

Tamoxifen诱导Cre-ERT2小鼠 使用指南

Cre-ERT2在无Tamoxifen诱导的情况下，在细胞质内处于无活性状态；当Tamoxifen诱导后，Tamoxifen的代谢产物4-OHT（雌激素类似物）与ERT结合，可使Cre-ERT2进核发挥Cre重组酶活性。

一、品系介绍：

Cre-ERT2小鼠是一类含有雌激素受体（estrogen receptor, ER）的配体结合区突变体（ERT）与Cre重组酶的融合蛋白表达的小鼠。Cre-ERT2在无Tamoxifen诱导的情况下，在细胞质内处于无活性状态；当Tamoxifen诱导后，Tamoxifen的代谢产物4-OHT（雌激素类似物）与ERT结合，可使Cre-ERT2进核发挥Cre重组酶活性。

利用不同的启动子，可以在不同组织或细胞中特异性调控表达Cre。如肝特异性表达Cre-ERT2：B6.129-Alb^{tm1(CreERT2)Srcmo}或B6.Cg-Tg(Afp-CreERT)Srcmo。

二、常规Tamoxifen诱导方法：

Tamoxifen用玉米油溶解。因Tamoxifen的光敏特性，操作和保存时应保持避光。

Tamoxifen (Sigma-Aldrich): CAS# 10540-29-1

玉米油 (Sigma-Aldrich) CAS# 8001-30-7

注意：不同品系的小鼠所用的Tamoxifen诱导方法可能不同，应根据实际使用效果、实验经验和设置加以调整和更改。此处提供的是初步的常规诱导方法，仅供参考。

1、腹腔注射：（以下方法来自Jax lab）

Tamoxifen (Sigma-Aldrich): CAS# 10540-29-1

Corn Oil (Sigma-Aldrich)

70% Ethanol (for disinfection)

1ml Syringe (BD)

3/8" beveled needle - 26 gauge (BD)

Procedure:

Dissolve tamoxifen in corn oil at a concentration of 20mg/ml by shaking overnight at 37°C. After tamoxifen is in solution, store at 4°C for the duration of injections.

Determine injection dose by weight, using approximately 75mg/kg body weight. For adult mice, a standard dose of 100ul tamoxifen/corn oil solution is effective for inducing recombination.

Administer tamoxifen via intraperitoneal injection (using an ACUC approved injection procedure) once every 24 hours for a total of 5 consecutive days. As a safeguard, sanitize injection site with 70% ethanol prior to injection.

Following the final injection, mice should be quarantined for 24 hours before returning to their normal animal room. Disposable pens and bedding should be discarded at this time. For Cre characterization work at the Jackson lab, there is a 7-day waiting period between the final injection and necropsy/histological analysis.

Throughout the course of tamoxifen injections and any post-injection wait period, mice should be closely monitored for any adverse reactions to the treatment.

References:

Madisen, J.L., et al., Nat Neurosci. 2010; 13(1): 133-140

Sohal, D.D., et al., Circ Res. 2001; 89: 20-25

2、灌胃给药:

(1) Tamoxifen was fed by gavage to mice at doses of 0.13-0.16mg/g.

Corn oil was used to dissolve tamoxifen at 37°C or room temperature (shaking) overnight and get the stock tamoxifen whose concentration is 20mg/ml.

Reference:

Feil R., et al., Proc Natl Acad Sci USA 1996; 93:10887-90.

Feil R., et al., Biochem Biophys Res Commun. 1997; 237:752-7.

Metzger D., et al., Proc Natl Acad Sci USA 1995; 92:6991-5.

Zhang Y., et al., Nucleic Acids Res. 1996; 24:543-8.

(2) 100ul/10g体重 (5mg/只) , 连续灌胃给药5天。等待至少一周后, 检测。

药品: TAMO: 20mg/ml Tamoxifen in 10% 乙醇/玉米油 (50度水浴助溶)

Oil: 10% 乙醇/玉米油

分装, 避光, - 20度冻存

备注:

1. Do not house the tamoxifen-treated mice together with vehicle-treated or untreated animals in the same cage.
2. 给药期间小鼠体重减轻, 停药后会恢复。
3. 观察到雄性小鼠给药后, 外观睾丸增大。
4. 小鼠体内各组织诱导效率有所差异, 通常上调约2.5-3倍