ALPHA DIAGNOSTIC INTERNATIONAL

Product Specification Sheet

Mouse Transferrin Receptor 1 (TfR1) Antibodies

□ Cat. # TFR16-M	**Rat** Monoclonal Anti-Mouse TfR1 IgG	**SIZE**: 100 ug
□ Cat. # TFR17-M	**Rat** Monoclonal Anti-Mouse TfR1 IgG-**Biotin** Conjugate	**SIZE**: 100 ug
□ Cat. # TFR17-C	Recombinant purified Mouse TfR1 protein control for WB	**SIZE**: 100 ul

Elemental iron is required for a variety of normal cellular functions and vital for proper growth and development. **Transferrin (Tf),** a serum glycoprotein of ~80 kDa and synthesized in the liver, is the primary protein of inter-organ transport of nonheme iron. Tf can bind two iron atoms. Tf binds to membrane **Transferrin receptors (TfRs)** and taken up by endocytosis. Iron is released from Tf, within acidic endosomes, into the cytoplasm apparently through the action of DMT1. The apoTf-TfR complex is returned to the cell surface, where, apo-Tf dissociates from TfR at the extracellular pH. The classical TfR, now termed **TfR1**, is a homodimeric (95 kDa subunits) type II membrane glycoprotein that binds two molecules of Tf. Human TfR1 (human 760 aa; mouse 763 aa) has a cytoplasmic domain 1-67aa, 68-88 aa TM, and 89-760 aa as extracellular domains. A monomeric serum form or **soluble TfR1** (~80 kDa) also exists that lacks residues 1-100 aa. Recently, a second Tf receptor, **TfR2**, has been cloned and characterized. TfR2 shares 45% identity with TfR1, and 27% with PMSA. Human TfR2 (human alpha 801 aa, Chromosome 7q22; mouse alpha 798 aa;) is predicted to contain a cytoplasmic domain of 1-80 aa, 1 TM domain followed by 105-801aa as the extracellular domain. It is highly expressed in liver and peripheral blood mononuclear cells. In contrast to Tfr1, expression of Tfr2 is not down regulated as a result of iron overload, consistent with the absence iron-responsive element in TfR2. It is alternatively spliced to **alpha and beta isoforms**. TfR2-beta protein lacked the N-terminal portion of the TfR2-alpha including the putative TM domain.

Source of Antigen and Antibodies

Purified **mouse** Tfr1 (~95 kDa) was used as immunogen to produce **monoclonal** antibodies in **rat**. A clone producing anti-mouse TfR1 (IgG2a) was used for harvesting antibodies from the culture medium. The antibody has been purified using Protein A/G column chromatography. Unconjugated IgG (**Cat # TFR16-M**) is supplied in PBS pH 7.2, containing 0.1% BSA and 0.1% azide in solution or in powder form.

Goat Anti-**rat** IgG-HRP, cat # 50320 (other conjugates also available) is recommend as 2-ab for cat # TFR16-M.

Antigen	Purified **mouse** Tfr1 (~95 kDa) protein
Ab Host/type	Rat monoclonal IgG2a **(#TFR16-M)** contains PBS/1% BSA/0.05% sodium azide
2-Ab	Goat **Anti-Rat IgG-HRP** conjugate Cat # 50320 (AP, biotin, FITC conjugates also available)
-ve control IgG	Cat # 20005-1, Rat (non-immune) Serum IgG, purified, suitable for ELISA, Western, IHC as –ve control

Purified anti-Tfr1 IgG IgG2a **(#TFR16-M)** was conjugated to **Biotin (cat # TFR17-M).** The conjugates are supplied at 0.5 mg/ml as solution in PBS pH 7.2, containing 0.1% BSA and 0.1% azide or in powder form.

Item # 20365 (Streptavidin-Peroxidase (HRP) conjugate) can be used to detect the biotinylated anti-Tfr1 IgG # TFR17-M.

#TFR17-C: Mouse TFR1 (protein accession Q62351 # ~78.3 kda, EC domain 89-763-aa (694-aa) was expressed as his-tag protein in HEK cell and purified (>95%). For Western blot +ve control (**Cat # TFR17-C**) is supplied in SDS-PAGE sample buffer (reduced). Load 10 ul/lane of **TFR17**-C for good visibility with antibody Cat # **TFR17**-M. 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 **TFR17**-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.

Storage

Short-term: unopened, undiluted vials for less than a week at 4oC.

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: **2-5 ug/ml**.
Flow Cytometry: 2-5 ug/ml using 1x10 ^{6} cells.
Histochemistry & Immunofluorescence: not tested. Suggested concn, 1-5 ug/ml.

Specificity & Cross-reactivity

TFR16-M epitope has not been determined. No significant reactivity is seen with TfR2. Antibody reactivity in other species is not established.

General References: Schneider C et al (1984) Nature 311, 675-678; McClelland A et al (1984) Cell 39, 267-274; Shih YJ et al (1990) JBC 265, 19077-19081; ; Nelson N et al (1999) EMBO J. 18, 4361-4371 (review); Cairo G and Pietrangelo A et al (2000) Biochem. J. 352, 241-250

*This product is for In vitro research use only.

Related material available from ADI
Human and mouse **Ferritin and Transferrin ELISA kits**
TFR16-17-M 131230A

Alpha Diagnostic Intl Inc., 6203 Woodlake Center Dr, San Antonio, TX 78244, USA; **Email**: service@4adi.com
☎ (800) 786-5777; (210) 561-9515; **Fax**: (210) 561-9544; **Web Site**: www.4adi.com