GemPharmatech™

Hus1 Cas9-KO Strategy

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Reviewer: Jia Yu

Design Date: 2023-10-18

Overview

Target Gene Name

- *Hus1*

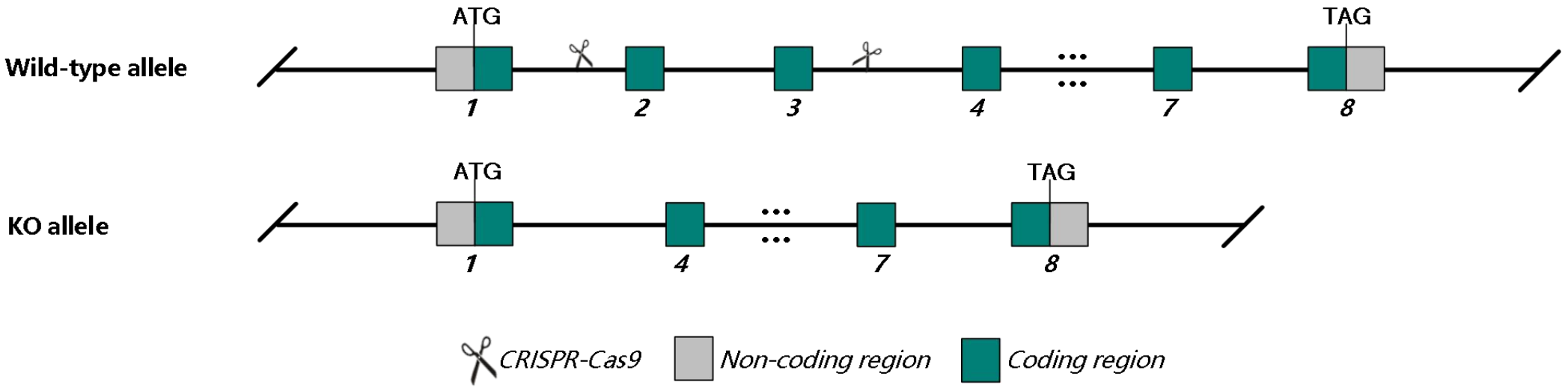
Project Type

- Cas9-KO

Genetic Background

- C57BL/6JGpt

Strain Strategy



Schematic representation of CRISPR-Cas9 engineering used to edit the *Hus1* gene.

Technical Information

- The *Hus1* gene has 6 transcripts. According to the structure of *Hus1* gene, exon 2-3 of *Hus1*-201 (ENSMUST00000020683.10) is recommended as the knockout region. The region contains 305 bp of coding sequence. Knocking out the region will result in disruption of gene function.
- In this project we use CRISPR-Cas9 technology to modify *Hus1* gene. The brief process is as follows: gRNAs were transcribed in vitro. Cas9 and gRNAs 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 on-target amplicon sequencing. A stable F1-generation mouse strain was obtained by mating positive F0-generation mice with C57BL/6JGpt mice and confirmation of the desired mutant allele was carried out by PCR and on-target amplicon sequencing.

Gene Information

Hus1 HUS1 checkpoint clamp component [*Mus musculus* (house mouse)]

Download Datasets

Gene ID: 15574, updated on 15-Oct-2023

Summary

Official Symbol	Hus1 provided by MGI
Official Full Name	HUS1 checkpoint clamp component provided by MGI
Primary source	MGI:MGI:1277962
See related	Ensembl:ENSMUSG00000020413 AllianceGenome:MGI:1277962
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	This gene encodes a component of a cell cycle checkpoint complex that causes cell cycle arrest in response to bulky DNA lesions and DNA replication blockage. Together with the proteins Rad9 and Rad1, the encoded protein forms a heterotrimeric complex known as the 9-1-1 complex. Mice lacking the encoded protein develop spontaneous chromosomal abnormalities resulting in embryonic lethality. Alternative splicing of this gene results in multiple transcript variants. [provided by RefSeq, Jan 2015]
Expression	Ubiquitous expression in CNS E11.5 (RPKM 4.3), limb E14.5 (RPKM 3.6) and 28 other tissues See more
Orthologs	human all

