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ProductInformation

ANTI-TRANCE (RANKL), MOUSE
Developed in Goat
Affinity Isolated Antibody

Product Number **T2692**

Product Description

Anti-TRANCE (RANKL) is developed in goat using purified recombinant mouse TRANCE, expressed in mouse NSO cells as immunogen. TRANCE specific IgG is purified from goat serum using mouse TRANCE affinity chromatography.

Anti-TRANCE (RANKL) recognizes recombinant mouse TRANCE (TNF-related activation-induced cytokine) by immunoblotting, ELISA, and neutralization. In immunoblotting, this antibody demonstrates no cross-reactivity with recombinant human CD40 ligand.

TRANCE, also called RANKL (RANK ligand), OPGL (Osteoprotegrin ligand), and ODF (Osteoclast differentiation factor), is a type II transmembrane signaling protein of the TNF superfamily. The extra-cellular domain contains two potential N-linked glycosylation sites. Mouse TRANCE cDNA encodes a 316 amino acid protein with a calculated molecular mass of approximately 28 kDa.[1] As a result of glycosylation, recombinant mouse TRANCE migrates as an approximately 36 kDa protein in SDS-PAGE.[1] Human and mouse TRANCE share approximately 85% amino acid sequence identity.

TRANCE was originally identified as an immediate early gene upregulated by T cell receptor stimulation. It is a key regulator of the immune system and of bone development and homeostasis.[2] Multi-functions of TRANCE include induction of activation of c-jun N-terminal kinase in T cells,[1] enhancement of T cell growth[3-5] and dendritic cell function,[6, 7] induction of osteoclastogenesis,[3, 4,8,9] and lymph node organo-genesis.[3, 4] The cell surface signaling receptor of TRANCE (RANKL) is RANK which undergoes receptor clustering during signal transduction.

TRANCE is highly expressed in thymus and lymph nodes but not in nonlymphoid tissues.[1] It is abundantly expressed in T cells but not in B cells.[1] TRANCE activates mature dendritic cells, inducing cytokine production, suggesting that it is a factor in the T cell-dentritic cell interaction during an immune response.[6] TRANCE expression is found on osteoblasts and regulates osteoclast differentiation function.

Reagents

Anti-TRANCE is supplied as approximately 100 µg of antiserum lyophilized from a 0.2 µm filtered solution in phosphate buffered saline.

Preparation Instructions

To one vial of lyophilized powder, add 1 ml of 0.2 µm-filtered solution of phosphate-buffered saline (PBS) to produce a 0.1 mg/ml stock solution of antibody. If aseptic technique is used, no further filtration should be needed for use in cell culture environments.

Storage/Stability

Prior to reconstitution, store at –20 °C. The reconstituted product may be stored at 2-8 °C for at least one month. For prolonged storage, freeze in working aliquots at –20 °C. Avoid repeated freezing and thawing.

Product Profile

To measure the ability of this antibody to neutralize the bioactivity of murine TRANCE, recombinant mouse TRANCE (30 ng/ml) is incubated with the antibody at concentrations of 0.001-1 µg/ml for 1 hour at 37 °C on a 96-well plate. At the end of this preincubation period, mouse splenocytes (1.5×10^6 cells/ml) and recombinant mouse M-CSF (20 ng/ml) are added. The mixture in a total volume of 100 µl/well is incubated at 37 °C for 6 days in a humidified CO_2 chamber. At the end of incubation, osteoclast differentiation is evaluated by TRAP (tartrate-resistant acid phosphatase) solution assay.

The ND_{50} of anti-mouse TRANCE is approximately 0.02-0.06 µg/ml in the presence of 30 ng/ml of recombinant mouse TRANCE, using mouse splenocytes.

The ND_{50} is the concentration of antibody required to yield one-half maximal inhibition of the cytokine activity on a responsive cell line, when the cytokine is present at a concentration just high enough to elicit a maximum response.

For ELISAs, a working concentration of 0.5-1.0 µg/ml detects approximately 0.3 ng/well of recombinant mouse TRANCE.

For immunoblotting, a working concentration of 0.1-0.2 µg/ml detects mouse TRANCE at approximately 20 ng/lane under non-reducing and reducing conditions.

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working dilutions by titration test.

Endotoxin: <10 ng/mg antibody determined by the LAL method.

References

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