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## Purified Mouse anti-FoxP3 Monoclonal Antibody

**CLX428AP**

**Lot:**

**Size:** 0.1 mg

**Clone:** 3G3

**Isotype:** Mouse IgG1

**Specificity:** The mouse monoclonal antibody 3G3 recognizes N-terminal region of FoxP3, a 47 - 55 kDa transcription factor, which is the master regulator in the development and function of regulatory T cells.

**Immunogen:** Full-length His-tagged recombinant murine FoxP3

**Species Reactivity:** Human, Mouse

**Application:** Flow Cytometry  
Application note:  
Staining method:  
- Perform staining of cell surface markers (CD25, CD4 etc.) for 20 min. in the dark and RT. Wash.  
- Add cold fixation buffer diluted to working concentration (if commercial reagent is used, follow the manufacturer's instructions) and incubate for 15 min. in 4°C.  
- Add cold permeabilization buffer diluted to working concentration (if commercial reagent is used, follow the manufacturer's instructions) to the pellet of cells.  
- Add 5-50µl of blocking solution (1% glycine, 0.1% gelatin) and incubate for 15 min. (4°C, in the dark).  
- Perform intracellular staining of FoxP3 for 30 min. (4°C, in the dark) with appropriate amount of 3G3 antibody (final concentration 2-10 µg/ml; optimal concentration for peripheral blood is 3 µg/ml).  
- 2 x wash with staining buffer (PBS with 0.2% BSA), centrifugate (1500 g, 5 min. 4°C)  
- Keep cold until measuring on a flow cytometry device with appropriate setting. (If necessary, for the same sample use mouse IgG1 isotype control (MOPC-21) in the same format as anti-FoxP3 antibody 3G3.)  
Western Blotting  
Recommended dilution: 2 µg/ml

*Continued...*

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- Purity:** > 95% (by SDS-PAGE)
- Purification:** Purified from ascites by protein-A affinity chromatography.
- Concentration:** 1 mg/ml
- Storage Buffer:** Phosphate buffered saline (PBS) with 15 mM sodium azide, approx. pH 7.4
- Storage / Stability:** Store at 2-8°C. Do not use after expiration date stamped on vial label. For long-term storage aliquot and store at -20°C. Avoid freeze/thaw cycles.
- Background:** FoxP3 (Forkhead box protein 3), a highly conserved forkhead/winged-helix transcription factor, plays a crucial role in maintaining immune homeostasis by governing the development and function of regulatory T cells. It is constitutively expressed at high level in CD25+ CD4+ Treg cells and at low level in a CD25-CD4+ T reg cell subset. Defects in gene encoding FoxP3 protein cause the scurfy phenotype in mice, and in human the IPEX syndrome (immune dysfunction, polyendocrinopathy, enteropathy, X-linked syndrome), also known as X-linked autoimmunity-allergic dysregulation (XLAAD) syndrome.
- References:**
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  - \*Law JP, Hirschhorn DF, Owen RE, Biswas HH, Norris PJ, Lanteri MC: The importance of Foxp3 antibody and fixation/permeabilization buffer combinations in identifying CD4+CD25+Foxp3+ regulatory T cells. *Cytometry A.* 2009 Dec;75(12):1040-50.

*This product is made available in collaboration with Exbio Praha, a.s.*

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