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DIO-SPOTlight in vivo ISR Activation with Tunicamycin v.250227

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Protocol status: Working

We use this protocol and it's working

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Abstract

Abstract

This protocol contains the steps and dosages used for the acute in vivo integrated stress response activation experiment using tunicamycin treatment in young mice (P12-14). (Wang et al., 2015) indicates may not be as effective in older mice.

In vivo tunicamycin treatment

- 1 Prepare tunicamycin (Tocris) stock solution in dimethyl sulfoxide (DMSO) to a concentration of 5 mg/ml. Prepare fresh, when possible and discard after 1 month.
 - a. On the day of treatment, dilute tunicamycin 1:25 (v/v) in 150 mM dextrose in PBS with 5% Tween-80
 - b. Prepare vehicle treatment by 1:25 (v/v) dilution of DMSO in 150 mM dextrose in PBS with 5% Tween-80
 - i. Sterile filter the tunicamycin and vehicle solutions through separate 0.22 μ m syringe filters
- 2 Starting around 3:00 PM, weigh P12 pups and record weights
- 3 Prepare sterile syringes to administer tunicamycin or vehicle solutions at a final concentration of 3 μ g/g bodyweight.
- 4 Scruff mouse pup and clean the injection site with an alcohol wipe.
- 5 Inject tunicamycin or vehicle solution subcutaneously at the base of the neck and return the mouse to the cage.
- 6 Perform a second identical injection 2 hours after the first.
- 7 Sacrifice mice 16 – 18 hours later.

Protocol references

Wang, H., Wang, X., Ke, Z. J., Comer, A. L., Xu, M., Frank, J. A., Zhang, Z., Shi, X., & Luo, J. (2015). Tunicamycin-induced unfolded protein response in the developing mouse brain. *Toxicology and Applied Pharmacology*, 283(3), 157–167. <https://doi.org/10.1016/j.taap.2014.12.019>