

CD4 (Helper/Inducer)

Concentrated and Prediluted Monoclonal Antibody
901-153-062917

BIOCARE
M E D I C A L

Catalog Number:	CM 153 AK, BK, CK	PM 153 AA	OAI 153 T60
Description:	0.1, 0.5, 1.0 ml, concentrated	6.0 ml, prediluted	60 tests, prediluted
Dilution:	1:50	Ready-to-use	Ready-to-use
Diluent:	Van Gogh Yellow	N/A	N/A

Intended Use:

For In Vitro Diagnostic Use

CD4 (Helper/Inducer) [BC/1F6] is a mouse monoclonal antibody that is intended for laboratory use in the qualitative identification of CD4 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:

CD4 is expressed in a T-cell subset (helper/inducer) and is found in approximately 80% of thymocytes and in 45% of peripheral blood lymphocytes. CD4 is expressed in the majority of T-cell lymphomas, including mycosis fungoides. CD4 has been used in lymphoma panels that include CD3, CD5, CD8, CD7 and TIA-1.

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. A secondary antibody may be applied to bind the primary antibody, followed by an enzyme labeled polymer; or an enzyme labeled polymer may be applied directly to bind the primary antibody. The detection of the bound primary antibody is evidenced by an enzyme-mediated colorimetric reaction.

Source: Mouse monoclonal

Species Reactivity: Human

Clone: BC/1F6

Isotype: IgG1

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

Epitope/Antigen: CD4

Cellular Localization: Cell surface/membrane

Positive Control: Tonsil or T-cell lymphoma

Known Applications:

Immunohistochemistry (formalin-fixed paraffin-embedded tissues)

Supplied As: Buffer with protein carrier and preservative
Van Gogh Yellow (P902)

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 (manual use):

Peroxide Block: Refer to **Primary Antibody** step.*

Pretreatment: Perform heat retrieval using Biocare's Borg or Reveal Decloaker. Refer to the Borg or Reveal Decloaker product data sheet for specific instructions.

Protein Block (Optional): Incubate for 5-10 minutes at RT with Biocare's Background Punisher.

Primary Antibody: Incubate for 30 minutes at RT.

*Block for 5 minutes with Peroxidized 1 after the primary antibody.

Probe: Incubate for 10 minutes at RT with a secondary probe.

Polymer: Incubate for 10-20 minutes at RT with a tertiary polymer.

Protocol Recommendations (manual use) Cont'd:

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.

Protocol Recommendations (ONCORE Automated Slide Staining System):

OAI153 is intended for use with the ONCORE Automated Slide Staining System. Refer to the ONCORE Automated Slide Staining System User Manual for specific instructions on its use. Protocol parameters in the ONCORE Automated Slide Stainer Protocol Editor should be programmed as follows:

Protocol Name: CD4

Protocol Template (Description): IHC Extras Template

Dewaxing (DS Option): DS2

Antigen Retrieval (AR Option): AR1, high pH; 103°C

Reagent Name, Time, Temp.: CD4, 30 min., 25°C

Use of **Mouse Amp HRP Detection (ORI6050)** is required for the above antibody protocol. Mouse HRP Detection (ORI6007) is not recommended.

Technical Note:

This antibody has been optimized for use with Biocare's MACH 4 Universal HRP-Polymer Detection and ONCORE Mouse Amp HRP Detection. Use TBS for washing steps.

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 protocol 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.

Precautions:

1. This antibody contains less than 0.1% sodium azide. Concentrations less than 0.1% are not reportable hazardous material according to U.S. 29 CFR 1910.1200, OSHA Hazard communication and EC Directive 91/155/EC. Sodium azide (NaN₃) 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 Disease Control, 1976, National Institute of Occupational Safety and Health, 1976) (4)

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Precautions Cont'd:

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 in contact with sensitive areas, wash with copious amounts of water. (5)
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 expiration date printed on the vial.
6. The SDS is available upon request and is located at <http://biocare.net>.

Quality Control:

Refer to CLSI Quality Standards for Design and Implementation of Immunohistochemistry Assays; Approved Guideline-Second edition (I/LA28-A2). CLSI Wayne, PA, USA (www.clsi.org). 2011.

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:

1. Rudiger T, *et al.* Peripheral T-cell lymphoma with distinct perifollicular growth pattern: a distinct subtype of T-cell lymphoma. *Am J Surg Pathol.* 2000 Jan; 24(1):117-22.
2. Izban KF, Hsi ED, Alkan S. Immunohistochemical analysis of mycosis fungoides on paraffin-embedded tissue sections. *Mod Pathol.* 1998 Oct; 11(10):978-82.
3. Macon WR, Salhany KE. T-cell subset analysis of peripheral T-cell lymphomas by paraffin section immunohistology and correlation of CD4/CD8 results with flow cytometry. *Am J Clin Pathol.* 1998 May; 109(5):610-7.
4. Center for Disease Control Manual. Guide: Safety Management, NO. CDC-22, Atlanta, GA. April 30, 1976 "Decontamination of Laboratory Sink Drains to Remove Azide Salts."
5. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline-Fourth Edition CLSI document M29-A4 Wayne, PA 2014.