anti-c-MYC (Y69) Rabbit Monoclonal Primary Antibody

REF 790-4628

06504612001

IVD Σ 50

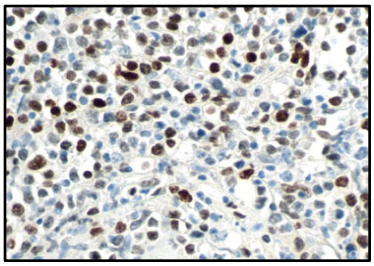

Figure 1. Anti-c-MYC (Y69) antibody nuclear staining of lymphoma tissue.

INTENDED USE

Anti-c-MYC (Y69) Rabbit Monoclonal Primary Antibody is intended for laboratory use in the qualitative immunohistochemical detection of c-MYC protein by light microscopy in sections of formalin-fixed, paraffin-embedded tissue stained on a BenchMark IHC/ISH instrument.

This product should be interpreted by a qualified pathologist in conjunction with histological examination, relevant clinical information, and proper controls.

This antibody is intended for in vitro diagnostic (IVD) use.

SUMMARY AND EXPLANATION

The MYC family of proteins, comprised of c-MYC, L-MYC and N-MYC, are master transcriptional regulators that control almost all cellular processes, including proliferation, differentiation, adhesion and survival.[1] L-MYC and N-MYC overexpression are restricted to small cell lung cancer and neural cancer, respectively.[1,2] However, c-MYC overexpression is observed in a broad array of human neoplasms, including hematologic malignancies (e.g. Burkitt lymphoma, diffuse large B-cell lymphoma (DLBCL), B-acute lymphocytic leukemia, multiple myeloma, primary plasma cell leukemia) and solid tumors (breast, ovarian, cervical, endometrial, colorectal, prostate, lung, gastric and pancreatic cancers; renal clear cell carcinoma and adrenal cell carcinoma; medulloblastoma, neuroblastoma and glioblastoma).[1-4] The oncogenic alterations leading to c-MYC overexpression vary widely in these neoplasms, and include chromosomal translocation, gene amplification, insertional mutation, enhanced translation, and increased protein stability.[2,4,5]

Chromosomal translocation of *c-MYC* is a defining feature of Burkitt lymphoma; the translocation juxtaposes *c-MYC* on chromosome 8 with one of three immunoglobulin genes located on chromosomes 14, 2 or 22.[6,7,8] Several studies have consistently reported high levels of c-MYC expression, as detected by immunohistochemistry (IHC), in Burkitt lymphoma specimens.[9-14] Additionally, several studies have demonstrated high concordance between c-MYC overexpression as detected by IHC, with *c-MYC* translocation as detected by FISH in Burkitt lymphoma specimens.[10,11,12,14] Thus, the detection of c-MYC IHC with anti-c-MYC (Y69) Rabbit Monoclonal Primary Antibody (anti-c-MYC (Y69) antibody) may be used as an aid in the identification of Burkitt lymphoma.

Per the 2016 revised WHO classification of lymphoid neoplasms, once the diagnosis of DLBCL is established, further characterization is required.[6] DLBCL can be characterized by cell of origin, molecular features, and genetic or mutational landscape.[9,15] c-MYC IHC is often performed in this context. Thus, the detection of c-MYC by IHC with anti-c-MYC (Y69) antibody may be used as an aid in the characterization of DLBCL.

Anti-c-MYC (Y69) antibody is a rabbit monoclonal antibody produced against the N-terminus of the 64 kDa c-MYC protein. The staining pattern for this antibody is nuclear. It may be used as part of a panel of IHC studies.

PRINCIPLE OF THE PROCEDURE

The anti-c-MYC (Y69) antibody binds to the N-terminus of the c-MYC protein in formalin-fixed, paraffin-embedded (FFPE) tissue sections. This antibody can be visualized using *ultra*View Universal DAB Detection Kit (Cat. No. 760-500 / 05269806001) or OptiView DAB IHC Detection Kit (Cat. No. 760-700 / 06396500001). Refer to the respective detection kit method sheet for further instructions.

MATERIAL PROVIDED

Anti-c-MYC (Y69) antibody contains sufficient reagent for 50 tests.

One 5 mL dispenser of anti-c-MYC (Y69) antibody contains approximately 120 µg of a rabbit monoclonal antibody.

The antibody is diluted in a TBS buffer containing carrier protein.

Specific antibody concentration is approximately 24 µg/mL.

Anti-c-MYC (Y69) antibody is a rabbit monoclonal antibody produced as purified cell culture supernatant.

Refer to the appropriate VENTANA detection kit method sheet for detailed descriptions of: Principle of the Procedure, Material and Methods, Specimen Collection and Preparation for Analysis, Quality Control Procedures, Troubleshooting, Interpretation of Results, and Limitations.

MATERIALS REQUIRED BUT NOT PROVIDED

Staining reagents, such as VENTANA detection kits and ancillary components, including negative and positive tissue control slides, are not provided.

Not all products listed in the method sheet may be available in all geographies. Consult your local support representative.

