

**My Account**

Login  
Create Account

**Resources**

View All (813)  
Adenoviruses (137)  
Antibodies (175)  
Bioimages (67)  
Genomics Studies (145)  
mESC Lines (68)  
Mouse Strains (120)  
Miscellaneous (46)  
Protocols (55)  
Research Data (4)  
Resource Tags (389)  
Visualization (9)

**Research & Cores**

Core Facilities (5)  
Research Highlights (5)  
Research Networks  
Research Objectives

**Information**

About the BCBC  
BCBC Events  
Branding & Logos  
Career Opportunities  
Health  
NIH hESC Registry  
Policies & Guidelines  
Member Publications  
Research Programs  
Research Investigators  
Member Directory  
Tutorials

**Ngn3-tTA - Mouse Strain RES239****Mouse Information**

<b>Common Name:</b>	Ngn3-tTA
<b>MGI Official Name:</b>	Ngn3 <sup>tm1(tTA)Hri</sup>
<b>Description:</b>	The mouse contains a Ngn3 knock-in allele. IRES-tTA-rabbit beta globin polyA signal replaced the entire Ngn3 cDNA coding sequence. This is therefore also a Ngn3 null allele. Ngn3 is a bHLH transcription factor that is required for the differentiation of endocrine islet and some neuronal cells.
<b>Categories:</b>	Tet

**Genetic Alterations****1) Targeted Mutagenesis**


<b>Type of Allele</b>	Gene Replacement
<b>Targeted Gene</b>	Neurogenin 3 (Neurog3 - <a href="#">NCBI GeneID:11925</a> )
<b>Targeted Allele</b>	Ngn3 tTA knockin (Ngn3tTA)

**Description of Targeting Vector**


DNA cloning follows standard molecular biology techniques. IRES-tTA was cloned using a two-step PCR. First, two PCR reactions were utilized to amplify IRES (using oligo1: 5'-ccgcttagttaaactcgagttaatta-3' and oligo2: 5'-atcaatctagacatggttgaggcaag-3') and tTA cDNA (using oligo3: 5'-acgcgtctaccaccgactctgctca-3' and oligo4: 5'-ctggcacaaccatgtctgattagat-3'). Templates used for PCR were pGpux1 (gift from G. Mellizer) and Pdx1-tTA (gift from R. MacDonald), respectively. The oligos used for PCR were designed so that a 15 base pairs overlapping between the two fragments was introduced. Second, the above PCR products were mixed 1:1 and used as template to generate IRES-tTA using oligo1 + oligo4. In parallel Ngn3-SpeI-eGFP was digested with SpeI and ligated with the 3.3Kb fragment from a pSL1180 vector digested with SpeI/XbaI to generate pSL1180-Ngn3-SpeI-eGFP vector (pSL1180 and pNgn3-SpeI-eGFP was gifts from G. Mellizer.). The IRES-tTA fragment was then inserted into pGpux1 using XhoI + MluI digestion to generate pGpux1-IRES-tTA. Finally Ascl + XhoI fragment from pGpux1-IRES-tTA (4.6kb) was ligated into Ascl + XhoI digested pSL1180-Ngn3-SpeI-eGFP vector, to obtain the targeting vector pSL1180Ngn3-IRES-tTA. The vector was linearized with Alol before electroporation. ES cell electroporation follow standard procedures. Southern blot was utilized to screen for targeted clones. Primary screen identified 9 positive clones from 384 clones screened. Six clones were expanded and recombination was verified with Southern blot probes located in both 3' Ngn3-arm and 5' Ngn3-arm (probes were PCR generated using the primers: 3' probe: 5'-cctggaagtgccaggagc-3' + 5'-cagtaccaccactactctc-3' and 5' probe: 5'-ccctctctcccttggc-3' + 5'-acacatggattggcactga-3'). Blastocyst injection and germline transmission tested following standard techniques.

<b>Targeting Vector Genbank File</b>	<a href="#">pngn3.IRES.tTA.gb</a>
<b>Citations</b>	Not Available

**Access Status**

 This resource is publicly viewable.

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Primary contributor: [Palle Serup](#)  
Co-contributed by:  
• [BCBC Mouse / ES Cell Core](#)

**Resource Tags**

mESC Core, mouse, mouse strain, Ngn3, Ngn3<sup>tm1(tTA)Hri</sup>, Ngn3-tTA, tTA

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**Resource History & Actions**

Approved on Jul 15, 2008  
Last modified on Feb 08, 2012

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**Related resources****BCBC**

No matching resources

**Other Consortia**

No matching resources

Data courtesy of [dkCOIN](#). Only public resources are displayed.

**Strain Information**

Strain Type:	Mixed
Chimera/Founder Genetic Background:	129
Current Genetic Background:	129 and ICR (CD1)(date recorded: 03/27/2015)
Strain Description:	The original knockin is in 129 (TL1 ES cells) background. It has been crossed with ICR mice over 4 generations. It is therefore at mostly ICR background.

**Associated Images**

Image 1	<p><b>Description:</b></p> <p><b>Ngn3-tTA derivation and tTA expression pattern detection.</b> (A) The <i>Ngn3</i> wild type and the knock-in allele (before removal of PuroR selection cassette). Note that a rabbit <math>\beta</math> globin polyadenylation signal is included at the 3' end of tTA. (B) Verification of the correct targeting of the <i>Ngn3</i> locus in six ES cell clones by genomic southern blot and PCR. The Southern blot pattern and PCR fragments are as expected from correctly targeted alleles (see Materials and Methods). P5 and P3 are templates used to make probes. (A) Whole mount LacZ staining at two embryonic stages and in seven-day-old postnatal pancreas. Left side panels are TetOLacZ single transgenic samples. Right side panels are pNgn3tTA; TetOlacZ littermates. The pancreatic region was circled with dotted lines. (B) LacZ expression in issue sections of corresponding ages to (A). Insets are boxed regions shown at higher magnification.</p> <p><b>Reference:</b> 19487660</p>
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**Repositories**

Serup Lab	<p>No URL supplied for repository</p> <p>Stock #: VUMC-KG Availability Notes: <i>Not provided</i></p>
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Email	<i>Not provided</i>

**Associated Publications**

*No publications associated*

**Comments**

*There are no comments for this entry.*

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