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FACS for CD45+ immune cells

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Protocol status: Working

We use this protocol and it's working

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Abstract

This protocol details FACS of intestinal immune cells (CD45+).

Attachments



[786-1993.pdf](#)

45KB

Materials

Reagents

 BD Pharmingen™ Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™) **BD Biosciences Catalog #553142**

 BD Horizon™ BUV395 Rat Anti-Mouse CD45 **BD Biosciences Catalog #564279**

 Propidium Iodide Solution **BioLegend Catalog #421301**

Equipment

Sony MA900 Multi-Application Cell Sorter



Procedure

52m

- 1 Resuspended cells isolated from the small intestine with  400 μL Fc block (BD Biosciences, Cat# 553142; Franklin Lakes, NJ), 1:200 in FACS buffer.
- 2 Incubate for  00:20:00  On ice . 20m

- 3 Wash cells in  500 μL FACS buffer, pellet at  1500 rpm, 00:02:00 , then resuspend in  600 μL of FACS buffer with 1:100 CD45 BUV395 antibody (BD Biosciences, Cat# 564279; Franklin Lakes, NJ). 2m

- 4 Incubate  On ice for  00:30:00 . 30m

- 5 After 30 minutes, wash cells in  500 μL FACS buffer. 
- 6 Resuspend in  500 μL of FACS buffer containing propidium iodide (BioLegend, Cat# 421301; San Diego, CA) at 1:10,000.
- 7 Sort viable, CD45+ cells on a Sony MA900 Multi-Application Cell Sorter.