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Antibodies to Thymocyte-expressed molecule involved in selection (THEMIS)

Thymocyte-expressed molecule involved in selection (THEMIS). Catalog # THMS-101 AP, FITC-THMS, P-THMS, PC-THMS.

Accession # *Q8N1K5* and Alternate Nomenclature: *TSEPA; C60rf190; C60rf207; thymocyte selection associated*

Thymocyte-expressed molecule involved in selection (THEMIS) is a protein coding gene that plays a regulatory role in positive and negative T-cell selection during late thymocyte development (1). THEMIS is located on human chromosome 6q22.33. It is a member of a new metazoan gene family and is required for completion of thymocyte positive selection (2). THEMIS contains a tandem repeat of a unique globular domain which includes cysteine motif that defines a family consisting of five uncharacterized vertebrate proteins that have orthologs in most animal species (2). THEMIS appears to be related to TCR signaling and, because it is quickly phosphorylated after TCR stimulation, it is necessary for optimal TCR-driven calcium mobilization as well as being necessary for activation of the Erk-Kinase (3).

Recently it was reported that THEMIS is essential for the transition of CD4⁺ CD8⁺ double-positive (DP) cells to MHC class I-restricted CD4⁺ CD8⁺ and MHC class II-restricted CD4⁺ CD8⁺ single positive (SP) cells (4). In agreement with these findings THEMIS deficient thymocytes showed defective positive selection which caused fewer thymocytes to reach maturation (5). In these mice, CD4SP thymocyte maturation was more severely affected than CD8SP due to the fact that sustained signaling is essential for CD4SP thymocyte development (6). A large percentage of THEMIS-deficient cells had a CD4(+)/CD25(+)/Foxp3(-) regulatory and CD62L(10)/CD44(hi) memory phenotypes than compared to wild type of T cells (3). THEMIS is expressed in thymocytes between the Pre-T cell antigen receptor (pre-TCR) and positive selection checkpoints. THEMIS is approximately a 77kDa protein (641 amino acids).

The THEMIS antibodies were generated against purified THEMIS protein. THEMIS are affinity purified over immobilized antigen based chromatography, and the purified immunoglobulins are stabilized in antibody stabilization buffer. FabGennix Int. Inc. will also provide limited quantities of antigenic blocking protein for THEMIS antibody. Antibodies to THEMIS (THMS-101AP) will label ~77kDa protein in Western blot positive control samples for THEMIS and several other tissues. FabGennix Inc. will also conjugate antibodies with secondary enzymes (alk-Pase or HRP) or fluorescent probes upon request at nominal cost.

Catalog #	Host Species	Nature	Cross reactivity	Quantity	Volume
THMS-101 AP	Rabbit	Affinity purified THEMIS antibodies	H, others n.d	100 ug	200ul
FITC- THMS	Rabbit	FITC-conjugated THEMIS antibody	H, others n.d	100ug	200ul
P-THMS	n/a	Antigenic blocking peptide for THMS -101AP	n/a	250 ug	100ul
PC- THMS	n/a	Western blotting positive control for THEMIS	n/a	For 5 appl	Inquire

R = rat; M = mouse; H = human; C = chicken; monk = monkey; * not all variants are labeled equally

- Immunogen:** Synthetic peptides corresponding to unique epitope on THEMIS. The THEMIS peptide was covalently modified post-synthetically covalently modified to achieve desired antigenicity.
- Concentration:** THMS -101AP: IgG concentration 0.64-0.72 mg/ml in antibody stabilization buffer.
- Applications:** Antibody THMS -101AP is ideal for WB and ELISA applications, other applications have not been tested. The species cross reactivity for these antibodies have not been examined fully. The dilutions for this antibody is for reference only, investigators are expected to determine the optimal conditions for specific assay. WB: > 1:500; IMM & i.p pull-down assays: n.d; IHC n.d. ELISA <1:10,000. Application of this antibody in protocols not listed here does not necessarily exclude its use in such procedures.
- Reactivity:** This antibody detects a single band of approximately 77kDa in PC- THEMIS samples.
- Protocols:** Standard protocol for various applications (WB; IMM and IHC) of this antibody is provided with the product specification sheet, however, FabGennix Inc. strongly recommends investigators to optimize conditions for use of this antibody in their laboratories.
- Form/Storage:** The antiserum is supplied in antibody stabilization buffer. The affinity-purified antibodies are isolated on immobilized antigen-affinity column and supplied as stabilized product. Store at -20°C for long-term storage. FabGennix Inc. does not recommend storage of very dilute antibody solutions unless they are prepared in specially formulated multi use antibody dilution buffer (Cat # DiluOBuffer). Working solutions of antibodies in DiluOBuffer should be filtered through 0.45um filter after every use for long-term storage.

Western Blot of THEMIS. The THEMIS antibody (Cat # THMS-101AP) at 1:500 in diluOBuffer was probed with PC-THMS sample. MW of THEMIS is approximately 77kDa

Notes: Briefly centrifuge to collect liquid, heat or boil PC-THEMIS tube for 1-2 minutes to dissolve any precipitate before use. This product is "ready-to-use" for electrophoresis. After thawing store at room temperature, Repeated freezing and thawing may result in appearance of higher MW immunoreactive bands.

New Reagents: Now you can recycle your western blots (nitrocellulose, supported membranes and PVDF membranes) by using our StripOBuffer (Cat FGI-1989). Each stripping is guaranteed to give better signal (up to 8 stripping). No strong pungent smell of reducing agents or heating required. Block in 5X diluOBuffer and you are ready for blotting with a new antibody

References:

1. Fu G, et al. Themis controls thymocyte selection through regulation of T cell antigen receptor-mediated signaling. *Nat Immunol.* August 2009; 10(8):848-856.
2. Johnson AL, et al. Themis is a member of a new metazoan gene family and is required for the completion of thymocyte positive selection. *Nat Immunol.* August 2009; 10(8):831-839.
3. Lesourne R, et al. Themis, a T cell-specific protein important for late thymocyte development. *Nat Immunol.* August 2009; 10(8):840-847.
4. Kakugawa K, et al. A novel gene essential for the development of single positive thymocytes. *Mol Cell Biol.* September 2009; 29(18):5128-5135.
5. Saito T, Watanabe N. Positive and negative thymocyte selection. *Crit Rev Immunol.* 1998; 18(4):359-370.
6. Yasutomo K, et al. The duration of antigen receptor signaling determines CD4+ versus CD8+ T-cell lineage fate. *Nature.* 2000; 404:506-510.

* For users who may require large amounts of THEMIS-101AP, please enquire about bulk material discounts.
This Product is for Research Use Only and is NOT intended for use in humans or clinical diagnosis.

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