Naphthalene injections
Updated 8/27/07
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Males and females have a slightly different response to naphthalene (timing of response, etc.). Also, use experimental and control animals of the same background, as background also influences the timing of the naphthalene response. Or, just make sure you have sex- and bkground-matched control and experimentals. Better to do all in one sex and one background if possible.

new info
-best to do injections consistently at same time of day, between 8am and noon
-100-200 mg/kg is sufficient to ablate Clara cells in terminal bronchioles, 275 mg/kg is more important for complete ablation in more proximal regions. We will do 200 mg/kg as a usual, but the dose should be optimized for each genotype/background.
-corn oil can become contaminated and cause death/systemic infections several days after IP injections, so corn oil should be aliquoted and frozen; thaw and make fresh solution each time

Prepare naphthalene solution—standard is 20 mg/mL in corn oil
(have done as concentrated as 30 mg/mL in corn oil).
Naphthalene is an irritant, toxic material and is a flammable solid. Wear gloves, a lab coat, and eye protection while the material is being prepared and used. Preparation and use should be performed in a chemical fume hood.

Thaw out corn oil aliquot(s) (found in -20 in hallway)
Weigh in hood (mothball smell!)
Prep in 50 mL conical with corn oil, then wrap cap with parafilm very securely, place on rocker to mix (might take about an hour to dissolve).
It is best to prepare a fresh solution each time.

Control injection will be just corn oil, at 10mL/kg, or 10 uL per g.

Take small scale, syringes, needles (20-25G), naphthalene solution, corn oil to mouse room.
Weigh the mouse, determine the appropriate dose.
For naphthalene, if want 200 mg/kg, use 10uL/g of the 20mg/mL stock.
Corn oil 10 mL/kg (10uL/g).

Do the injection in the hood!
Draw up solution with syringe, then put a 20-25G needle on the syringe to inject by IP. Go very slowly, as the solutions are very thick, and if you push too hard, the pressure in the syringe will make the needle come off, mess, etc.

After IP injection, dab the injection site with paper towel so the mouse won't lick up too much naphthalene or corn oil.
Place an orange surgery card on the cage, stating naphthalene and date of administration, put transgel in bottom of cage

Important time points and info:
-some mice will die from liver toxicity within a day or two of injection
-mice will appear hunched, ruffled fur, sickly when Clara cell depletion is maximal, but they will recover
-48 hours: first BrdU+ cells in terminal bronchioles (all BASCs)
-52 hours: maximal Clara cell ablation
-72 hours: BrdU+ Clara cells also
-1-2 weeks: you will see repair underway; terminal bronchioles are covered, but not full of nice columnar cells yet
-6 weeks: 90% recovery of Clara cells

Information on repeated dosing to come…..