NEW **Try the new Gene table**
Try the new Transcript table

Genomic context

Location: 11 A1; 11 5.74 cM — See Hus1 in Genome Data Viewer

Exon count: 9

https://www.ncbi.nlm.nih.gov/gene/15574

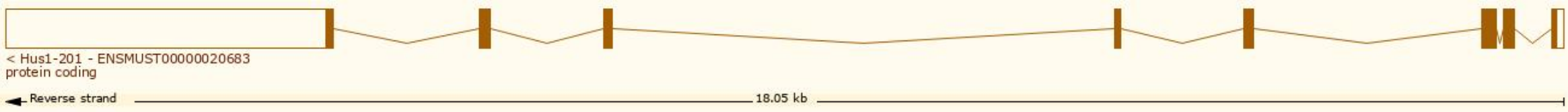
Transcript Information

The gene has 6 transcripts, all transcripts are shown below:

Show/hide columns (1 hidden) Filter

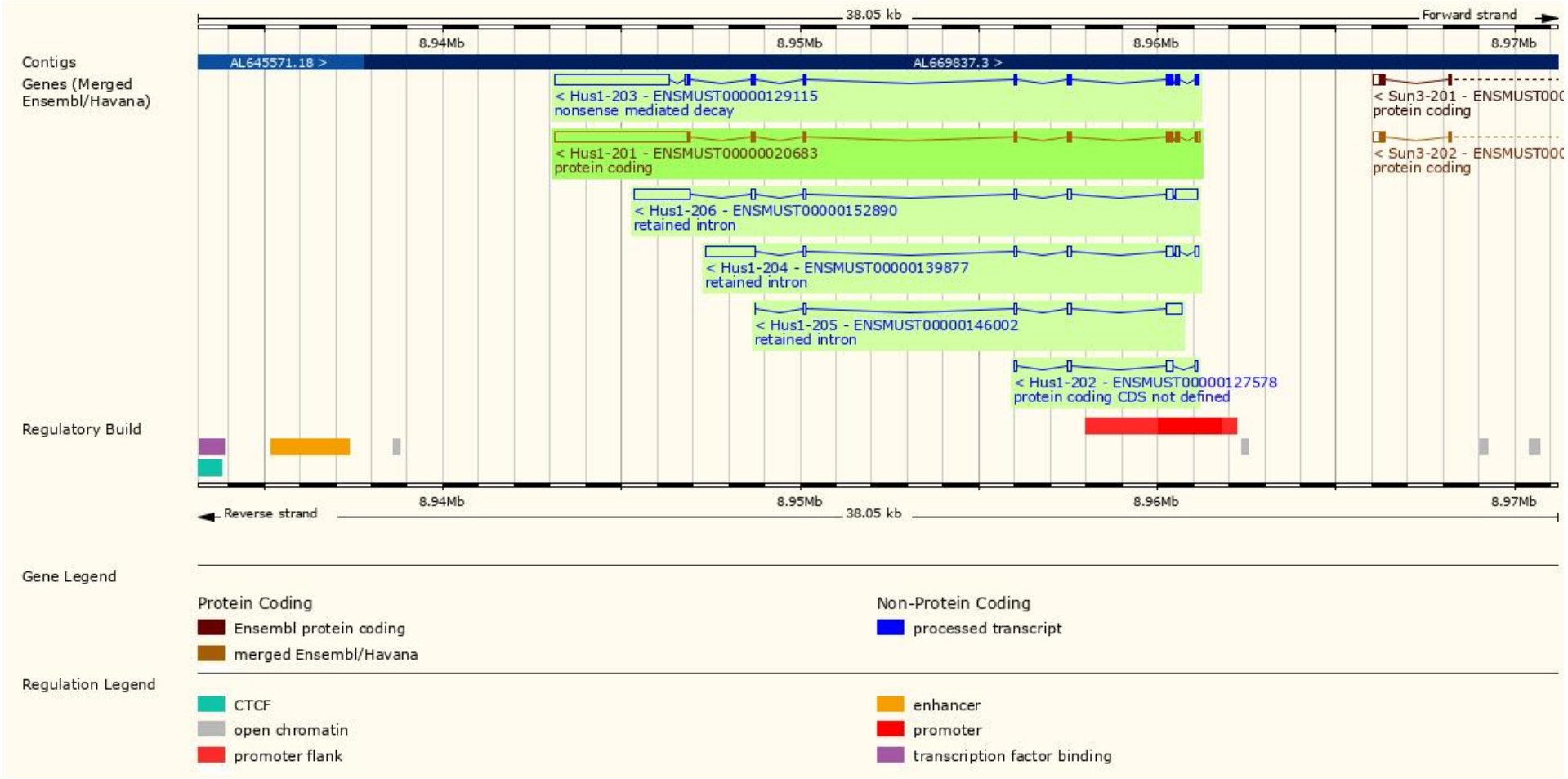
Transcript ID	Name	bp	Protein	Biotype	CCDS	UniProt Match	Flags
ENSMUST00000020683.10	Hus1-201	4640	281aa	Protein coding	CCDS24429	Q8BQY8-2	Ensembl Canonical, GENCODE basic, APPRIS P1, TSL:1
ENSMUST00000129115.2	Hus1-203	4172	281aa	Nonsense mediated decay	CCDS24429	Q8BQY8-2	TSL:1
ENSMUST00000127578.2	Hus1-202	433	No protein	Protein coding CDS not defined		-	TSL:5
ENSMUST00000152890.8	Hus1-206	2792	No protein	Retained intron		-	TSL:5
ENSMUST00000139877.8	Hus1-204	2114	No protein	Retained intron		-	TSL:2
ENSMUST00000146002.8	Hus1-205	742	No protein	Retained intron		-	TSL:5

