

# CD269 (BCMA) antibodies, mouse

For research use only

9 µg equal 60 tests, 30 µg equal 200 tests. One test corresponds to labeling of 10<sup>6</sup> cells.

Product	Content	Order no.
CD269 (BCMA)-VioBright FITC	9 µg in 300 µL	130-108-327
CD269 (BCMA)-VioBright FITC	30 µg in 1 mL	130-108-304
CD269 (BCMA)-PE	9 µg in 300 µL	130-108-323
CD269 (BCMA)-PE	30 µg in 1 mL	130-108-300
CD269 (BCMA)-APC	9 µg in 300 µL	130-108-324
CD269 (BCMA)-APC	30 µg in 1 mL	130-108-301
CD269 (BCMA)-PE-Vio770	9 µg in 300 µL	130-108-325
CD269 (BCMA)-PE-Vio770	30 µg in 1 mL	130-108-302
CD269 (BCMA)-APC-Vio770	9 µg in 300 µL	130-108-326
CD269 (BCMA)-APC-Vio770	30 µg in 1 mL	130-108-303
CD269 (BCMA)-Biotin	9 µg in 300 µL	130-108-322
CD269 (BCMA)-Biotin	30 µg in 1 mL	130-108-299

## Warnings

Reagents contain sodium azide. Under acidic conditions sodium azide yields hydrazoic acid, which is extremely toxic. Azide compounds should be diluted with running water before discarding. These precautions are recommended to avoid deposits in plumbing where explosive conditions may develop.

## Technical data and background information

<b>Antigen</b>	CD269 (BCMA)
<b>Clone</b>	REA550
<b>Isotype</b>	recombinant human IgG1
<b>Isotype control</b>	REA Control antibodies
<b>Alternative names of antigen</b>	TNFRSF17, Bcm, BCMA, Tnfrsf13, Tnfrsf13a
<b>Molecular mass of antigen [kDa]</b>	20
<b>Distribution of antigen</b>	B cells, lymphocytes
<b>Product format</b>	Reagents are supplied in buffer containing stabilizer and 0.05% sodium azide.
<b>Fixation</b>	Cells should be stained prior to fixation, if formaldehyde is used as a fixative.
<b>Storage</b>	Store protected from light at 2–8 °C. Do not freeze.

Clone REA550 recognizes the mouse CD269 antigen, which is also known as B cell maturation protein BCMA. CD269 is a member of the TNF receptor superfamily. Its binding to BLyS and APRIL activates

transcription factor NF- $\kappa$ B and stimulates IgM production by peripheral blood B cells. A soluble form of BCMA, which inhibits the proliferative activity of APRIL *in vitro*, decreases tumor cell proliferation in nude mice. CD269 has been detected in immune organs such as spleen, thymus, bone marrow, and heart, as well as at lower levels in kidney and lung and is preferentially expressed in mature B lymphocytes. Additional information: Clone REA550 displays negligible binding to Fc receptors.

