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 phenylmethysulfonyl fluoride (PMSF), 0.02% (w/v) NaN₃) at a cell density of 10⁷ 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 4x10⁷ 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.