

Anti-SIT Purified SIT1 Monoclonal Antibody

Catalog Number: M05696

About SIT1

SIT (SHP2-interacting transmembrane adaptor protein) is expressed exclusively in lymphoid organs and acts either as a positive or as a negative regulatory element in T cell activation and in T cell development. Binding to Grb2 plays a pivotal role in signal transduction. Hubener et al. (2001) determined that the SIT gene contains 5 exons and spans 1.8 kb of genomic DNA. The SIT promoter demonstrated strong transcriptional activity and potential binding sites for both ubiquitous and lymphoid-specific transcription factors.

Overview

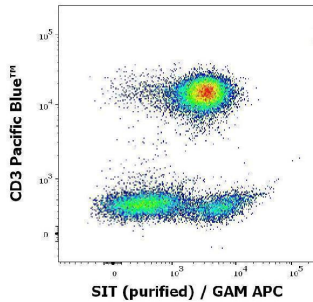
Product Name	Anti-SIT Purified SIT1 Monoclonal Antibody
Reactive Species	Human
Description	Boster Bio Anti-SIT Purified SIT1 Monoclonal Antibody (Catalog# M05696). Tested in Flow Cytometry, IP, WB application(s). This antibody reacts with Human.
Application	Flow Cytometry, IP, WB
Clonality	Monoclonal SIT-01
Formulation	Phosphate buffered saline (PBS), pH 7.4, 15 mM sodium azide
Storage Instructions	Store at 2-8°C. Do not freeze.
Host	Mouse
Uniprot ID	Q9Y3P8

Technical Details

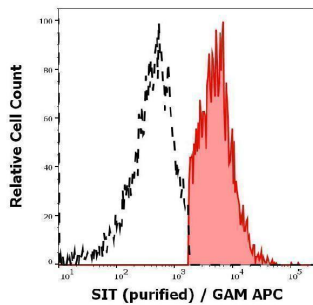
Immunogen	Bacterially produced recombinant intracellular fragment of human SIT. The antibody SIT-01 reacts with an intracellular epitope of SHP2-interacting transmembrane adaptor protein (SIT) expressed exclusively in lymphoid organs.
Predicted Reactive Species	Primate
Cross Reactivity	This antibody weakly cross-reacts with murine SIT.
Isotype	Mouse IgG1
Form	Liquid
Concentration	1 mg/ml
Purification	Purified by protein-A affinity chromatography.
Suggested Dilutions	Flow cytometry: 1-5 ug/ml, intracellular staining.

Western blotting: 1-2 ug/ml, reducing conditions.

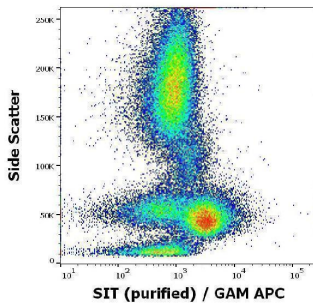
Anti-SIT Purified SIT1 Monoclonal Antibody (M05696) Images



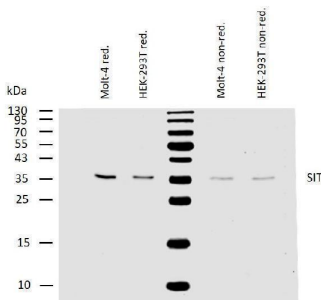
Flow cytometry multicolor intracellular staining of human peripheral whole blood stained using anti-SIT (SIT-01) purified antibody (concentration in sample 9 µg/ml, GAM APC) and anti-human CD3 (UCHT1) Pacific Blue™ antibody (20 µl reagent / 100 µl of peripheral whole blood).



Separation of human CD3 negative SIT positive lymphocytes (red-filled) from CD3 negative SIT negative lymphocytes (black-dashed) in flow cytometry analysis (intracellular staining) of peripheral whole blood stained using anti-SIT (SIT-01) purified antibody (concentration in sample 9 µg/ml, GAM APC).

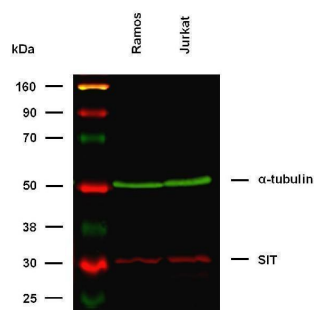


Flow cytometry intracellular staining pattern of human peripheral whole blood using anti-SIT (SIT-01) purified antibody (concentration in sample 9 µg/ml, GAM APC).

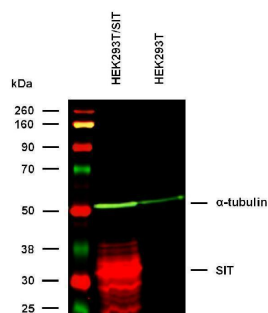


Western blotting analysis of human SIT using mouse monoclonal antibody SIT-01 on lysates of Molt-4 and HEK-293T cells under reducing and non-reducing conditions. Nitrocellulose membrane was probed with 2 µg/ml of mouse anti-SIT monoclonal antibody followed by IRDye800-conjugated anti-mouse secondary antibody. SIT was detected around 36 kDa.

Anti-SIT Purified (clone SIT-01) works in WB application. Western blotting analysis was performed on whole cell extracts (RIPA lysis buffer) of Ramos and Jurkat cell lines, mixed and heated (100°C, 5 min) with reducing (2-mercaptoethanol) SDS-loading buffer. Samples were resolved using 10% SDS-PAGE gel. Nitrocellulose membrane blot was probed simultaneously with mouse IgG1



monoclonal antibody SIT-01 (2 µg/ml), and rat IgG2a anti-tubulin monoclonal antibody YOL1/34 (1 µg/ml) used as the loading control. Subclass-specific secondary antibodies IRDye 800CW Goat-anti-Rat IgG (green) and IRDye 680LT Goat-anti-Mouse IgG (red) were used for multiplex fluorescent Western blot detection. SIT was detected at ~32 kDa in tested cell lines.



Anti-SIT Purified (clone SIT-01) specificity verification by WB. The specificity of SIT-01 antibody was assessed by comparing binding signals in HEK293T cells overexpressing the target SIT protein to wild type cells (control) with low level of endogenous protein expression. Western blotting analysis was performed on whole cell extracts (urea lysis buffer) of transfected and control cells, mixed and heated (100°C, 5 min) with reducing (2-mercaptoethanol) SDS-loading buffer. Samples were resolved using 10% SDS-PAGE gel. Nitrocellulose membrane blot was probed simultaneously with mouse IgG1 monoclonal antibody SIT-01 (2 µg/ml), and rat IgG2a anti-tubulin monoclonal antibody YOL1/34 (1 µg/ml) used as the loading control. Subclass-specific secondary antibodies IRDye 800CW Goat-anti-Rat IgG (green) and IRDye 680LT Goat-anti-Mouse IgG (red) were used for multiplex fluorescent Western blot detection.

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Anti-SIT Purified SIT1 Monoclonal Antibody

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