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Mouse model of post-colitis (DNBS) chronic visceral hypersensitivity.

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This protocol is used to administer DNBS (Dinitrobenzene sulfonic acid) to the colorectum of mice via intracolonic enema administration to induce mild colonic inflammation.

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protocol

Grundy, L et al. Chronic linaclotide treatment reduces colitis-induced neuroplasticity and reverses persistent bladder dysfunction JCI Insight. 2018;3(19):e121841. <https://doi.org/10.1172/jci.insight.121841>.

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2,4-Dinitrobenzenesulfonic acid hydrate (DNBS), MERCK Sigma-Aldrich Catalogue number: 556971

KY Lubricating Jelly, Durex

- 1 Fast mice overnight by removing bedding and food from cages and placing a wire fasting rack at the bottom of cage. Provide 5% glucose drinking water. Record weight before fasting.
- 2 Weigh mouse following overnight fasting, if mouse has lost more than 10% of prefast weight, or is under 22 grams do not treat animal, rehouse in a cage with fresh bedding, free access to food and water.
- 3 Weigh out the desired amount of DNBS to achieve a concentration of 6.5mg/ml and in a volume enough to administer 0.10mL /mouse. Dissolve DNBS in the desired volume of 35% ethanol. Make sure to wear gloves during handling of DNBS and weigh in an appropriate Fume Cupboard.
- 4 Load a 1 ml syringe with 0.10mL DNBS. Attach a 3cm cannula to syringe and remove dead space.
- 5 Place mouse in induction box. Induce anesthesia with 5% isoflurane, 2% oxygen. Once the mouse is fully anaesthetized (absence of pedal reflex upon pinch) lower isoflurane to 1.5 % for maintenance.
- 6 Within induction box, place animal head down into a 50 mL syringe so that the mouse receives anesthetic whilst the lower trunk and rectum is accessible. Check pedal reflexes to ensure mouse is deeply anaesthetized.
- 7 Lubricate both the mouse rectum (KY Jelly, Durex) and the sides of the cannula and slowly insert the cannula into the rectum until it is 3 cm from the mouse rectum opening. Take care not to puncture the colon wall. If resistance to cannula insertion is felt, stop insertion and take note of the depth inserted. Resistance may be due to some residual faeces remaining in the colon even though the mouse has been fasted overnight, which may make it difficult to move the whole 3 cm.
- 8 Very slowly inject 0.10mL of DNBS solution as slowly withdrawn cannula out of the colorectum. As the cannula is withdrawn, the DNBS solution will gradually leak out of the cannula and not expose past the targeted area.
- 9 Remove mouse from tube and hold vertically for 30 seconds to prevent leakage of instilled DNBS due to gravity and allow uniform colorectal exposure.

- 10 Place mouse into a new cage with clean bedding and monitor recovery until anesthetic wears off and mouse is ambulatory.
- 11 Post-DNBS instillation mice are to be housed individually with free access to food and 5% glucose water, soaked food and sunflower seeds. Cages are to be placed on heat pads for 2-3 days post-DNBS instillation.
- 12 Monitor mice (including weigh) at least twice per day for 7 days post-DNBS enema as per approved post procedure monitoring requirements. Monitor and record for score severity of colitis (and related discomfort), such as weight loss, diarrhoea, constipation, blood in stools, GRIMACE (pain score), lack of movement and grooming (hunched/ruffled)