



Cell Stimulation for Cytokine Production Protocol

Cell Stimulation Cocktail (500X) Cat. No. TNB-4975-UL100

Note: Cell Stimulation Cocktail is supplied as a 500X solution in Ethanol. Product should be stored at -20°C, protected from light and tightly sealed to prevent evaporation.

Other Materials Required

- RPMI complete medium (RPMI with 10% FBS, 1% pen/strep, 50 uM 2-ME)

For activation of mouse splenocytes:

1. Harvest spleen and prepare single cell suspension in RPMI complete medium at approximately $5-10 \times 10^6$ cells/mL.
2. Add 1X (2 μ L/mL) Cell Stimulation Cocktail (500X) directly to the cell suspension.
3. Culture overnight (6-18 hours) at 37°C, 5% CO₂. We typically leave this culture overnight which will generate a nice population of IL-2 and IFN-gamma expressing cells.