BioLegend
The path to legendary discovery™

Product Data Sheet

FOXP3 Fix/Perm Buffer Set

Catalog # / Size: 421403 / 100 tests

Storage: The FOXP3 fix/perm buffer set solution should be stored between 2°C and 8°C. **Do not freeze.** This set is guaranteed for three months.

This product has a shelf-life of 12 months or less. Please use our Expiration Lookup Tool (www.biolegend.com/concentrationlookup) to verify the expiration date of your lot of product.

Applications:

Applications: ICFC - *Quality tested*

Recommended Usage: The FOXP3 Fix/Perm buffer(4X) must be diluted to 1X working solution with PBS (10 mM sodium phosphate, 150 mM sodium chloride, pH 7.2) prior to staining procedures. *e.g.* Dilute one (1) part FOXP3 Fix/Perm buffer(4X) to three (3) parts PBS. The FOXP3 Perm buffer (10X) should be diluted to 1X working solution with PBS (10 mM sodium phosphate, 150 mM sodium chloride, pH 7.2) prior to staining procedures. *e.g.* Dilute one (1) part FOXP3 Perm buffer(10X) to nine (9) parts PBS.
NOTE: The FOXP3 Perm buffer (10X) may have crystalization or precipitation oberserved when it is stored at 2-8°C, however, it's normal and does not affect the buffer performance. If there is a heavy precipitation observed after diluted to 1X working solution, it can be filtered to clarify the solution.

Application Notes: **FOXP3 Intracellular Staining Procedures:**
1. Perform cell surface staining as described in BioLegend's Cell Surface Immunofluorescence Staining Protocol.
2. 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.
3. Wash once with cell staining buffer (Cat. No. 420201) by spin at 250Xg for 5 minutes and remove the supernatant.
4. Wash once with 1ml 1X BioLegend's FOXP3 Perm buffer.
5. 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 ul of 1X BioLegend's FOXP3 Perm buffer.
6. Add appropriate amount of flurochrome conjugated anti-FOXP3 antibody and incubate at room temperature in the dark for 30 minutes.
7. Wash twice with cell staining buffer, and resuspend in 0.5ml cell staining buffer then analyze with flow cytometer with appropriate instrument setting.

Application References:

1. Yang ZZ, *et al.* 2006. *Blood* 107:3639.PubMed
2. Groh V, *et al.* 2006. *Nat. Immunol.* 7:755.PubMed
3. Bamias G, *et al.* 2007. *J. Immunol.* 178:1809.PubMed
4. MacDonald K PA, *et al.* 2007. *Blood* doi:10.1182/blood-2007-01-067249.PubMed
5. Müller M, *et al.* 2007. *J. Immunol.* 179:2774.
6. Dang Y,*et al.*2007.*Clin Cancer Res.*13:1883.PubMed
7. Basu S, *et al.* 2008. *J. Immunol.* 180:5974. PubMed
8. Takamura S, *et al.* 2010. *J. Immunol.* 184:4696. PubMed
9. Beavis, PA., *et al.* 2011. *PNAS.* 108:16717. PubMed
10. Kling J, *et al.* 2013. *Exp Parasitol.* 129:270. PubMed
11. Troutman TD, *et al.* 2012. *PNAS.* 109:273. PubMed
12. Faustman DL, *et al.* 2012. *PLoS One.* 7:e41756. PubMed
13. Liu Y, *et al.* 2012. *Food Chem Toxicol.* 50:1920. PubMed
14. Kawamoto N, *et al.* 2013. *Int Immunol.* 25:53. PubMed
15. Morandi B, *et al.* 2013. *J. Immunol.* 191:4858. PubMed
16. Shiba T, *et al.* 2014. *Toxicol Appl Pharmacol.* 274:191. PubMed
17. Cook KW, *et al.* 2014. *Gut.* PubMed
18. Shibuya KC, *et al.* 2014. *PLoS One.* 9:96565. PubMed
19. Cecil DL, *et al.* 2014. *Cancer Res.* 74:2710. PubMed
20. Linnerbauer S, *et al.* 2014. *PLoS Pathog.* 10:1004068. PubMed