The strategy is based on the design of *Hus1*-201 transcript, the transcription is shown below:



Source: http://asia.ensembl.org/

Genomic Information



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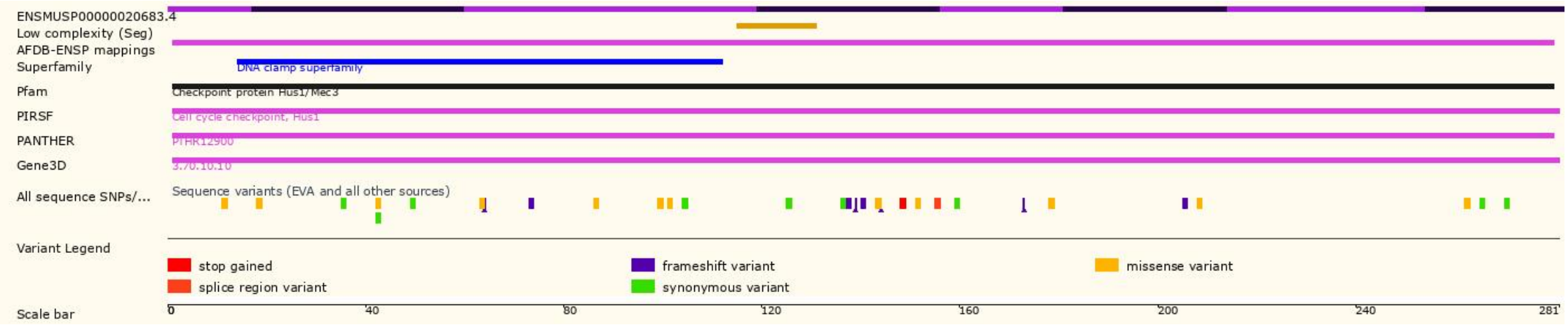
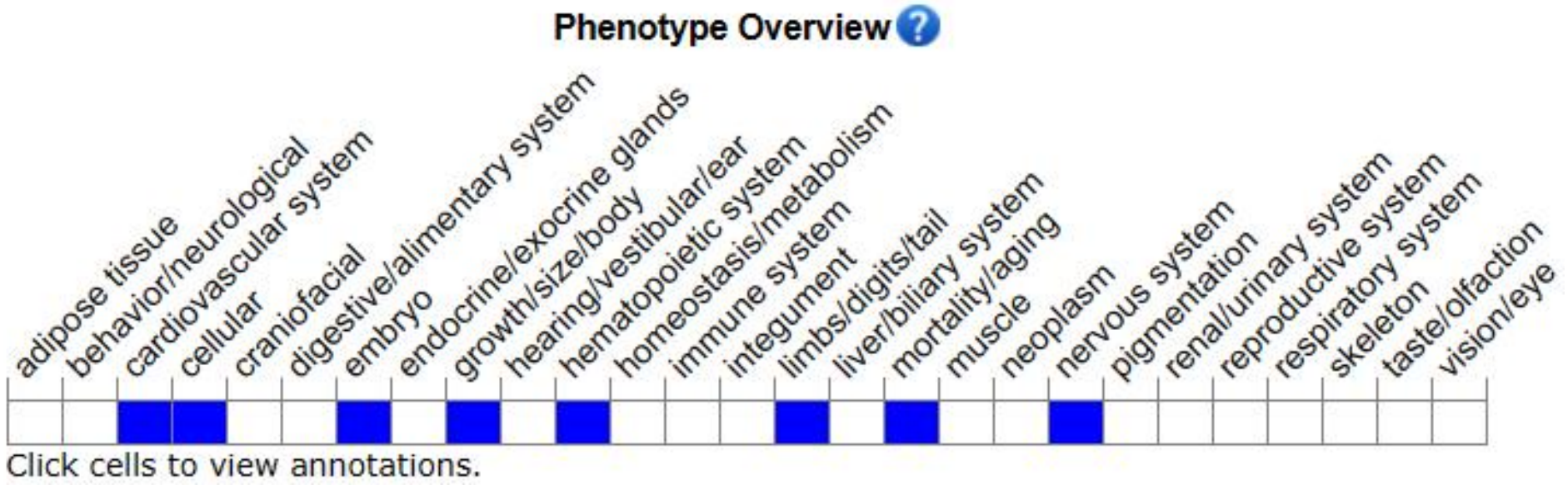
Source: http://asia.ensembl.org/

