B6.Cg-<sup>trnO.1(tetO-COP4*EGFP)Hze</sup>-J

Stock No: 024100 | Ai90(TITL-Chronos)-D or Ai90D

- Congenic, Targeted Mutation

**CRYO RECOVERY**

**PLACE ORDER**

Typically mice are recovered in 10-14 weeks. Contact Customer Service to place an order or for more information.
Overview

Also Known As: Ai90(TITL-Chronos)-D or Ai90D

Ai90(TITL-Chronos)-D (also called Ai90D) mice are a Cre/Tet-dependent, optogenetic line, created by targeted insertion at the *Igs7* locus (TIGRE; an intergenic region on mouse chromosome 9 that allows reporter expression to be tightly regulated). Following Cre-mediated removal of the STOP cassette, they may be used to generate Tet-Off/Tet-On mutant animals with conditional (inducible/reversible) expression of the channelrhodopsin/EGFP fusion protein (Chronos/EGFP). Subsequent illumination of Chronos-expressing (EGFP fluorescent) cells with blue-to-green light leads to reversible photostimulation of action potential firing/neural activity in these cells.

Donating Investigator

Hongkui Zeng, Allen Institute for Brain Science
Detailed Description

Ai90(TITL-Chronos)-D mice are a Cre/Tet-dependent, optogenetic line, created by targeted insertion at the Igs7 locus (TIGRE; an intergenic region on mouse chromosome 9 that allows reporter expression to be tightly regulated). Ai90D mice harbor the TIGRE-Ins-TRE-LSL-Chronos conditional allele, designed with a modified Tet response element (TRE or tetO) and loxP-flanked STOP cassette upstream of an improved Chronos/EGFP fusion protein (see detailed description below). When bred with other mice expressing Cre recombinase, tetracycline-controlled transactivator protein (tTA) and/or reverse tetracycline-controlled transactivator protein (rtTA), Chronos expression in cells/tissues where the expression patterns of the individual promoters driving Cre and tTA/rtTA overlap can be regulated with tetracycline or its analog doxycycline (dox). Following induction of Chronos expression (EGFP fluorescence), illuminating neurons with blue-to-green light leads to reversible photostimulation of action potential firing/neural activity in these cells.

Specifically, the donating investigator reports Ai90D mice have no reported levels of EGFP fluorescence in absence of Cre and tTA. When Ai90D mice are bred with tTA-driver and Cre-driver lines to create triple transgenic animals (Chronos+/tTA+/Cre+), cells expressing both tTA and Cre exhibit robust fluorescence. Light-induced expression of the activation opsin is expected to occur at levels sufficient to effectively depolarize/activate cortical neurons. Chronos expression/function in tissues other than brain has not yet been evaluated by the donating investigator (March 2014).

Heterozygous Ai90D mice are viable and fertile with no reported gross physical or behavioral abnormalities. The donating investigator has not attempted to generate homozygous Ai90D mice to date (March 2014).

The bacterial opsins are retinal-binding proteins that combine a light-sensitive domain with an ion channel or pump; providing light-dependent ion transport, membrane potential alteration, and sensory functions to bacteria. The Stigeoclonium helveticum-derived channelrhodopsin ShChR (Chronos) is a blue-to-green light-driven cation channel that depolarizes the cell and causes action potentials. As such, illuminating Chronos-expressing cells with blue-to-green light (~470-530 nm) leads to rapid and reversible photostimulation of robust action potential firing activity in these cells. Chronos has high light sensitivity and faster kinetics than previous channelrhodopsins. The action spectrum of Chronos is shifted to longer wavelengths compared to ChR2. In addition, Chronos is especially well suited to be paired with a red-shifted channelrhodopsin in order to allow for two light color activation of independent populations of neurons. The Chronos/EGFP fusion gene is composed of a mammalian codon-optimized Chronos fused in-frame to the N-terminus of EGFP.

Development

Expression Data

Control Suggestions

Selected References

Genetics

Igs7tm90.1(tetO-COP4*EGFP)Hze

Disease/Phenotype
Genotyping Protocols
Standard PCR: Igs7
Genotyping resources and troubleshooting

Breeding Considerations
When maintaining a live colony, heterozygous mice may be bred to wildtype mice from the colony or to C57BL/6J inbred mice (Stock No. 000664). The donating investigator has not attempted to generate homozygous mice to date (March 2014).

Additional Breeding and Husbandry Support

Citation
When using the Ai90(TITL-Chronos)-D or Ai90D mouse strain in a publication, please cite the originating article(s) and include JAX stock #024100 in your Materials and Methods section.

Animal Health Reports
Facility Barrier Level Descriptions
Production of mice from cryopreserved embryos or sperm occurs in a maximum barrier room, G200

Pricing & Availability

Typically mice are recovered in 10-14 weeks. Contact Customer Service to place an order or for more information.

Cryo Recovery

<table>
<thead>
<tr>
<th>SERVICE/PRODUCT</th>
<th>DESCRIPTION</th>
<th>PRICE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cryo Recovery</td>
<td>Heterozygous or Wildtype forIgs7&lt;tm90.1(tetO-COP4*/EGFP)Hze&gt;</td>
<td>$2,854.50</td>
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Related Products and Services

Frozen Mouse Embryo

<table>
<thead>
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<th>SERVICE/PRODUCT</th>
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<tr>
<td>Frozen Mouse Embryo</td>
<td>B6.Cg-Igs7&lt;tm90.1(tetO-COP4*/EGFP)Hze&gt;/J</td>
<td>$2,595.00</td>
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</tbody>
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