Description: BioLegend's FOXP3 Fix/Perm Buffer Set has been specially formulated for intracellular staining FOXP3 with minimum effect on the surface fluorochrome staining. It contains one 30 ml 4X concentrated FOXP3 Fix/Perm buffer (Cat. No. 421401) and one 25 ml 10X concentrated FOXP3 Perm buffer (Cat. No. 421402). The FOXP3 Fix/Perm buffer(4X) must be freshly diluted to 1X working solution with PBS (10 mM sodium phosphate, 150 mM sodium chloride, pH 7.2) prior to staining procedures. *e.g.* Dilute one (1) part FOXP3 Fix/Perm buffer(4X) to three (3) parts PBS. The FOXP3 Perm buffer (10X) should be diluted to 1X working solution with PBS (10 mM sodium phosphate, 150 mM sodium chloride, pH 7.2) prior to staining procedures. *e.g.* Dilute one (1) part FOXP3 Perm buffer(10X) to nine (9) parts PBS.
Caution: The FOXP3 Fix/Perm buffer contains paraformaldehyde, which is toxigenic and mutagenic. Please handle with caution and wear gloves, lab coat and necessary protection to avoid direct body contact.

Related Products:

Product	Clone	Application
Cell Staining Buffer		FC, ICC, ICFC

For research use only. Not for diagnostic use. Not for resale. BioLegend will not be held responsible for patent infringement or other violations that may occur with the use of our products.

*These products may be covered by one or more Limited Use Label Licenses (see the BioLegend Catalog or our website, www.biolegend.com/ordering#license). BioLegend products may not be transferred to third parties, resold, modified for resale, or used to manufacture commercial products, reverse engineer functionally similar materials, or to provide a service to third parties without written approval of BioLegend. By use of these products you accept the terms and conditions of all applicable Limited Use Label Licenses. Unless otherwise indicated, these products are for research use only and are not intended for human or animal diagnostic, therapeutic or commercial use.

BioLegend Inc., 9727 Pacific Heights Blvd, San Diego, CA 92121 www.biolegend.com
Toll-Free Phone: 1-877-Bio-Legend (246-5343) Phone: (858) 768-5800 Fax: (877) 455-9587

Purified anti-mouse/rat/human FOXP3	150D	WB, ICFC
Alexa Fluor® 488 anti-mouse/rat/human FOXP3	150D	ICFC
Alexa Fluor® 647 anti-mouse/rat/human FOXP3	150D	ICFC
Purified anti-human FOXP3	206D	WB, ICFC, IHC
Purified anti-human FOXP3	259D	WB, IHC, ICFC
Alexa Fluor® 488 anti-human FOXP3	259D	ICFC
Alexa Fluor® 488 anti-human FOXP3	206D	ICFC
Alexa Fluor® 647 anti-human FOXP3	206D	ICFC

For research use only. Not for diagnostic use. Not for resale. BioLegend will not be held responsible for patent infringement or other violations that may occur with the use of our products.

*These products may be covered by one or more Limited Use Label Licenses (see the BioLegend Catalog or our website, www.biolegend.com/ordering#license). BioLegend products may not be transferred to third parties, resold, modified for resale, or used to manufacture commercial products, reverse engineer functionally similar materials, or to provide a service to third parties without written approval of BioLegend. By use of these products you accept the terms and conditions of all applicable Limited Use Label Licenses. Unless otherwise indicated, these products are for research use only and are not intended for human or animal diagnostic, therapeutic or commercial use.

BioLegend Inc., 9727 Pacific Heights Blvd, San Diego, CA 92121 www.biolegend.com
Toll-Free Phone: 1-877-Bio-Legend (246-5343) Phone: (858) 768-5800 Fax: (877) 455-9587