BOB-1

Concentrated and Prediluted Monoclonal Antibody
901-418-110817

BIOCARE MEDICAL

Catalog Number:	**CM 418 A, B**	**PM 418 AA**
Description:	0.1, 0.5 ml, concentrated	6.0 ml, prediluted
Dilution:	1:50	Ready-to-use
Diluent:	Renoir Red	N/A

Intended Use:
For In Vitro Diagnostic Use
BOB-1 [TG14] is a mouse monoclonal antibody that is intended for laboratory use in the qualitative identification of B-cell specific octamer binding protein-1 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:
BOB-1 (B-cell specific octamer binding protein-1) protein is a B-lymphocyte-specific transcriptional coactivator. It interacts with Oct-1 and Oct-2 transcription factors. BOB-1 and Oct-2 play essential roles in germinal center formation and immunoglobulin production. BOB-1 has been reported to be detectable in all B-cell populations found in reactive lymphoid tissues, the strongest expression being found in germinal center B-cells and plasma cells. BOB-1 and Oct-2 are most useful for the B-lineage determination of CD20-plasmablastic, or primary effusion subtypes of diffuse large B-cell lymphoma (DLBCL). Other studies have shown BOB-1, CD79a and Cyclin E are the most appropriate markers to discriminate classical Hodgkin's lymphoma from primary mediastinal large B-cell lymphoma. The strong nuclear expression of BOB-1 and Oct-2 by germinal center-derived lymphomas makes these antibodies a novel class of broad spectrum B-lineage immunohistochemical markers in the differential diagnosis of lymphomas.

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, a secondary antibody is added to bind to the primary antibody. An enzyme label is then added to bind to the secondary antibody; this detection of the bound antibody is evidenced by a colorimetric reaction.

Source: Mouse monoclonal
Species Reactivity: Human; others not tested
Clone: TG14
Isotype: IgG2b
Total Protein Concentration: ~10 mg/ml. Call for lot specific Ig concentration.
Epitope/Antigen: Prokaryotic recombinant protein containing 116 amino acids of the C-terminus of BOB-1
Cellular Localization: Nuclear
Positive Tissue Control: Tonsil
Known Applications:
Immunohistochemistry (formalin-fixed paraffin-embedded tissues)
Supplied As: Buffer with protein carrier and preservative
Storage and Stability:
Store at 2ºC to 8ºC. 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 2ºC to 8ºC.

Protocol Recommendations:
Peroxide Block: Block for 5 minutes with Biocare's Peroxidazed 1.

Protocol Recommendations Cont'd:
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 30-45 minutes at RT.
Probe: Incubate for 10 minutes at RT with a secondary probe.
Polymer: Incubate for 10-20 minutes at RT with a tertiary 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 TBS buffer for washing steps.
2. When using Reveal, a 45-10-20 detection procedure is recommended.
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/LA28-A2) 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 (NaN_3) 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) (6)
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. (7)

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Precautions Cont'd:
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:
1. Hoeller S, *et al*. BOB.1, CD79a and Cyclin E are the most appropriate markers to discriminate classical Hodgkin's lymphoma from primary mediastinal large B-cell lymphoma. Histopathology. 2010 Jan; 56(2):217-28.
2. Advani AS, *et al*. OCT-2 expression and OCT-2/BOB.1 co-expression predict prognosis in patients with newly diagnosed acute myeloid leukemia. Leuk Lymphoma. 2010 Apr; 51(4):606-12.
3. McCune RC, *et al.* Expression profiling of transcription factors Pax-5, Oct-1, Oct-2, BOB.1 and PU.1 in Hodgkin's and non-Hodgkin's lymphomas: a comparative study using high throughput tissue microarrays. Mod Pathol. 2006; 19(7):1010-8.
4. Chu PG, *et al.* Lineage determination of CD20- B-Cell neoplasms: an immunohistochemical study. Am J Clin Pathol. 2006 Oct; 126(4):534-44.
5. Browne P, *et al*. The B-cell transcription factors BSAP, Oct-2, and BOB.1 and the pan-B-cell markers CD20, CD22, and CD79a are useful in the differential diagnosis of classic Hodgkin lymphoma. Am J. Clin Pathol. 2003 Nov; 120(5):767-77.
6. 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."
7. 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.

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