

## **FUGENE TRANSFECTIONS of HeLa Cells**

1. Never start with an overconfluent flask
2. If transfecting for a pulse chase, I like to have 1 dish per time point.
3. Day 1: Spit cells into 10cm dishes with a final cell count of  $1.5 \times 10^6$
4. Day 2: Transfect with 10ug of DNA as followed
  - a. Take Fugene out and leave at RT for 15min
  - b. Mix 30ul of Fugene with 470ul of warm serum free DMEM
  - c. Incubate at RT for 5 min
  - d. Add drop wise to DNA
  - e. Incubate at RT for 10 min
  - f. Add 500ul drop wise to cells
5. Day 3: Split cells 1:2 \*Do not discard any cells
6. Day 4: Harvest