

Certificate of Analysis

Product: Affinity Purified Anti-ECSIT (Rabbit)

Code: 600-401-950

Lot #: 19836

Size: 100 µg

Physical State: Liquid (sterile filtered)

Antibody Concentration: 1.0 mg/ml (by UV absorbance at 280 nm)

Buffer: 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2

Stabilizer: None

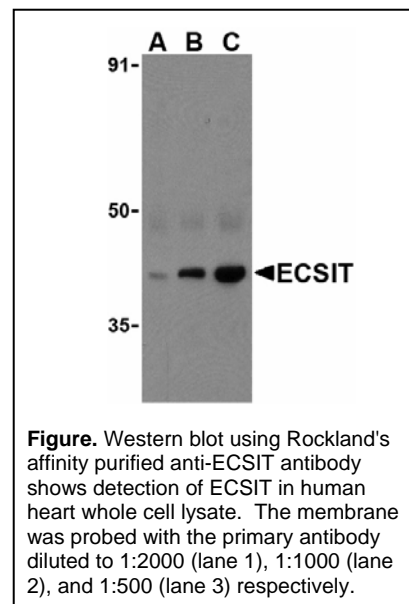
Preservative: 0.02% (w/v) Sodium Azide

Storage Conditions: Store vial at -20° C prior to opening. Dilute only prior to immediate use. For extended storage aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Expiration date is one (1) year from date of opening.

Background Information: Activation of NF-κB as a result of Toll-like receptor (TLR) and IL-1 receptor signaling is a major component of innate immune responses (reviewed in 1). Signals from these receptors are relayed by a number of adapter molecules such as TRIF, TIRAP, and MyD88 (2) to kinases such as IRAK (3) and other intermediates such as TNF receptor associated factor (TRAF)-6 (4). ECSIT (evolutionarily conserved signaling intermediate in Toll pathways) was initially identified as a cytoplasmic protein interacting specifically with TNF receptor associated factor (TRAF)-6 in the TLR pathway (5). Recently however, ECSIT has also been shown to be required for bone morphogenetic protein (Bmp) signaling and mesoderm formation during mouse embryogenesis (6), indicating the possibility of cross-talk between the TLR/IL-B and Bmp signaling pathways.

Recommended Dilutions:

| | |
|-----------------------------|--------------------|
| ELISA | 1:5,000 - 1:20,000 |
| WESTERN BLOT | 1:500 - 1:2,000 |
| IMMUNOHISTOCHEMISTRY | User Optimized |
| OTHER APPLICATIONS | User Optimized |



Application Note(s): This affinity-purified antibody has been tested for use in ELISA, immunohistochemistry and western blot. Specific conditions for reactivity should be optimized by the end user. Expect a band approximately 45 kDa in size corresponding to ECSIT protein by western blotting in the appropriate cell lysate or extract.

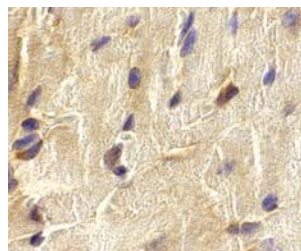


Figure. Immunohistochemistry using Rockland's affinity purified anti-ECSIT antibody shows staining of ECSIT in mouse heart tissue. Paraffin embedded formalin fixed tissue was reacted with primary antibody diluted 1:500.

Purity and Specificity: This affinity-purified antibody is directed against human ECSIT protein. The product was purified from monospecific antiserum by immunoaffinity chromatography. A BLAST analysis was used to suggest cross reactivity with ECSIT protein from human and mouse sources based on 100% homology with the immunizing sequence. Reactivity against homologues from other sources is not known.

Immunogen: This affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to a region near the carboxy terminus of human ECSIT protein.

Relevant Links: NCBI [NP_057665](#)

Swis-Prot [Q9NY11](#)

Predicted Molecular Weight: Human ECSIT, MW 49.1 kDa (431 aa)

General References:

1. Takeda K, Kaisho T, and Akira S. Toll-like receptors. *Annu. Rev. Immunol.* 2003; **21**:335-76.
2. Vogel SN, Fitzgerald KA, and Fenton MJ. TLRs: differential adapter utilization by toll-like receptors mediates TLR-specific patterns of gene expression. *Mol. Interv.* 2003; **3**:466-77.
3. Janssens S and Beyaert R. Functional diversity and regulation of different interleukin-1 receptor-associated kinase (IRAK) family members. *Mol. Cell.* 2003; **11**:293-302.
4. Sato S, Sugiyama M, Yamamoto M, et al. Toll/IL-1 receptor domain-containing adaptor inducing IFN-beta (TRIF) associates with TNF receptor-associated factor 6 and TANK-binding kinase 1, and activates two distinct transcription factors, NF-kappa B and IFN-regulatory factor-3, in the Toll-like receptor signaling. *J. Immunol.* 2003; **171**(8):4304-10.
5. Kopp E, Medzhitov R, Carothers J, et al. ECSIT is an evolutionarily conserved intermediate in the Toll/IL-1 signal transduction pathway. *Genes Dev.* 1999; **13**:2059-71.
6. Xiao C, Shin JH, Kluppel M, et al. Ecsit is required for Bmp signaling and mesoderm formation during mouse embryogenesis. *Genes Dev.* 2003; **17**:2933-49.

Related Products:

| | |
|------------------------------|--|
| #200-401-949 | IgG Fraction of Anti-ADAM10 (Rabbit) |
| #600-401-950 | Affinity Purified Anti-ECSIT (Rabbit) |
| #200-401-951 | IgG Fraction of Anti-IRAK (Rabbit) |
| #600-401-952 | Affinity Purified Anti-IRAK-2 (Rabbit) |
| #600-401-953 | Affinity Purified Anti-IRAK-4 (Rabbit) |
| #600-401-954 | Affinity Purified Anti-IRAK-M (Rabbit) |
| #600-401-955 | Affinity Purified Anti-MyD88 (Rabbit) |
| #600-401-956 | Affinity Purified Anti-TLR2 (Rabbit) |
| #600-401-957 | Affinity Purified Anti-TLR4 (Rabbit) |
| #600-401-958 | Affinity Purified Anti-TLR8 (Rabbit) |
| #611-703-127 | Peroxidase Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (DONKEY) MX10 |
| #611-132-122 | IRDye800 Conjugated Affinity Purified Anti-RABBIT IgG (H&L) (GOAT) MX10 |
| #B501-0500 | 500 g BLOTTO |
| #BSA-30 | 500 ml 30% BOVINE SERUM ALBUMIN SOL'N in 0.85% sodium chloride (no preservative or stabilizer) |
| #B304 | 10 ml NORMAL GOAT SERUM (NGS) |
| #KIA-003 | MaxTag TM Anti-RABBIT IgG Kit for Immunoblotting |
| #MB-070 | Blocking Buffer for Fluorescent Western Blotting |

Note: This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information.

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