Making Virus

1. Talk to Alison Killilea in the Cell Culture Facility about getting SF-9 cells and media that you will need. If you have never worked in the Cell Culture Facility also ask her or an experienced lab mate for an orientation session.
2. Ask for some ESF 921 medium with 2% Fetal Bovine Serum and fungicides (PSF) and some ESF 921 medium without any supplements. Store extra medium in fridge.
3. For each transfection, you will want one well of a 6-well plate seeded at $1.5x10^6$. Allow cells to attach for at least one hour.
4. Prepare in snap-cap polystyrene tubes:
   Solution A: for each transfection 5 ul mini-prep bacmid+100 ul warmed ESF 921 without supplements.
   Solution B: for each transfection dilute 6 ul Cellfectin and 100 ul warmed ESF 921 without supplements. Note: Mix Cellfectin thoroughly before dilution.
5. Combine the solutions, mix gently and incubate 30-45 minutes at room temp.
6. Wash cells once with 2 ml ESF 921 without FBS or antibiotics.
7. For each transfection, add 0.8 ml ESF 921 without supplements to each tube with lipid-DNA complex. Mix gently.
8. Aspirate medium from cells and overlay 1 ml lipid-DNA complex onto cells.
9. Incubate 5 hr at 27°C.
10. Aspirate media from cells and overlay 3 ml ESF 921 with supplements.
11. Incubate 72 -96 hr. Remove supernatant to sterile tube. You can also scrape the cells into PBS and look for protein expression but don't expect too much.
12. Clarify the sup by spinning 5 min 1k in clinical centrifuge.
13. Move sup to new tube and store at 4°C in box in fridge.