transcription is shown below：



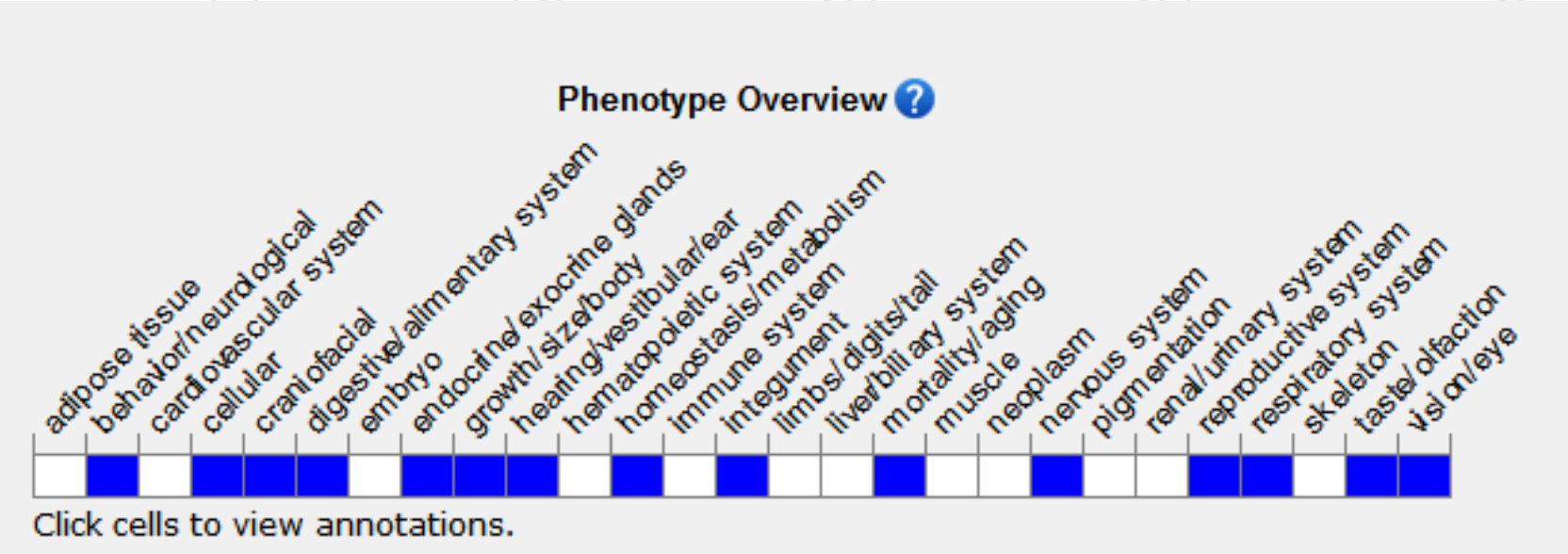
Genomic location distribution



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 targeted null alleles exhibit sensory neuron losses affecting coordination, balance, hearing, taste, and breathing, cerebellar abnormalities, increased sympathetic neuron number, and postnatal lethality. Carriers show mild defects.Homozygotes for targeted null alleles exhibit sensory neuron losses affecting coordination, balance, hearing, taste, and breathing, cerebellar abnormalities, increased sympathetic neuron number, and postnatal lethality. Carriers show mild defects.

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

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