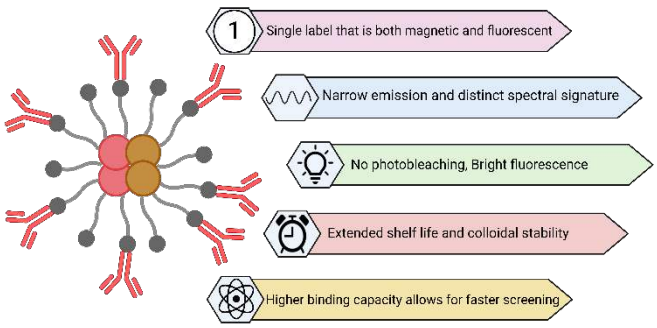
MagDot Labeling and Separation Protocol in CoreMag2T Magnet

Introduction

MagDot is a unique reagent that combines inorganic semiconductor nanocrystals and superparamagnetic iron oxide particles within the same nanoparticle. They have increased sensitivity, high fluorescence intensity, resistance to photobleaching, and improved shelf-life stability. Owing to its dual nature, it is possible to perform cell separation on MagDot labeled cells via Magnet Activated Cell Sorting (MACS) and immediately transfer the cells to Fluorescence Activated Cell Sorting (FACS) for further purification without any additional fluorescent labeling procedure.

CoreMag2T is an open magnetic system designed to be used with MagDot reagents. It has a field strength of 2 Tesla in the middle of the magnetic core and can hold 12 x 75 mm (5 mL) flow cytometry tubes. The inlet portion of the magnet has a rubber gasket to hold the tube firmly while being easily removable. The design enables the magnet to be kept in a half cylinder instead of a complete cylinder. As a result, the user can visualize the separation and confidently remove the supernatant.



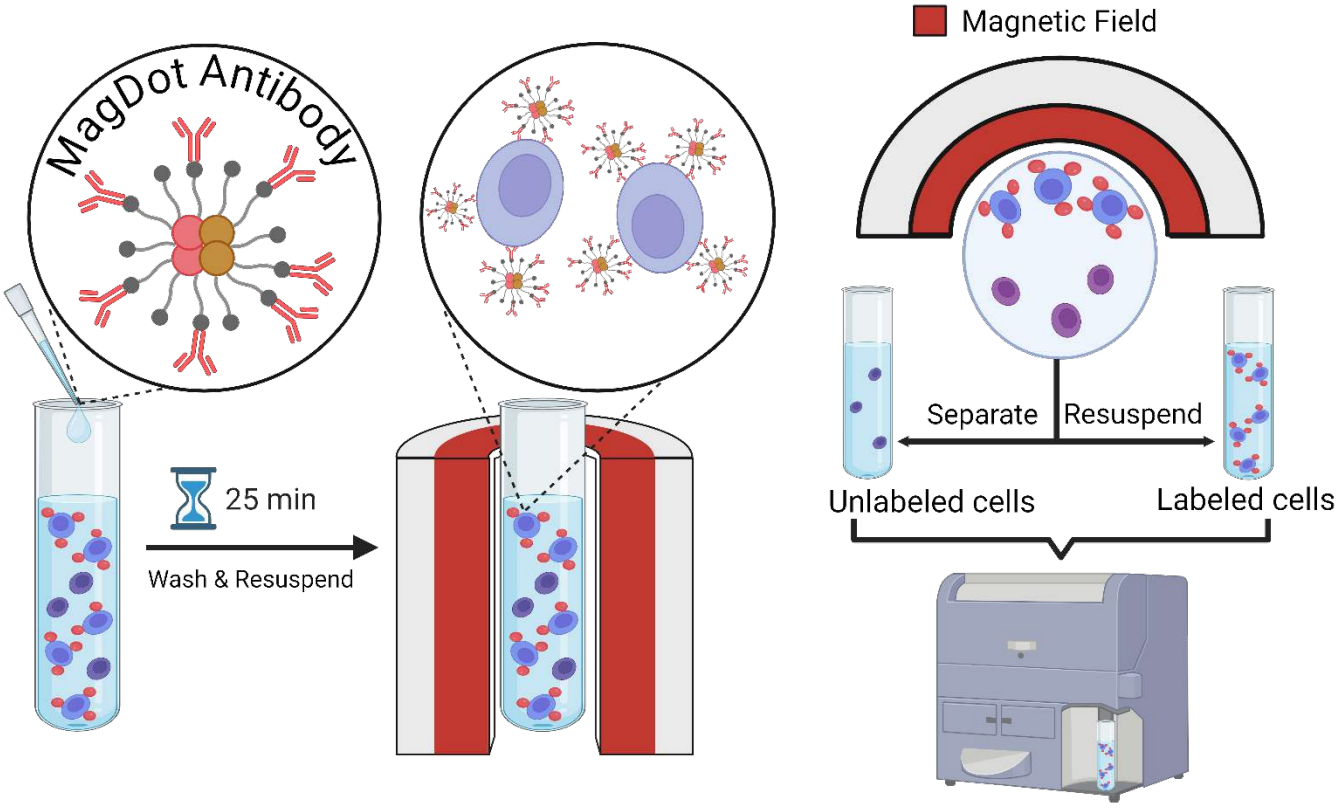
Reagents and Instruments

- Target cell suspension in PBS or buffer of choice
- Staining Buffer: PBS + 0.5% BSA + 2 mM EDTA
- MagDot antibody
- 12x75mm round bottom polystyrene tubes, 50 mL centrifuge tube
- Centrifuge, Vortex, Flow cytometer
- CoreMag2T (Core Quantum Technologies Inc.)

Direct Labeling and Separation in CoreMag2T

Sample Preparation: Prepare single-cell suspension in staining buffer with desired concentration of cells.

1. Resuspend 1E+06 cells in 200 µL of staining buffer in a flow tube.
2. Add appropriately titrated MagDot antibody to the flow tube, and pipette briefly to mix.
   Note: 25 µL of MagDot antibody for a million cells is recommended
3. Incubate for 25 minutes at room temperature.
   Note: If cells are very phagocytic, incubate at 4°C
4. Post incubation, add 300 µL of staining buffer to make the volume up to 500 µL.
5. Insert the flow cytometry tube in the CoreMag2T magnet for 30 minutes.
6. Post 30 minutes of separation, aspirate the solution containing unlabeled (non-magnetic) cell population and transfer it to another tube.
7. Remove the tube from the magnet and resuspend the labeled (magnetic) cell population in 400 µL of staining buffer.
8. Directly analyze the cells in flow cytometer.



MagDot antibody added to cell suspension

Cell suspension mixture transferred to CoreMag2T

Cells separated magnetically and directly analysed in a Flow Cytometer

*Image components are not to scale

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