

Protocol for Lysing Red Blood Cells

Red Blood Cell Lysing Buffer, containing ammonium chloride, is formulated to lyse Red Blood Cells from human and murine samples while leaving the lymphocytes intact. This buffer, supplied at 10X dilution, should be diluted to 1X to produce the desired effect.

1. Dilute the 10X buffer to a 1X dilution using distilled water.
2. Warm the solution to room temperature.
3. Aliquote your sample of whole blood into a tube.
4. Add the Fluorescent conjugated antibodies to stain directly to the sample. Mix well and incubate at room temperature for 30 minutes, protected from light.
5. Add 2 mL of room temperature 1X buffer and pulse vortex.
6. Incubate for 10 to 15 minutes, protected from light.
7. Centrifuge at 500 xg for 5 minutes at room temperature and pour out the supernatant.
8. Resuspend the pellet with 1X with PBS containing 2% Fetal Bovine Serum and 0.02% Sodium Azide.
9. Centrifuge cells and resuspend to proceed with further analysis.