## Reagent requirements

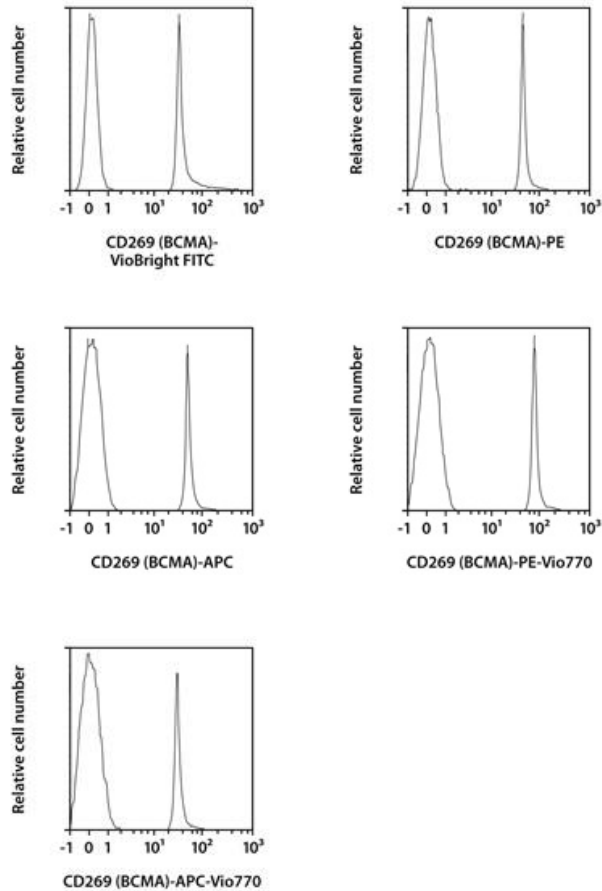
- Buffer: Prepare a solution containing phosphate-buffered saline (PBS), pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS<sup>®</sup> BSA Stock Solution (# 130-091-376) 1:20 with autoMACS<sup>®</sup> Rinsing Solution (# 130-091-222). Keep buffer cold (2–8 °C).  
Note: EDTA can be replaced by other supplements such as anticoagulant citrate dextrose formula-A (ACD-A) or citrate phosphate dextrose (CPD). Buffers or media containing Ca<sup>2+</sup> or Mg<sup>2+</sup> are not recommended for use.
- (Optional) Fluorochrome-conjugated anti-biotin antibodies, e.g., Anti-Biotin-PE (# 130-090-756) as secondary antibody reagent in combination with biotinylated antibodies.
- (Optional) Propidium Iodide Solution (# 130-093-233) for flow cytometric exclusion of dead cells without fixation.
- (Optional) Fixation and Dead Cell Discrimination Kit (# 130-091-163) for cell fixation and flow cytometric exclusion of dead cells.

## Protocol for cell surface staining

- The recommended antibody dilution for labeling of cells and subsequent analysis by flow cytometry is 1:10 for up to 10<sup>6</sup> cells/50  $\mu$ L of buffer.
  - Volumes given below are for up to 10<sup>6</sup> nucleated cells. When working with fewer than 10<sup>6</sup> cells, use the same volumes as indicated. When working with higher cell numbers, scale up all reagent volumes and total volumes accordingly (e.g. for 2 $\times$ 10<sup>6</sup> nucleated cells, use twice the volume of all indicated reagent volumes and total volumes).
1. Determine cell number.
  2. Centrifuge cell suspension at 300 $\times$ g for 10 minutes. Aspirate supernatant completely.
  3. Resuspend up to 10<sup>6</sup> nucleated cells per 45  $\mu$ L of buffer.
  4. Add 5  $\mu$ L of the antibody.
  5. Mix well and incubate for 10 minutes in the dark in the refrigerator (2–8 °C).  
Note: Higher temperatures and/or longer incubation times may lead to non-specific cell labeling. Working on ice requires increased incubation times.
  6. Wash cells by adding 1–2 mL of buffer and centrifuge at 300 $\times$ g for 10 minutes. Aspirate supernatant completely.
  7. (Optional) If biotinylated antibody was used, resuspend the cell pellet in 100  $\mu$ L of buffer, add 10  $\mu$ L of fluorochrome-conjugated anti-biotin antibody, and continue as described in steps 5 and 6.
  8. Resuspend cell pellet in a suitable amount of buffer for analysis by flow cytometry or fluorescence microscopy.

## Examples of immunofluorescent staining

Latex beads were coated with recombinant mouse CD269 (BCMA) protein and then stained with CD269 (BCMA) antibodies or with the corresponding REA Control antibodies (left peak). Flow cytometry was performed using the MACSQuant<sup>®</sup> Analyzer.



## References

1. **Madry, C. et al.** (1998) The characterization of murine BCMA gene defines it as a new member of the tumor necrosis factor receptor superfamily. *Int. Immunol.* 10(11): 1693–1702.
2. **Rennert, P. et al.** (2000) A soluble form of B cell maturation antigen, a receptor for the tumor necrosis factor family member april, inhibits tumor cell growth. *J. Exp. Med.* 192: 1677–1684.
3. **Marsters, S. A. et al.** (2000) Interaction of the TNF homologues BLyS and APRIL with the TNF receptor homologues BCMA and TACI. *Curr. Biol.* 10(13): 785–788.

## Warranty

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