




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# 🌐 Injection of cholera toxin subunit B in the interscapular brown adipose fat and Stellate Ganglion dissection

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1 Works for me

 Share[dx.doi.org/10.17504/protocols.io.rm7vzykoxlx1/v1](https://dx.doi.org/10.17504/protocols.io.rm7vzykoxlx1/v1) Daniele Neri

## ABSTRACT

Protocol to deliver cholera toxin subunit B into the interscapular brown fat of mice

## DOI

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### Preparation

- 1 Prepare the surgery area by placing a clean paper towel on top of a heat pad
- 2 Anesthetize the animal under isoflurane until there's no toe pinch reflex
- 3 Transfer the mouse to the surgery area, flat on its stomach on the heatpad. Place the nose cone to maintain anesthesia
- 4 Shave the interscapular area of the mouse and sterilize it with three alternating wipes of betadine and 70% isopropyl alcohol
- 5 Apply eye ointment
- 6 Administer analgesics. We use buprenorphine 0.1 mg/kg subcutaneously
- 7 Load up cholera toxin subunit B (CTB) in a glass pulled needle connected to a mouth pipette (10 uL per side)

### Surgery

- 8 Make a 1 cm horizontal incision above the interscapular area
- 9 Visually identify the white adipose layer of interscapular pad, and use scissors to cut it and expose the underlying brown adipose tissue

- 10 While holding the tissue to expose the brown fat, inject 10 uL of CTb on each side
- 11 Gently return the fat pads to their original position, and close the wound with a 4-0 PS-4 suture
- 12 Clean the wound with betadine and 70% isopropyl alcohol and monitor the animal for any sign of distress

#### Tissue harvesting

- 13 Euthanize the mouse after 72 hours by intraperitoneal injection of 2-2-2-Tribromoethanol (500 mg/kg)
- 14 Open the ribcage and remove the lungs for easier access to the sympathetic chain
- 15 Carefully dissect the Stellate Ganglion from each side