Protein Information



Source: https://www.ensembl.org

Mouse Phenotype Information (MGI)



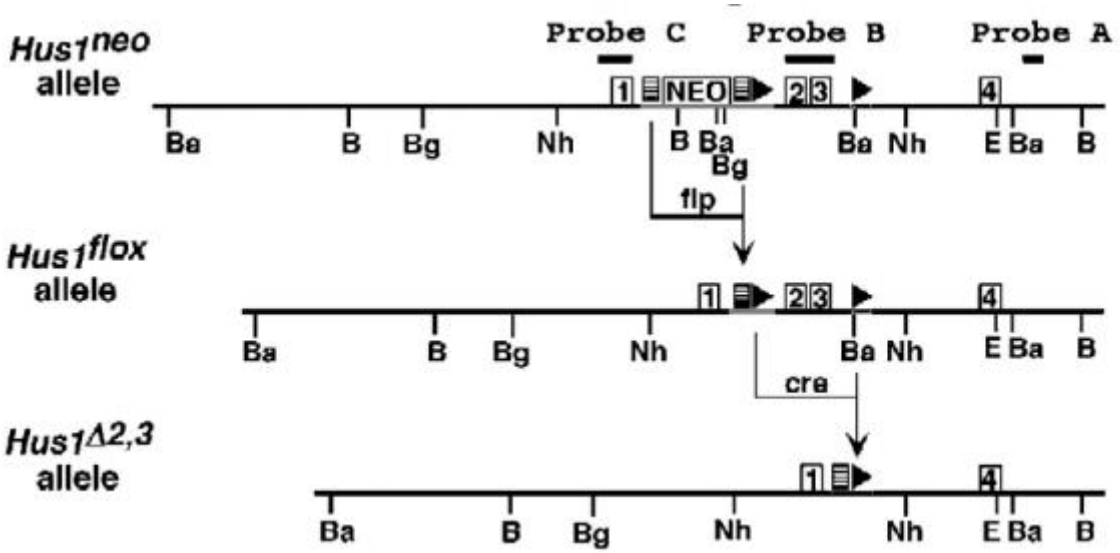
Homozygotes for a targeted null mutation exhibit defects in yolk sac vascularization, placental abnormalities, extensive apoptosis, and midgestational lethality. Mutant cells show increased chromosomal abnormalities.

Source: https://www.informatics.jax.org

Important Information

- *Hus1* is located on Chr 11. If the knockout mice are crossed with other mouse strains to obtain double homozygous mutant offspring, please avoid the situation that the second gene is on the same chromosome.
- This strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risks of the mutation on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Reference



Swiss Webster foster mothers. Chimeric male mice were mated to 129S6 females and the targeted allele was subsequently maintained on a 129S6 inbred background. Removal of the *neo* cassette was accomplished by crossing $Hus1^{+/neo}$ mice with Flp-expressing $R26^{+/Fki}$ mice [35]. *Hus1* exons two and three were deleted by crossing $Hus1^{+/flox}$ or $Hus1^{flox/flox}$ mice with either $EIIa\text{-}cre^{+/-}$ [36] or $Hs\text{-}cre6^{+/-}$ mice [38].

[1] Levitt PS, Liu H, Manning C, Weiss RS. Conditional inactivation of the mouse Hus1 cell cycle checkpoint gene. Genomics. 2005 Aug;86(2):212-24. doi: 10.1016/j.ygeno.2005.04.007. PMID: 15919177.