

Phospho-Histone H3

Concentrated and Prediluted Polyclonal Antibody

Control Number: 902-404-091917

Catalog Number:	ACR 404 A,C	APR 404 AA
Description:	0.1, 1.0 ml, concentrated	6.0 ml, prediluted
Dilution:	1:100-1:200	Ready-to-use
Diluent:	Da Vinci Green	N/A

Intended Use:

For Research Use Only. Not for use in diagnostic procedures.

Summary and Explanation:

Phospho-Histone H3 (pHH3) is a recently described immunomarker specific for cells undergoing mitosis. The phosphorylation of histone H3 plays an important role in gene expression, chromatin remodeling, chromosome condensation and cell division. Histone H3 phosphorylation may initiate at different phases of cell division in different organisms, but metaphase chromosomes are always found to be heavily phosphorylated. H3 phosphorylation at serine 10 and serine 28 is coupled with mitotic chromosome segregation and condensation. Prognostic significance of the mitotic index (MI) using the Mitosis marker anti-PHH3 antibody have been reported to be of great value in breast cancer, melanoma and meningiomas. pHH3 staining may also be a useful method in which accurate determination of proliferation potential is relevant to tumor grading or clinical treatment decision-making.

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 polyclonal

Species Reactivity: Human

Clone: N/A

Isotype: N/A

Epitope/Antigen: PhosphoSer10.

Positive Control: Melanoma

Cellular Localization: Nuclear

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

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.

Staining Protocol Recommendations:

Peroxide Block:

Block for 5 minutes with Biocare's Peroxidized 1.

Pretreatment Protocol:

Heat Retrieval Method:

Retrieve sections under pressure using Biocare's Decloaking Chamber, followed by a wash in distilled water; alternatively, steam tissue sections for 45-60 minutes. Allow solution to cool for 10 minutes then wash in distilled water.

Protein Block:

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

Primary Antibody: Incubate for 30 minutes at RT.

Staining Protocol Recommendations Cont'd:

Probe: N/A

Polymer: Incubate for 30 minutes at RT with a polymer.

Chromogen:

Incubate for 5 minutes at RT when using Biocare's DAB.

Technical Note:

This antibody has been standardized with Biocare's MACH 2 detection system. It can also be used on an automated staining system and with other Biocare polymer detection kits. Use TBS buffer 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) (5)

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. (6)

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>.

Technical Support:

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

References:

1. Skaland I, *et al.* The prognostic value of the proliferation marker Phosphohistone H3 (PPH3) in luminal, basal-like and triple negative phenotype invasive lymph node-negative breast cancer. *Cellul Oncol.* 2009;31:261-71.

2. Nasr M, *et al.* Comparison of pHH3, Ki-67, and Survivin immunoreactivity in benign and malignant melanocytic lesions. *Am J. Dermatopathol.* 2008;30(2):117-22.

3. Skaland I, *et al.* Phosphohistone H3 expression has stronger prognostic value than classical prognosticators in invasive lymph node-negative breast cancer patients less than 55 years of age. *Mod Pathol.* 2007;20:1307-15.

4. Kim Y.J. *et al.* Prognostic significance of the mitotic index using the mitosis marker anti-phosphohistone H3 in meningiomas. *Am J.Clin Pathol.* 2007;128:118-25.

5. 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."

6. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory workers from occupationally Acquired Infections; Approved guideline-Third Edition CLSI document M29-A3 Wayne, PA 2005.