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Flow Cytometry Staining Protocol For Initial Kidney Studies

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Abstract

Protocol for flow cytometry cell staining for initial kidney studies

Troubleshooting

1 Cell suspensions were centrifugated (400xg, 5 min) and then stained concurrently with 5 µg/mL anti-mouse CD45-AF488 (clone 30-F11, BioLegend, San Diego, CA), 7 µg/mL EpCAM-PE (clone G8.8, BioLegend, San Diego, CA), and 5 µg/mL TER119-AF647 (clone TER-119, BioLegend, San Diego, CA) monoclonal antibodies in PBS+1% BSA (PBS+) for 30 minutes.

2 Samples were then washed twice with ~2mL PBS+ by centrifugation (400xg, 5 min).

3 Cell suspensions were then stained with 3.33 µg/mL 7-AAD viability dye (BD Biosciences, San Jose, CA) on ice for at least 10 minutes.

4 Cell suspensions were then analyzed on a Novocyte 3000 Flow Cytometer (ACEA Biosciences, San Diego, CA).