**PD-L1**
Concentrated and Prediluted Rabbit Monoclonal Antibody
901-3171-021517

**Catalog Number:**
- ACI 3171 A, C
- API 3171 AA

**Description:**
- 0.1, 1.0 ml, concentrated
- 6.0 ml, prediluted

**Dilution:**
- 1:100
- Ready-to-use

**Diluent:**
- Renoir Red
- N/A

**Intended Use:**
For In Vitro Diagnostic Use

PD-L1 [CAL10] is a rabbit monoclonal antibody that is intended for laboratory use in the qualitative identification of PD-L1 protein by immunohistochemistry (IHC) in formalin-fixed paraffin-embedded (FFPE) human tissues. The clinical interpretation of any staining or its absence should be complemented by morphological studies using proper controls and should be evaluated within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist.

**Summary and Explanation:**
Programmed death ligand 1 (PD-L1, also known as CD274) inhibits tumor-reactive T cells via binding to the programmed death-1 (PD-1) receptor, rendering tumor cells resistant to CD8+ T cell-mediated lysis (1). Studies have shown that the inhibitory receptor PD-1 is expressed on tumor-infiltrating lymphocytes (TIL) while PD-L1 is expressed on tumor cells. Assessment of PD-L1 expression in combination with CD8+ TIL density may be a useful predictive metric in multiple cancers, including stage III NSCLC, hormone receptor negative breast cancer and sentinel lymph node melanoma (2-4). Clinical trials utilizing humanized chimeric antibodies that block inhibitory checkpoints, such as anti-PD-1 and anti-PD-L1, have demonstrated delayed tumor growth and increased survival (5). While identification of PD-L1 overexpression by IHC is not yet standardized, it has become increasingly important to identify these tumors, as a directed therapy may improve clinical outcomes in these patients (6). In cutaneous melanoma, the targeting of PD-1/PD-L1 has provided meaningful clinical benefit for patients in just the past 5-10 years (7). The use of IHC for protein identification, along with novel therapies, such as checkpoint inhibitors and vaccines, are generating new options for the treatment of cancer patients. The PD-L1 [CAL10] clone does not cross react with PD-L2.

**Principle of Procedure:**
Antigen detection in tissues and cells is a multi-step immunohistochemical process. The initial step binds the primary antibody to its specific epitope. After labeling the antigen with a primary antibody, an enzyme labeled polymer is added to bind to the primary antibody. The detection of the bound antibody is evidenced by a colorimetric reaction.

**Source:** Rabbit monoclonal

**Species Reactivity:** Human; others not tested

**Clone:** CAL10

**Isotype:** IgG

**Total Protein Concentration:** ~10 mg/ml. Call for lot specific Ig concentration.

**Epitope/Antigen:** Peptide corresponding to the region within human PD-L1

**Cellular Localization:** Membranous/cytoplasmic

**Positive Tissue Control:** Lung adenocarcinoma or tonsil

**Known Applications:**
Immunohistochemistry (formalin-fixed paraffin-embedded tissues)

**Supplied As:** Buffer with protein carrier and preservative

**Storage and Stability:**
Store at 2°C to 8°C. Do not use after expiration date printed on vial. If reagents are stored under conditions other than those specified in the package insert, they must be verified by the user. Diluted reagents should be used promptly; any remaining reagent should be stored at 2°C to 8°C.

**Protocol Recommendations:**
- **Peroxide Block:** Block for 5 minutes with Biocare's Peroxidized 1.
- **Pretreatment:** Perform heat retrieval using Biocare's Diva Decloaker. Refer to the Diva Decloaker data sheet for specific instructions.
- **Primary Antibody:** Incubate for 5 minutes at RT with Biocare's Background Punisher.
- **Protein Block (Optional):** Incubate for 5-10 minutes at RT with Biocare's Background Punisher.
- **Probe:** N/A
- **Polymer:** Incubate for 30 minutes at RT with a secondary-conjugated polymer.
- **Chromogen:**
  - Incubate for 5 minutes at RT with Biocare's DAB – OR – Incubate for 5-7 minutes at RT with Biocare's Warp Red.
- **Counterstain:**
  - Counterstain with hematoxylin. Rinse with deionized water. Apply Tacha's Bluing Solution for 1 minute. Rinse with deionized water.

**Technical Note:**
1. This antibody has been standardized with Biocare's MACH 4 detection system. Use TBST buffer for washing steps.
2. This clone demonstrates a weak protein expression in endothelial cells.

**Limitations:**
The optimum antibody dilution and protocols for a specific application can vary. These include, but are not limited to fixation, heat-retrieval method, incubation times, tissue section thickness and detection kit used. Due to the superior sensitivity of these unique reagents, the recommended incubation times and titers listed are not applicable to other detection systems, as results may vary. The data sheet recommendations and protocols are based on exclusive use of Biocare products. Ultimately, it is the responsibility of the investigator to determine optimal conditions. The clinical interpretation of any positive or negative staining should be evaluated within the context of clinical presentation, morphology and other histopathological criteria by a qualified pathologist. The clinical interpretation of any positive or negative staining should be complemented by morphological studies using proper positive and negative internal and external controls as well as other diagnostic tests.

**Quality Control:**
Refer to CLSI Quality Standards for Design and Implementation of Immunohistochemistry Assays; Approved Guideline-Second edition (I/LS82-2) CLSI Wayne, PA USA (www.clsi.org). 2011

**Precautions:**
1. This antibody contains less than 0.1% sodium azide. Concentrations less than 0.1% are not reportable hazardous materials according to U.S. 29 CFR 1910.1200, OSHA Hazard communication and EC Directive 91/155/EC. Sodium azide (NaH) used as a preservative is toxic if ingested. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. Upon disposal, flush with large volumes of water to prevent azide build-up in plumbing. (Center for
Precautions Cont’d:
Disease Control, 1976, National Institute of Occupational Safety and Health, 1976) (8)
2. Specimens, before and after fixation, and all materials exposed to them should be handled as if capable of transmitting infection and disposed of with proper precautions. Never pipette reagents by mouth and avoid contacting the skin and mucous membranes with reagents and specimens. If reagents or specimens come into contact with sensitive areas, wash with copious amounts of water. (9)
3. Microbial contamination of reagents may result in an increase in nonspecific staining.
4. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change.
5. Do not use reagent after the expiration date printed on the vial.
6. The SDS is available upon request and is located at http://biocare.net.

Troubleshooting:
Follow the antibody specific protocol recommendations according to data sheet provided. If atypical results occur, contact Biocare’s Technical Support at 1-800-542-2002.

References: