

Mouse Treg Flow™ Kit (FOXP3 Alexa Fluor® 488/CD4 APC/CD25 PE)

Catalog # / Size: 320018 / 25 tests

Clone: 150D

Reactivity: Mouse

Storage: Upon receipt, store at 4°C and protected from prolonged exposure to light. Do not freeze.

Application: FC, ICFC

Application Notes:

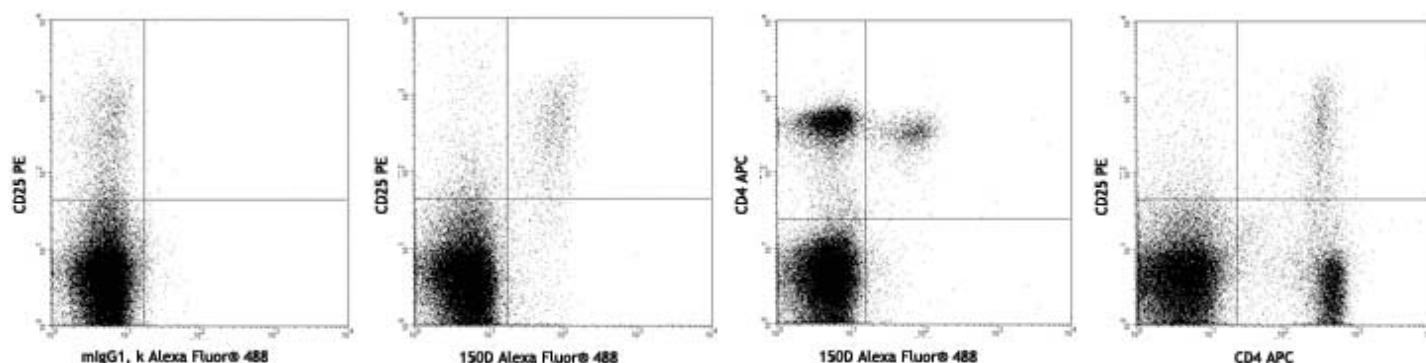
Immunofluorescence Staining Procedures:

1. Distribute 0.5-1 X 10⁶ cells/100 µl/tube into 12 X 75 mm plastic testing tubes.
2. Add 20 µl of CD4 APC/CD25 PE cocktail to each tube, vortex and incubate at room temperature in the dark for 20 minutes.
3. Wash once with 1 ml cell staining buffer by spin at 250Xg for 5 minutes and discard the supernatant.
4. Prepare 1X working solutions of FOXP3 Fix/Perm buffer and FOXP3 Perm buffer prior to staining. The FOXP3 Fix/Perm buffer (4X) must be freshly diluted by diluting one (1) part FOPX3 Fix/Perm buffer (4X) with three (3) parts PBS. The FOXP3 Perm buffer (10X) should be diluted by diluting one (1) part FOXP3 Perm buffer (10X) with nine (9) parts of PBS.

Caution: The FOXP3 Fix/Perm buffer contains paraformaldehyde, which is toxic and mutagenic.

5. Add 1 ml of 1X BioLegend's FOXP3 Fix/Perm solution to each tube, vortex and incubate at room temperature in the dark for 20 minutes, then spin down the cells and remove the supernatant.
6. Wash once with cell staining buffer by spin at 250Xg for 5 minutes and remove the supernatant.
7. Wash once with 1ml 1X BioLegend's FOXP3 Perm buffer.
8. Re-suspend cells in 1ml 1X BioLegend's FOXP3 Perm buffer, incubate at room temperature in the dark for 15 minutes, spin down cells and discard the supernatant, then resuspend the pellet in 100 µl of 1X BioLegend's FOXP3 Perm buffer.
9. Add 5 µl Alexa Fluor® 488 anti-mouse FOXP3 antibody or 5 µl Alexa Fluor® 488 mouse IgG1, k isotype control into appropriate tube and incubate at room temperature in the dark for 30 minutes.
10. Wash twice with cell staining buffer, and resuspend in 0.5ml cell staining buffer then analyze with flow cytometer with appropriate instrument setting.

*** Alexa Fluor® is a registered trademark of Molecular Probes, Inc. Alexa Fluor® dye antibody conjugates are sold under license from Molecular Probes, Inc. for research use only, except for use in combination with microarrays and high content screening, and are covered by pending and issued patents.



BALB/c mouse splenocytes stained with Mouse Treg Flow™ Kit (FOXP3 Alexa Fluor® 488/CD4 APC/CD25 PE)



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Application References:

1. Roncador, G., et al., 2005 Eur. J. Immunol. 35:1681.
2. Luke, P.P.W., et al. 2006. Amer. J. Transplant. 6(9):2023.
3. Bamias, G., et al., 2007. J. Immunol. 178:1809.
4. Valencia, X., et al., 2007. J. Immunol. 178:2579.
5. Davidson, T.S., et al., 2007. J. Immunol. 178:4022.
6. MacDonald, K. PA., et al., 2007. Blood doi:10.1182/blood-2007-01-067249.
7. Müller, M., et al., 2007. J. Immunol. 179:2774.
8. Jaffar, Z., et al., 2007. J. Immunol. 179:6193

Description:

T regulatory (Treg) cells are a subset of T lymphocytes which is characterized by CD4+/CD25+/FOXP3+. These naturally occurring Treg cells originate in the thymus, and comprise 2-10% of peripheral CD4+ T cells. It has been shown that Treg cells are able to inhibit T cells proliferation and cytokine production and play critical roles in preventing autoimmunity as well as in controlling tumor immunity and transplantation tolerance. Impaired Treg function or Treg cell deficiency will develop variety of autoimmune diseases, while higher frequency of Treg cells will cause hypo-immune response to pathogens.

BioLegend's Mouse Treg Flow™ Kit is designed and formulated specifically for immunofluorescence staining and flow cytometric analysis of mouse Treg cells in a mixed lymphocyte population. This kit is composed of fluoro-chrome conjugated anti-mouse CD4, CD25 and FOXP3 antibodies and the critical buffers. It is easy to use for identification of Treg cells.

Materials Provided:

1. Alexa Fluor® 488 anti-mouse FOXP3, 25 tests (Cat. No. 320011)
2. Alexa Fluor® 488 Mouse IgG1, k isotype control, 25 tests (Cat. No. 400133)
3. FOXP3 Fix/Perm buffer and FOXP3 Perm buffer, 100 tests (Cat. No. 421403)
4. anti-mouse CD4 APC/CD25 PE Cocktail, 50 tests (Cat. No. 121301)

Materials not included:

Cell Staining Buffer (Cat. No. 420201)

Other Names:

Forkhead box protein P3, Scurfin, JM2, IPEX, Zinc finger protein JM2, T regulatory cells, Regulatory T cells

Antigen References:

1. Hori, S., et al. 2003. Science 299:1057.

Related Products

	Clone	Application
Cell Staining Buffer (FBS)	Soln4202	FC, ICC, ICFC



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