

Human Erythrocyte Lysing Solution

(Ammonium Chloride Lysing Solution, 10x)

(McCoy, J.P., 1998. Handling, storage and preparation of human blood cells. *Curr. Protocol. Cytom.* 5:5.1.1-5.1.13)

Reagents:

- 80.2 g NH₄CI (1.5 M)
- 8.4 g NaHCO₃ (100 mM)
- 3.7 g disodium EDTA (10 mM)
- Distilled Water
- 1 N HCI
- 1 N NaOH

Method:

- 1) Combine NH₄Cl, NaHCO₃ and disodium EDTA in 900ml distilled water. Mix to dissolve completely.
- 2) Adjust to pH 7.4 with 1 N HCl or 1 N NaOH.
- 3) Add distilled water to 1 liter final volume.

Store ≤6 months at 4° C

Working solution: Dilute 1:10 with distilled water. Make working lysing solution fresh before each use. Keep working solution cold and discard any unused portion.

Notes:

- This solution is used to lyse erythrocytes.
- Never store lysing solution at <10x concentration, as it will form ammonium carbonate, which is ineffective.
- Sodium bicarbonate may be replaced by 10 g KHCO₃ (100 mM) and disodium EDTA by 3.66 g tetrasodium EDTA (8.2 mM). If desired, lysing solution may be adjusted to a specific pH using 1 N HCl or 1 N NaOH.
- Some researchers prefer to use a 10-fold lower concentration of EDTA (I mM disodium EDTA or 0.82 mM tetrasodium EDTA in the 10x recipe).