

CD33

Concentrated and Prediluted Monoclonal Antibody
902-3116-020226



Available Product Formats				
Format	Catalog Number	Description	Dilution	Diluent
Concentrate	ACR 3116 A, C	0.1, 1.0 mL	1:50	Renoir Red
Predilute	APR 3116 AA	6.0 mL	Ready-to-use	N/A
NeoPATH PRO	NPAR 3116 T40	40 tests	Ready-to-use	N/A

Intended Use:

For Research Use Only. Not for use in diagnostic procedures.
This mouse antibody is intended to detect CD33.

Background Information:

CD33 or Siglec-3 is a 67 kD glycosylated transmembrane receptor expressed on myeloid-specific cells.^{5,6} The CD33 antibody was also found to be a useful marker in the workup of myeloid sarcomas.⁵⁻⁷ In normal bone marrow trephine biopsies, clone PWS44 stains myeloid, myelomonocytic hemopoiesis and mature macrophages; cells of the erythroid and megakaryocytes series are negative for CD33.⁵

Known Applications:

Immunohistochemistry (Formalin-fixed paraffin-embedded tissues). Other applications have not been tested.

Supplied As:

Concentrate:

Buffered saline solution, pH 7.2 – 7.4, contains a protein carrier and less than 0.1% sodium azide preservative. See Safety Data Sheet for additional details.

Ready-to-use:

Buffered saline solution, pH 6.1 – 6.3, contains a protein carrier and less than 0.1% sodium azide preservative. See Safety Data Sheet for additional details.

Materials and Methods:

Reagents Provided:

Host Source: Mouse monoclonal

Species Reactivity: Human; other species not tested.

Clone: PWS44

Isotype: IgG2b

Protein Concentration: Contact Biocare's Technical Support for specific Ig concentration

Specificity: Prokaryotic recombinant protein corresponding to a region of the C2 domain on human CD33

Reconstitution, Mixing, Dilution, and Titration:

Prediluted antibody reagent is optimally diluted for use with the above listed staining systems. Further dilution may result in loss of antigen staining. The user must validate any such change. Differences in tissue processing and technical procedures in the user's laboratory may produce significant variability in results necessitating regular performance of in-house controls. Concentrated reagent requires dilution as indicated in table above.

Storage and Stability:

Store at 2°C to 8°C. The product is stable to the expiration date printed on the vial label when stored under these conditions. Do not use after expiration date. Storage under any condition other than those specified must be verified. Diluted reagents should be used promptly; store any remaining reagent at 2°C to 8°C. The stability of user diluted reagents has not been established by Biocare.

Staining Protocol Recommendations (NeoPATH PRO):

NPAR3116 is compatible for use with the NeoPATH PRO. Below are programming and protocol recommendations to assist the user when staining

using Biocare's NeoPATH PRO Automated Staining Platform for research applications. The user is responsible for further optimizations of the protocol.

Chromogen Option: DAB

Antibody Protocol: CD33-BC, 20 min at RT

Template: HRP_HIGH_105C_20MINAB+BLOCK

Dewax: Dewax STD; 20 min at 75°C

Antigen Retrieval (HIER Option): HIGH_105C_30MIN

Enzyme: N/A

Block Option: NeoPATH Pro Background Punisher, 10 min at RT

Detection: HRP_20AB+BLOCK (Amplifier; 10 min at RT, Polymer; 25 min at RT)

Chromogen: 7 min DAB + 2 min DAB Enhancer at RT.

Hematoxylin: 7 min at RT

Staining Protocol Recommendations (intelliPATH FLX® and manual use):

Below are programming and protocol recommendations to assist the user when staining manually and/or using Biocare's intelliPATH FLX Automated Staining Platform for research applications. The user is responsible for further optimizations of the protocol.

Peroxide Block: Block for 5 minutes with a peroxide blocking reagent.

Pretreatment: Perform heat retrieval if applicable. When using automated equipment, refer to the equipment operator's manual for specific equipment use instructions.

Protein Block (Optional): Incubate for 5-10 minutes at RT with a protein blocking reagent.

Primary Antibody: Incubate for 30 minutes at RT.

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

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

Chromogen: Incubate for 5-7 minutes at RT with desired chromogen.

Counterstain: Counterstain with hematoxylin. Rinse with deionized water. Apply Bluing Solution for 1 minute. Rinse with deionized water.

Technical Note:

This antibody, for intelliPATH FLX and manual use, has been standardized with MACH 4 detection system. Use TBS for washing steps.

Limitations:

This product is provided for Research Use Only (RUO) and is not for use in diagnostic procedures. Suitability for specific applications may vary and it is the responsibility of the end user to determine the appropriate application for its use.

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 (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)¹
2. Handle materials of human or animal origin as potentially biohazardous and dispose of such materials with proper precautions. In the event of exposure, follow the health directives of the responsible authorities where used.^{2,3}
3. 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



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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.⁴

4. Microbial contamination of reagents may result in an increase in nonspecific staining.
5. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change.
6. Do not use reagent after the expiration date printed on the vial.
7. To prevent evaporation and ensure maximum test capacity, promptly cap and remove reagents from automated instruments after each run. Leaving reagents exposed can reduce their effectiveness and the number of tests they can provide. Always store reagents as directed to maintain their integrity.
8. Dispose of all used reagents and any other contaminated disposable materials following procedures for infectious or potentially infectious waste. It is the responsibility of each laboratory to handle solid and liquid waste according to their nature and degree of hazardousness and to treat and dispose of it (or have them treated and disposed of) in accordance with any applicable regulations.
9. Follow local disposal regulations for your location along with recommendations in the Safety Data Sheet to determine the safe disposal of this product
10. The SDS is available upon request and is located at <http://biocare.net>.
11. Report any serious incidents related to this device by contacting the local Biocare representative and the applicable competent authority of the Member State or country where the user is located.

Technical Support:

Contact Biocare's Technical Support at 1-800-542-2002 for questions regarding this product.

References:

1. 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."
2. Occupational Safety and Health Standards: Occupational exposure to hazardous chemicals in laboratories. (29 CFR Part 1910.1450). Fed. Register.
3. Directive 2000/54/EC of the European Parliament and Council of 18 September 2000 on the protection of workers from risks related to exposure to biological agents at work.
4. 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.
5. Hoyer JD, *et al.* CD33 detection by immunohistochemistry in paraffin-embedded tissues: a new antibody shows excellent specificity and sensitivity for cells of myelomonocytic lineage. *Am J Clin Pathol.* 2008 Feb; 129(2):316-23.
6. Rollins-Raval MA, Roth CG. The value of immunohistochemistry for CD14, CD123, CD33, myeloperoxidase and CD68R in the diagnosis of acute and chronic myelomonocytic leukaemias. *Histopathology.* 2012 May; 60(6):933-42.
7. Amador-Ortiz C, *et al.* Use of classic and novel immunohistochemical markers in the diagnosis of cutaneous myeloid sarcoma. *J Cutan Pathol.* 2011 Dec; 38(12):945-53.
8. Brotelle T, *et al.* Gemtuzumab ozogamicin for treatment of acute myeloid leukemia. *Bull Cancer.* 2014 Feb; 101(2):211-8.



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