

# Human and Mouse Reactive T-bet Monoclonal Antibodies

## Features

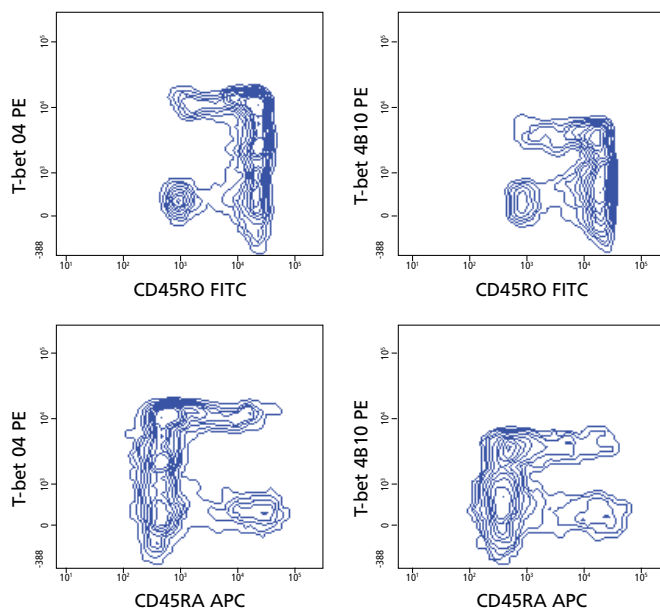
Useful marker for the detection of Th1, NK, and other IFN- $\gamma$  expressing cells.

Both the 4B10 and high-sensitivity clone O4-46 react with human and mouse T-bet.

Suitable for use in several applications including flow cytometry, Western blot, and immunohistochemistry.

Available in several formats including purified, PE, and Alexa Fluor® 647. High-sensitivity clone O4-46 is also available in Alexa Fluor® 488, BD Horizon™ V450, and PerCP-Cy™5.5.

Compatible with BD Phosflow™ buffer systems including BD Phosflow Lyse/Fix Buffer and BD Phosflow Permeabilization Buffers I through IV.



**Figure 1.** Whole blood was treated with BD Phosflow Lyse/Fix Buffer (Cat. No. 558049). Cells were permeabilized with BD Phosflow Perm Buffer III (Cat. No. 558050), washed, and stained with PE Mouse anti-T-bet Clone O4-46 (Cat. No. 561268) or Clone 4B10 (Cat. No. 561265), FITC Mouse Anti-Human CD45RO (Cat. No. 555492), APC Mouse Anti-Human CD45RA (Cat. No. 550855), BD Horizon™ V450 Mouse Anti-Human CD3 (Cat. No. 560365), and PerCP Mouse Anti-Human CD4 (Cat. No. 347324) antibodies. Contour plots showing CD45RO (top plot) or CD45RA (bottom plot) versus T-bet expression patterns for CD3<sup>+</sup>CD4<sup>+</sup> lymphocytes were generated using a BD FACSCanto™ II flow cytometer.

BD Biosciences now offers two clones, O4-46 and 4B10, for the study of the Th1 signature transcription factor, T-bet, in human and mouse samples. T-bet is expressed in response to antigen triggered T cell receptor (TCR) signaling and several cytokines. T-bet expression leads to many cellular responses including the expression of interferon- $\gamma$  (IFN- $\gamma$ ).

## T-bet (clones O4-46 and 4B10) purified and fluorochrome-conjugated antibodies

The new BD Pharmingen™ T-bet monoclonal antibodies (O4-46 and 4B10) react with human and mouse T-bet (T-box gene expressed in T cells), a master regulatory transcription factor that is also known as TBX21 (T-box21) and TBLYM (T-box transcription factor, expressed in lymphocytes). Human and mouse T-bet protein sequences are 88% homologous.<sup>1</sup>

Available conjugates include Alexa Fluor® 488, BD Horizon V450, and PerCP-Cy5.5 (clone O4-46 only), Alexa Fluor® 647, and PE formats to enable maximum flexibility for design of multicolor fluorescent antibody staining panels in combination with any BD FACS™ brand flow cytometer.

