

HISTONE EXTRACTION PROTOCOL

1. Harvest cells and wash twice with ice-cold PBS. PBS can be supplemented with 5mM Sodium Butyrate to retain levels of histone acetylation.
2. Resuspend cells in Triton Extraction Buffer (TEB: PBS containing 0.5% Triton X 100 (v/v), 2mM phenylmethylsulfonyl fluoride (PMSF), 0.02% (w/v) NaN_3) at a cell density of 10^7 cells per ml.
3. Lyse cells on ice for 10 minutes with gentle stirring.
4. Centrifuge at 2000rpm for 10 minutes at 4°C. Remove and discard the supernatant.
5. Wash the cells in half the volume of TEB and centrifuge at before.
6. Resuspend the pellet in 0.2N HCl at a cell density of 4×10^7 cells per ml.
7. Acid extract the histones over night at 4°C.
8. Centrifuge samples at 2000rpm for 10 minutes at 4°C.
9. Removed the supernatant and determine protein content using the Bradford assay.
10. Store aliquots at -20°C.