

## Product Information

<b>Description:</b>	This anti-BTLA antibody is a purified recombinant human monoclonal antibody which recognizes the human BTLA protein. BTLA (B- and T-lymphocyte attenuator) belongs to the CD28 immunoglobulin superfamily (IgSF) and is an immune-regulatory receptor that binds to HVEM (Herpesvirus entry mediator, also known as TNFRSF14, Tumor necrosis factor receptor superfamily member 14). This antibody has been tested for specific binding to purified human BTLA protein and neutralizes the interaction between BTLA and HVEM.
<b>Concentration:</b>	1.34 mg/ml
<b>Species:</b>	Human
<b>Isotype:</b>	IgG1
<b>Formulated In:</b>	8 mM phosphate, 110 mM NaCl, 2.2 mM KCl, pH 7.4, and 20% glycerol
<b>Expression System:</b>	HEK293
<b>Clonality:</b>	Monoclonal
<b>Purification:</b>	Protein A affinity chromatography from HEK293 supernatants
<b>Cross Reactivity:</b>	This antibody recognizes human BTLA. It has not been tested with other species.
<b>Format:</b>	Aqueous buffer solution
<b>Stability:</b>	At least 12 months at -80°C. Avoid freeze/thaw cycles.
<b>Storage:</b>	-80°C
<b>MW:</b>	150 kDa
<b>Purity:</b>	≥90%
<b>Assay Conditions:</b>	To determine the binding of this antibody to BTLA, we performed a binding assay described in the following steps:

1. Purified anti-BTLA antibody was coated onto a clear 96-well plate overnight at 4°C (1 µg/ml in PBS).
2. The next day, each well was washed with 100 µl of BPS Immuno Buffer 1 (BPS Bioscience #79311) three times. The plate was tapped upside down onto paper towels to remove excess buffer.
3. Each well was blocked with 100 µl of Blocking Buffer 2 (BPS Bioscience #79728) for 1 hour at room temperature (with slow shaking).
4. Serial dilutions of the antibody (150 nM to 0 nM in 3-fold dilutions) were incubated in each well for 1 hour at room temperature (with slow shaking).
5. Next, wells were washed three times as in step #2 and incubated with HRP-Streptavidin (1:1000, BPS Bioscience #79742) diluted in Blocking Buffer 2 for 30 minutes at room temp (with slow shaking).
6. Plate was washed again three times, then tapped to dry.

7. For detection, the wells were incubated with Colorimetric HRP Substrate (BPS Bioscience #79651) for 1-10 minutes until a blue color developed in the positive control.
8. The reaction was immediately quenched with an equal volume of 1N HCL and the absorbance was measured at 450 nm.

To determine the ability of this antibody to neutralize the binding of BTLA and HVEM, we performed the BPS Bioscience BTLA:HVEM[Biotinylated] Inhibitor Screening Assay Kit (BPS Bioscience, #72008). In short:

1. Purified BTLA (BPS Bioscience, #71141) was coated onto an opaque 96-well plate overnight at 4°C (2 ng/μl in PBS).
2. The next day, each well was washed with 100 μl of BPS Immuno Buffer 1 (BPS Bioscience #79311) three times. We then tapped upside down on paper towels to remove excess buffer.
3. Each well was washed with 100 μl of Blocking Buffer 2 (BPS Bioscience #79728) for 1 hour at room temperature (with slow shaking).
4. Serial dilutions of the neutralizing antibody (75 nM to 0 nM in 2-fold dilutions) were incubated in each well for 1 hour at room temperature (with slow shaking).
5. An equal volume of biotin labeled-HVEM (BPS Bioscience, #71143) diluted to 1 ng/μl in 1x BPS Immuno Buffer 1 was added to each well. The plate was incubated at room temperature for two hours with slow shaking.
6. Next, wells were washed three times as in step #2 and each well was blocked for 10 minutes at room temperature with 100 μl of Blocking Buffer 2.
7. HRP-Streptavidin (1:1000, BPS Bioscience #79742) was diluted in Blocking Buffer 2 and added to each well. The plate was incubated for 1 hour at room temp (with slow shaking).
8. Next, wells were washed three times as in step #2 and each well was blocked for 10 minutes at room temperature with 100 μl of Blocking Buffer 2.
9. Just before use, equal volumes of HRP Chemiluminescent Substrate A and HRP Chemiluminescent Substrate B were combined and 100 μl was added to each well.
10. The plate was immediately read on a luminometer.

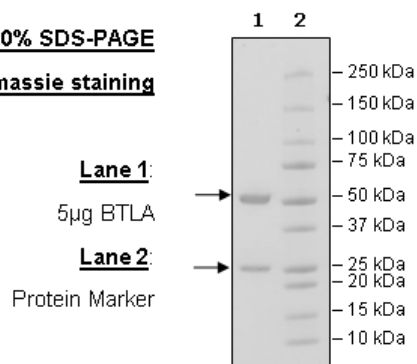
**Applications:**

This product is for research use only. It is not suitable for human, diagnostic, or therapeutic use.

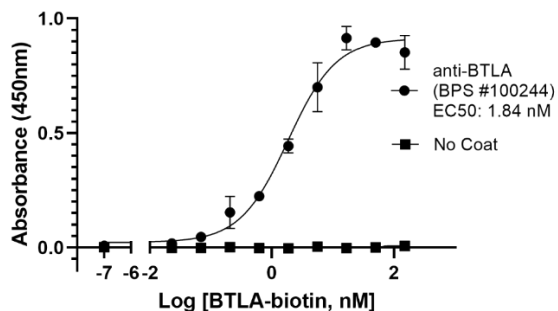
## Quality Control Data

### 4-20% SDS-Page Coomassie Staining

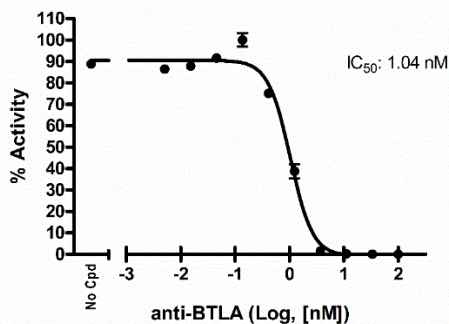
**4-20% SDS-PAGE**  
**Coomassie staining**



### anti-BTLA vs. BTLA-biotin Binding Assay



### BTLA:HVEM-biotin anti-BTLA Neutralization Assay



The graphs above represent the binding assay and neutralization assay described above in "Assay Conditions". Data shown is representative. For lot-specific information, please contact BPS Bioscience, Inc. at [support@bpsbioscience.com](mailto:support@bpsbioscience.com).