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# **Product Information**

#### Anti-Tie 2

produced in goat, affinity isolated antibody

Catalog Number T3192

# **Product Description**

Anti-Tie 2 is produced in goat using as immunogen a purified recombinant human Tie 2 extracellular domain expressed in mouse myeloma NSO cells. Affinity isolated antibody is obtained from goat anti-Tie 2 antiserum by immunospecific purification which removes essentially all goat serum proteins, including immunoglobulins, which do not specifically bind to the peptide.

Anti-Tie 2 recognizes human Tie 2 by various immunochemical techniques including immunoblotting, ELISA, immunohistochemistry, and neutralization. By immunoblotting, this antibody exhibits no cross-reactivity with recombinant human Tie 1.

Tie 1/Tie (tyrosine kinase with Ig and EGF homology domains 1) and Tie 2/Tek define a new class of the receptor tyrosine kinase (RTK) subfamily with unique structural characteristics: two immunoglobulin-like domains flanking three epidermal growth factor (EFG)-like domains followed by three fibronectin type III-like repeats in the extracellular region and a split tyrosine kinase domain in the cytoplasmic region. Human Tie 2/Fc is a 1124 amino acid residue precursor protein of approximately 100 kDa. From glycosylation, the protein migrates to ~165 kDa in SDS-PAGE under reducing conditions.

Tie 1 and Tie 2, expressed primarily on endothelial and hematopoietic progenitor cells, play important roles in angiogenesis, vasculogenesis, and hematopoiesis. In developing vascular endothelial cells, Tie 1 and Tie 2 are specifically expressed.<sup>2,3</sup> Two ligands that bind Tie have been identified, angiopoietin-1 and angiopoietin-2. Based on gene-targeting studies, the *in vivo* functions of Tie 1 are related to endothelial cell differentiation.<sup>2</sup> The receptor tyrosine kinase Tie also plays a role in the survival and integrity of the endothelium.<sup>4</sup>

# Reagent

Supplied lyophilized from a 0.2  $\mu$ m filtered solution in phosphate buffered saline with 5% trehalose.

## **Preparation Instructions**

To one vial of lyophilized powder, add 1 mL of sterile phosphate buffered saline to produce a 0.1 mg/mL stock solution of antibody.

## Storage/Stability

Prior to reconstitution, store at -20 °C. Reconstituted product may be stored at 2-8 °C for up to one month. For prolonged storage, freeze at -20 °C in working aliquots. Avoid repeated freezing and thawing. Do not store in a frost-free freezer.

#### **Product Profile**

Anti-Tie 2 has the ability to neutralize receptor-ligand interaction in a functional ELISA assay. Approximately 5-10  $\mu$ g/mL of antibody will block 50% of the binding of 40 ng/mL of recombinant human angiopoietin-2 to immobilized recombinant human Tie 2/Fc chimera (100  $\mu$ L of a 4  $\mu$ g/mL solution coated in each well).

 $\underline{Immunoblotting} \hbox{: a working antibody concentration of } 0.1\mbox{-}0.2~\mu g/mL \hbox{ is recommended. The detection limit for human Tie 2 is $\sim 1$ ng/lane under non-reducing and reducing conditions.}$ 

<u>ELISA</u>: a working antibody concentration of 0.5-1.0  $\mu$ g/mL is recommended. The detection limit for human Tie 2 is ~0.2 ng/well.

 $\frac{Immunohistochemistry}{concentration of 3-10 \ \mu g/mL is recommended using paraffin-embedded human tissue sections.}$ 

**Note**: In order to obtain the best results in various techniques and preparations, we recommend determining optimal working dilutions by titration.

### References

- Partanen, J., et al., A novel endothelial cell surface receptor tyrosine kinase with extracellular epidermal growth factor homology domains. *Mol. Cell Biol.*, 12, 1698-1707 (1992).
- 2. Sato, T.N., et al., Distinct roles of the receptor tyrosines Tie-1 and Tie-2 in blood vessel formation. *Nature*, **376**, 70-74 (1995).
- Sato, T.N., et al., Tie-1 and Tie-2 define another class of putative receptor tyrosine kinase genes expressed in early embryonic vascular system. *Proc. Natl. Acad. Sci. USA*, 90, 9355-9358 (1993).
- 4. Puri, M.C., et al., The receptor tyrosine kinase Tie is required for integrity and survival of vascular endothelial cells. *EMBO J*, **14**, 5884-5891 (1995).

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