

## Technical Data Sheet

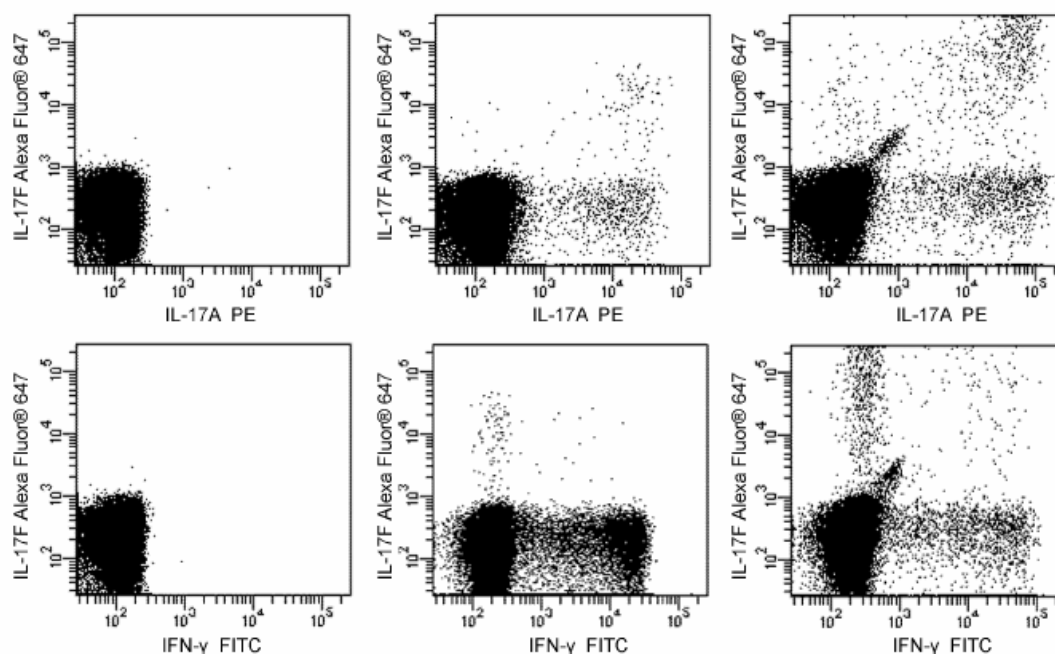
## Alexa Fluor® 647 Mouse anti-Human IL-17F

## Product Information

<b>Material Number:</b>	<b>561333</b>
<b>Alternate Name:</b>	Interleukin-17F; cytokine ML-1; ML-1; IL-24
<b>Size:</b>	100 tests
<b>Vol. per Test:</b>	5 µl
<b>Clone:</b>	O33-782
<b>Immunogen:</b>	Human IL-17F
<b>Isotype:</b>	Mouse IgG1, κ
<b>Reactivity:</b>	QC Testing: Human
<b>Storage Buffer:</b>	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

## Description

The O33-782 monoclonal antibody specifically binds to Interleukin-17F (IL-17F). IL-17F is a member of the IL-17 family of cytokines. IL-17F is encoded by the *IL17F* gene located in chromosome 6 (location: 6p12). IL-17F is a proinflammatory cytokine that is produced by activated T cells including differentiated CD4+ T helper 17 (Th17) cells. Activated Th17 cells can express disulfide-linked IL-17F and IL-17A homodimers as well as IL-17A/IL-17F heterodimers. These IL-17 dimers act by binding to and signaling through IL-17 receptor complexes (IL-17R). IL-17R are comprised of transmembrane IL-17RA and IL-17RC protein subunits that are expressed by a variety of target cells including epithelial and endothelial cells, keratinocytes, fibroblasts, and granulocytes. IL-17F can induce target cells to produce proinflammatory cytokines such as IL-1β, IL-6, G-CSF, GM-CSF, and TNF and chemokines including CXCL1/Gro-α, CXCL2/Gro-β, and CXCL8/IL-8 that attract and activate leukocytes, eg, neutrophils. Th17 and other IL-17F-producing cells play protective roles in the clearance of extracellular pathogens, including bacteria and fungi. IL-17F can also play adverse roles in inflammation associated with asthma and autoimmune diseases.



**Flow cytometric analysis of IL-17F expression by resting or activated human peripheral blood mononuclear cells (PBMC).** Human PBMC were either unstimulated (Left Panels) or stimulated with Phorbol 12-Myristate 13-Acetate (PMA; Sigma P-8139) plus Ionomycin (Sigma; I-0634) in the presence of BD GolgiStop™ Protein Transport Inhibitor (Cat. No. 554724) for 5 hours (Middle Panels) or cultured in Th17 polarization conditions and restimulated with PMA and Ionomycin in the presence of BD GolgiStop™ for 5 hours (Right Panels). Cells were then fixed and permeabilized using BD Cytotfix/Cytoperm™ reagents (Cat. No. 554714) followed by staining with Alexa Fluor® 647 Mouse anti-Human IL-17F (Cat. No. 561333), PerCP-Cy5.5 Mouse anti-Human CD4 (Cat. No. 341654), FITC Mouse Anti-Human IFN-γ (Cat. No. 554700) and PE Mouse anti-Human IL-17A (Cat. No. 560486). Two-color flow cytometric dot plots showing the correlated expression patterns of IL-17F versus IL-17A or IFN-γ were derived from CD4+ gated events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometry was performed using a BD™ LSR II Flow Cytometry System. Other compatible fixation and permeabilization treatments are listed in the "Recommended Assay Procedure."

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## Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed.

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

## Application Notes

### Application

Intracellular staining (flow cytometry)	Routinely Tested
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### Recommended Assay Procedure:

This antibody conjugate is suitable for intracellular staining of human peripheral blood mononuclear cells using BD Cytotfix/Cytoperm™ reagents, BD Pharmingen™ Human FoxP3 Buffer Set or BD™ Phosflow fixation and permeabilization buffers (Fix buffer I with Perm/Wash Buffer I, Perm Buffer II, or Perm Buffer III).

### Suggested Companion Products

Catalog Number	Name	Size	Clone
554724	Protein Transport Inhibitor (Containing Monensin)	0.7 ml	(none)
554714	BD Cytotfix/Cytoperm™ Fixation/Permeabilization Kit	250 tests	(none)
341654	PerCP-Cy5.5 Mouse anti-Human CD4	50 tests	SK3
554700	FITC Mouse Anti-Human IFN-γ	0.1 mg	B27
560486	PE Mouse anti-Human IL-17A	100 tests	N49-653
560098	Human FoxP3 Buffer Set	100 tests	(none)
557870	Fix Buffer I	250 ml	(none)
557885	Perm/Wash Buffer I	125 ml	(none)
558052	Perm Buffer II	125 ml	(none)
558050	Perm Buffer III	125 ml	(none)

### Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use  $1 \times 10^6$  cells in a 100-μl experimental sample (a test).
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Please refer to [www.bdbiosciences.com/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.
4. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
5. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
6. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
7. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
8. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
9. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

### References

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Wang YH, Liu YJ. The IL-17 cytokine family and their role in allergic inflammation. *Curr Opin Immunol.* 2008; 20(6):697-702. (Biology)

Wright JF, Bennett F, Li B, et al. The human IL-17F/IL-17A heterodimeric cytokine signals through the IL-17RA/IL-17RC receptor complex. *J Immunol.* 2008; 181(4):2799-2805. (Biology)

Wright JF, Guo Y, Quazi A, et al. Identification of an interleukin 17F/17A heterodimer in activated human CD4+ T cells. *J Biol Chem.* 2007; 282(18):13447-13455. (Biology)