

Splitting adherent mammalian cells

1. Wash cells with 1x PBS pH 7.4 twice.
2. Add 5 mL trypsin-EDTA (Irvine Scientific cat# 9341) or more depending on size of dish.
3. Place dish of cells in 37°C incubator.
4. Check cells if they have detached after 5-10 minutes. This may take longer depending on the confluence of cells at trypsinization.
5. Shake dish to further detach cells if necessary.
6. Once all of the cells have detached, add an equal amount of media with serum.
7. Resuspend cells in this 1:1 mixture of media and trypsin.
8. Centrifuge cells to pellet.
9. Aspirate supernatant.
10. Resuspend cells in fresh media.
11. Plate cells according to your needs. If cells are for protein expression, split into many large flasks. If cells are for cell biology experiments, plate according to desired conditions.

Note: Always dispose of unwanted cells according to current state regulations!
Autoclave all used tissue culture wares before disposing as regular trash.