

Product Specification Sheet

Duodenal Cytochrome b (Dcytb) Antibodies

Cat. # DCYTB11-P	Mouse DCYTB Control Peptide # 1	SIZE: 100 ug
Cat. # DCYTB11-S	Rabbit Anti-Mouse DCYTB antiserum # 1	SIZE: 100 ul
Cat. # DCYTB11-A	Rabbit Anti-Mouse DCYTB IgG # 1 (aff pure)	SIZE: 100 ug

Iron is an essential element for a variety physiological process. Most dietary iron exists in the form of ferric iron complexes which must be reduced to yield ferrous ion before it can be taken up by protein transporters such as DCT1 (divalent cation transporter)/NRAMP2/DMT1. These transporters are especially active in small intestine where most dietary iron absorption is conducted. Ferrous Fe (II) is very unstable at physiological pH and quickly oxidized to ferric Fe (III). Therefore, highly specialized transmembrane electron transport system, maintained by ferric reductases, is required for the availability of intracellular ferrous ion. Most recently, a new gene called **Dcytb (for duodenal cytochrome b)** has been cloned and characterized in mouse duodenum that may function as mammalian ferric reductase.

Dcytb (human 286 aa, rat 290 aa) is a 6-transmembrane domains protein with ~45% sequence identity with cytochrome b561 reductase. However, no sequence homology exists with plant and yeast Dcytb. The N-terminus of mouse Dcytb is almost identical with the rabbit's p30 protein called cytochrome b558. Putative binding sites for cytochrome b561 substrates (ascorbic acid and semidehydroascorbic acid) are partially conserved in Dcytb and b561. Unlike most proteins involved in iron metabolism, Dcytb gene lacks a definable iron-responsive element. Dcytb is highly expressed in the brush border membrane of duodenal enterocytes.

Source of Antigen and Antibodies

Antigen	A 20 AA Peptide (designated DCYTB11-P; control peptide) of mouse DCYTB conjugated to KLH; epitope location ~ C-terminus
Ab Host/type	Rabbit, Polyclonal unpurified antiserum (#DCYTB11-S) and IgG, purified over antigen-agarose (Cat # DCYTB11-A)
2-Ab	Cat # 20320, goat anti-rabbit IgG-HRP (AP, biotin, FITC conjugates also available).
-ve control IgG	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as -ve control

Form & Storage of Antibodies/Peptide Control

Antiserum (unpurified)

100ul solution lyophilized powder
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 (1:1K-5K for neat serum and 1-10 ug/ml for affinity pure antibody using ECL technique). Dcytb is approx. 28 kDa protein in the duodenum (1).

ELISA: Control peptide can be used to coat ELISA plates at 1 ug/ml and detected with antibodies (1:10-50K for neat serum and 0.5-1 ug/ml for affinity pure).

Histochemistry & Immunofluorescence: Not tested.

Specificity & Cross-reactivity

The 20 AA mouse DCYTB11-P control peptide sequence is 70% conserved in human Dcytb. No significant sequence homology exists with b558 or other reductases. Antibody cross-reactivity in various species has not been established. 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 (see detailed protocol at the web site.).

General References: McKie AT et al (2001) Science 291, 1755-1759; Escriou V et al (1994) JBC 269, 14007; Royer-pokora B (1986) Nature 322, 32-38; Dinauer MC (1987) Nature 327, 717-720

(2) Citations of ADI's Antibodies (see web site for updated list)

Theurl I, 2005, Pathways for the regulation of body iron homeostasis in response to experimental iron overload, J. Hepatology, In Press, IHC, mouse tissues, no changes

*This product is for In vitro research use only.

Related material available from ADI

Antibodies NRAMP1/2, MTP1, IRP1/2, HFE, Transferrin, and receptor, Ferritin (H/L chains)

DCYTB11-S-A-P 71223A

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