

Product Specification Sheet

**Tyrosine Protein Kinase Receptor/Tunica internal endothelial cell kinase (TIE-1) Antibodies**

<b>Cat.</b> TIE12-S	Rabbit Anti-Human Tie-1 Antiserum # 2	<b>SIZE:</b> 100 ul
<b>Cat.</b> TIE12-A	Rabbit Anti-Human Tie-1 IgG # 2 (aff pure)	<b>SIZE:</b> 100 ug
<b>Cat.</b> TIE12-P	Human Tie-1 Control/blocking peptide # 2	<b>SIZE:</b> 100 ug
<b>Cat.</b> TIE12-C	Recombinant Human Tie-1-Fc protein for WB	<b>SIZE:</b> 100 ul

Embryonic vascular system undergoes a series of complex, highly regulated series of events involving differentiation, migration and association of primitive endothelial cells. This process is termed vasculogenesis. A further remodeling of the primitive vascular system forms the mature cardiovascular system. This process is known as angiogenesis (sprouting of new capillary vessels from pre-existing vasculature). A family of receptor tyrosine kinases **TIE1 and TIE 2** or Tek has been identified in vascular endothelium and hematopoietic cells. Mice lacking TIE 1 or TIE 2 are lethal. Ties may represent the earliest endothelial cell lineage marker and may regulate the endothelial cell proliferation, differentiation, and proper patterning during vasculogenesis. TIEs appear to be acting downstream of the VEGFRs. Tie-1 (human 1138 AA; mouse 1134 aa) is a type 1 membrane receptor protein specifically expressed in developing vascular endothelial cells. Tie-1 extracellular portion (25-279 aa) contains 3 fibronectin type III-like and 2 Ig-like C2-type, and 3-EGF-like domains.

**Source of Antigen and Antibodies**

<b>Antigen</b>	21aa peptide of Human TIE1 ; <b>Designated (TIE12-P or control peptide)</b> . conjugated to KLH; epitope location ~ N-terminus, Extracellular domain
<b>Ab Host/type</b>	Rabbit, polyclonal Unpurified antiserum (cat #TIE12-S) Aff pure IgG1 ( <b>cat #TIE12-A</b> ) purified over antigen-agarose column
<b>2-ab</b>	<b>Goat Anti-rabbit IgG-HRP</b> cat # 20320 (AP, biotin, FITC conjugates also available)
<b>-ve control</b>	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as -ve control

Recombinant human Tie-1 protein (22-760 aa, EC domain) was expressed as His-tagged Fc protein Chimera and purified (>90%). Recombinant chimeric Tie-1 protein migrates as ~130-140 kda under reducing conditions. For Western blot +ve control (**Cat # TIE12-C**) is supplied in SDS-PAGE sample buffer (reduced). Load 10 ul/lane of **TIE12-C** for good visibility with antibody Cat # **TIE12-S** or other antibodies. Store at -20oC in suitable size aliquots. SDS may crystallize in cold conditions. It should redissolve by warming before taking it from the stock. It should be heated once prior to loading on gels. If the product has been stored for several weeks, then it may be preferable to add 5 ul of fresh 2x sample buffer per 10 ul of the **TIE12-C** solution prior to heating and loading on gels. This preparation is not biologically active. It is not suitable for ELISA or other applications where native protein is required. Do not freeze, thaw, or heat repeatedly.

**Form & Storage of Antibodies/Peptide Control Antiserum (unpurified)**

100ul	solution	lyophilized powder
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Supplied 0.05% azide, **Reconstitute** powder in 100 ul PBS

**Affinity pure IgG**  
100 ug/100ul solution lyophilized powder  
Supplied in **Buffer:** PBS+0.1% BSA  
**Reconstitute powder** in PBS at 1mg/ml

**Control/blocking peptide**  
100 ug/100 ul solution lyophilized powder  
Supplied in Buffer: PBS pH 7.5,  
**Reconstitute powder in PBS at 1 mg/ml.**

**Storage**  
**Short-term:** unopened, undiluted liquid vials at -20OC and powder at 4oC or -20oC..

**Long-term:** at -20C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

**Stability:** 6-12 months at -20oC or below.  
**Shipping:** 4oC for solutions and room temp for powder

**Recommended Usage**

**Western Blotting** (1-10 ug/ml for affinity pure using Chemiluminescence technique). NIH/3T3 cells can be used as positive control. Tie-1 ~135-140 kDa. Recombinant Tie-1 fusion protein (cat # TIE12-C) migrate as 130-140 kDa due to glycosylation.

**ELISA** (1:10K-1:100K; using 50-100 ng of control peptide/well).

**Histochemistry & Immunofluorescence:** Not tested. We recommend the use of affinity purified antibody at 2-20 ug/ml in formaldehyde fixed tissue.

**Specificity & Cross-reactivity**

Human TIE12-P sequence is 90% conserved in mouse, 63% in bovine Tie-1. No significant sequence homology is seen with Tie-2. Antibody crossreactivity in various species is not established. The Control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-10 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity at the web site Human Tie-1-Fc protein control (#TIE12-C) for Western may be used for control studies.

**General References:** (1) Partenen J et al (1992) Mol. Cell. Biol. 12, 1698; Sato TN et al (1993) PNAS 90, 12056; Iwama A et al (1993) BBRC 195, 301; Korhonen J et al (1995) Blood 86, 1828; deVries C et al (1992) Science 255, 989.

*\*This product is for in vitro research use only.*

**Related materials available from ADI**

Antibodies to Ang-1, Ang-2, Tie-1, Tie-2, Recombinant Mouse and Human VEGFs, Anti-flk-1, Flt-1, and Flt-4 (VEGFRs 1-3)  
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