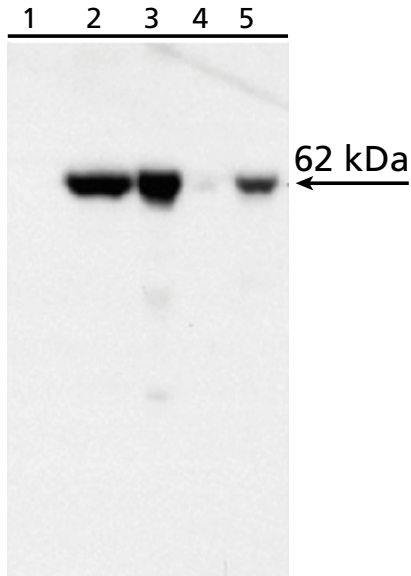
T-bet was discovered through a yeast one-hybrid screen using the Th1-specific portion of the IL-2 promoter linked to a reporter gene. T-bet was then transformed with constructs from an activated Th1 cell cDNA library fused to an activation domain. After further selection, eight clones were identified that contained sequences that were related to or identical to T-bet.<sup>1</sup>

Expression of T-bet leads to the expression of IFN- $\gamma$  through binding to enhancers on the promoter of the *Ifn $\gamma$*  gene. T-bet further stabilizes the Th1 phenotype through binding to a silencer on the *il4* gene, preventing the expression of the Th2 signature cytokine IL-4.<sup>2</sup>

In addition to Th1 cells, T-bet is expressed by and activates transcriptional activities within other hematopoietic cells, including stem cells, NK, and NKT cells and subsets of thymocytes, primed/activated CD4<sup>+</sup> T cells, CD8<sup>+</sup> T cells, and  $\gamma\delta$  T cells, B cells, and dendritic cells.

Visit [bdbiosciences.com/tcell](http://bdbiosciences.com/tcell) for more information.

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**Figure 2.** Western blot analysis of T-bet expressed by human and mouse cells. Lysates from mouse D10.G4.1 (Th2, lane 1) and 2D6 (Th1, lane 2) cells and from untreated human NK-92 (lane 3) and Jurkat (lane 4) cells and PBMCs (lane 5) were electrophoresed (15 µg total cellular protein/lane) and blotted using Purified Mouse anti-T-bet (Clone O4-46, Cat. No. 561263) antibody (1 µg/mL). T-bet is identified as a band of ~62 kDa.

### References

1. Szabo SJ, Kim ST, Costa GL, Zhang X, Fathman CG, Glimcher LH. A novel transcription factor, T-bet, directs Th1 lineage commitment. *Cell*. 2000;100:655-669.
2. Amsen D, Spilianakis CG, Flavell RA. How are T(H)1 and T(H)2 effector cells made? *Curr Opin Immunol*. 2009;21:153-160.

### Ordering Information

Description	Clone	Isotype	Format	Size	Cat. No.
Human and Mouse T-bet	O4-46	Mouse IgG <sub>1</sub> , κ	Purified	0.1 mg	561263
			Alexa Fluor® 488	50 tests	561266
			Alexa Fluor® 647	50 tests	561267
			BD Horizon™ V450	50 tests	561312
			PE	50 tests	561268
			PerCP-Cy5.5	50 tests	561316
Human and Mouse T-bet	4B10	Mouse IgG <sub>1</sub> , κ	Purified	0.1 mg	561262
			Alexa Fluor® 647	50 tests	561264
			PE	50 tests	561265

### Support Products

Description	Clone	Isotype	Format	Size	Cat. No.
Mouse IgG <sub>1</sub> , κ Isotype Control	MOPC-21	Mouse IgG <sub>1</sub> , κ	Alexa Fluor® 488	50 tests	557782
			Alexa Fluor® 647	50 tests	557783
			PE	100 tests	559320
			BD Horizon V450	0.1 mg	560373
			PerCP-Cy5.5	0.1 mg	550795
BD Phosflow Lyse/Fix Buffer 5X				250 mL	558049
BD Phosflow Perm/Wash Buffer I				125 mL	557885
BD Phosflow Perm Buffer II				125 mL	558052
BD Phosflow Perm Buffer III				125 mL	558050
BD Phosflow Perm Buffer IV, 10X				50 mL	560746



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23-11733-01

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