## **Technical Data Sheet**

# **Biotin Rat Anti-Human CD120b**

#### **Product Information**

Material Number: 552477

Alternate Name: TNF Receptor Type II

 Size:
 0.5 mg

 Concentration:
 0.5 mg/ml

 Clone:
 hTNFR-M1

Immunogen: COS-expressed recombinant human TNFRII

Isotype:Rat IgG2b,  $\kappa$ Reactivity:QC Testing: Human

Storage Buffer: Aqueous buffered solution containing ≤0.09% sodium azide.

### Description

The hTNFR-M1 antibody reacts with the extracellular domain of the 75 kDa transmembrane receptor for the human cytokines, tumor necrosis factor (TNF or TNF- $\alpha$ ) and lymphotoxin-alpha (LT- $\alpha$ 3, aka, lymphotoxin or TNF- $\beta$ ). This receptor is referred to as the p75 or Type II Tumor Necrosis Factor Receptor (TNFRII) [aka, CD120b]. Human TNFRII proteins are expressed by hematopoietic cells including macrophages, neutrophils, lymphocytes, thymocytes and mast cells. TNFRII is expressed by a variety of other cell types including endothelial cells, cardiac myocytes and prostate cells. Naive B cells express very low or undetectable levels of TNFRII whereas mature erythrocytes and platelets are uniformly negative for TNFRII expression. The immunogen used to generate the hTNFR-M1 hybridoma was COS- expressed recombinant human TNFRII.

### **Preparation and Storage**

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. The antibody was conjugated with biotin under optimum conditions, and unreacted biotin was removed. Store undiluted at  $4^{\circ}$  C and protected from prolonged exposure to light. Do not freeze.

### **Application Notes**

### Application

Application	
ELISA Detection	Routinely Tested
Flow cytometry	Reported

#### **Recommended Assay Procedure:**

**ELISA Detection:** Biotinylated hTNFR-M1 (Cat. No. 552477) serves as the detection antibody in a sandwich ELISA for measuring human TNFRII protein levels. hTNFR1-M1 (Cat. No. 552477) can be paired with purified MR2-2 anti-human TNFRII using the recombinant human soluble TNFRII as the standard. This detection antibody should be titrated between 0.5 -1 μg/ml to determine its optimal concentration for ELISA detection. To obtain linear standard curves, doubling dilutions of recombinant soluble human TNFRII ranging from 1000 to 10 pg/ml are recommended for inclusion in each ELISA plate. For specific methodology, please visit the protocols section or chapter on ELISA in the Immune Function Handbook, both of which are posted on our web site, www.bdbiosciences.com.

*Note:* This ELISA antibody pair shows no cross-reactivity with the following recombinant human cytokines: CD14,CD40, CD40L, IL-1Ra, IL-1RII, IL-1α, IL-1β, IL-2, IL-3, IL-4, sIL-4R, IL-5, IL-6, IL-6R, IL-7, IL-8, IL-10, IL-12p40, IL-12p70, IL-13, IL-15, TNF, LT-α, sTNFRI, IFNγ, TRAIL, GM-CSF, TGFβ, Trx. This ELISA antibody pair also shows no cross-reactivity with recombinant mouse sTNFRI, recombinant mouse sTNFRII, recombinant mouse TNF.

Immunofluorescent Staining and Flow Cytometric Analysis: The purified hTNFR-M1 (Cat. No. 551311) antibody can be used for the immunofluorescent staining ( $\leq 1~\mu g$  antibody/10e6 cells) and flow cytometric analysis of human nucleated cells to measure their expressed levels of surface TNFRII. An appropriate immunoglobulin isotype control is clone R35-38 (Cat. No. 555846). Other formats available for immunofluorescent staining include, PE (Cat. No. 552418). Please note also that as a consequence of in vivo or in vitro activation, cell surface TNFRII can either be shed by cells or transiently expressed at higher levels. As a result, cellular activation can affect the cell's overall expressed level of surface TNFRII.

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#### **Product Notices**

- 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 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.

#### References

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