The following reagents and materials may be required for staining but are not provided:

1. Recommended control tissue
2. Microscope slides, positively charged
3. Rabbit Monoclonal Negative Control Ig (Cat. No. 790-4795 / 06683380001)
4. OptiView DAB IHC Detection Kit (Cat. No. 760-700 / 06396500001)
5. *ultra*View Universal DAB Detection Kit (Cat. No. 760-500 / 05269806001)
6. Amplification Kit (Cat. No. 760-080 / 05266114001)
7. EZ Prep Concentrate (10X) (Cat. No. 950-102 / 05279771001)
8. Reaction Buffer Concentrate (10X) (Cat. No. 950-300 / 05353955001)
9. LCS (Predilute) (Cat. No. 650-010 / 05264839001)
10. ULTRA LCS (Predilute) (Cat. No. 650-210 / 05424534001)
11. Cell Conditioning Solution (CC1) (Cat. No. 950-124 / 05279801001)
12. ULTRA Cell Conditioning Solution (ULTRA CC1) (Cat. No. 950-224 / 05424569001)
13. Hematoxylin II (Cat. No. 790-2208 / 05277965001)
14. Bluing Reagent (Cat. No. 760-2037 / 05266769001)
15. Permanent mounting medium
16. Cover glass
17. Automated coverslipper
18. General purpose laboratory equipment
19. BenchMark IHC/ISH instrument

STORAGE AND STABILITY

Upon receipt and when not in use, store at 2-8°C. Do not freeze.

To ensure proper reagent delivery and the stability of the antibody, replace the dispenser cap after every use and immediately place the dispenser in the refrigerator in an upright position.

Every antibody dispenser is expiration dated. When properly stored, the reagent is stable to the date indicated on the label. Do not use reagent beyond the expiration date.

SPECIMEN PREPARATION

Routinely processed FFPE tissues are suitable for use with this primary antibody when used with VENTANA detection kits and a BenchMark IHC/ISH instruments. The recommended tissue fixative is 10% neutral buffered formalin.[16] Sections should be cut at approximately 4 µm in thickness and mounted on positively charged slides. Slides should be stained immediately, as antigenicity of cut tissue sections may diminish over time. Ask your Roche representative for a copy of “Recommended Slide Storage and Handling” for more information

It is recommended that positive and negative controls be run simultaneously with unknown specimens.

WARNINGS AND PRECAUTIONS

1. For in vitro diagnostic (IVD) use.
2. For professional use only.
3. CAUTION: In the United States, Federal law restricts this device to sale by or on the order of a physician. (Rx Only)

4. Do not use beyond the specified number of tests.
5. Positively charged slides may be susceptible to environmental stresses resulting in inappropriate staining. Ask your Roche representative for more information on how to use these types of slides.
6. Materials of human or animal origin should be handled as biohazardous materials and disposed of with proper precautions. In the event of exposure, the health directives of the responsible authorities should be followed.[17,18]
7. Avoid contact of reagents with eyes and mucous membranes. If reagents come in contact with sensitive areas, wash with copious amounts of water.
8. Avoid microbial contamination of reagents as it may cause incorrect results.
9. For further information on the use of this device, refer to the BenchMark IHC/ISH instrument User Guide, and instructions for use of all necessary components located at dialog.roche.com.
10. Consult local and/or state authorities with regard to recommended method of disposal.
11. Product safety labeling primarily follows EU GHS guidance. Safety data sheet available for professional user on request.
12. To report suspected serious incidents related to this device, contact the local Roche representative and the competent authority of the Member State or Country in which the user is established.

STAINING PROCEDURE

VENTANA primary antibodies have been developed for use on BenchMark IHC/ISH instruments in combination with VENTANA detection kits and accessories. Refer to the tables below for recommended staining protocols.

This antibody has been optimized for specific incubation times but the user must validate results obtained with this reagent.

The parameters for the automated procedures can be displayed, printed and edited according to the procedure in the instrument User Guide. Refer to the appropriate VENTANA detection kit method sheet for more details regarding immunohistochemistry staining procedures.

For more details on the proper use of this device, refer to the inline dispenser method sheet associated with P/N 790-4628.

Table 1. Recommended staining protocol for anti-c-MYC (Y69) antibody with *ultra*View Universal DAB Detection Kit on a BenchMark IHC/ISH instrument.

Procedure Type	Method	
	XT	ULTRA or ULTRA PLUS [a]
Deparaffinization	Selected	Selected
Cell Conditioning (Antigen Unmasking)	CC1, Standard	ULTRA CC1 64 minutes, 95°C
Antibody (Primary)	16 minutes, 37°C	16 minutes, 36°C
Amplification	Selected	Selected (Rabbit)
Counterstain	Hematoxylin II, 4 minutes	
Post Counterstain	Bluing, 4 minutes	

[a] Concordance was demonstrated between BenchMark ULTRA and BenchMark ULTRA PLUS instruments using representative assays.

Table 2. Recommended staining protocol for anti-c-MYC (Y69) antibody with OptiView DAB IHC Detection Kit on a BenchMark IHC/ISH instrument.

Procedure Type	Method	
	XT	ULTRA or ULTRA PLUS [a]
Deparaffinization	Selected	Selected
Cell Conditioning (Antigen Unmasking)	CC1 64 minutes	ULTRA CC1 64 minutes, 100°C
Pre-Primary Peroxidase Inhibitor	Selected	Selected
Antibody (Primary)	16 minutes, 37°C	16 minutes, 36°C
OptiView HQ Linker	8 minutes (default)	
OptiView HRP Multimer	8 minutes (default)	
Counterstain	Hematoxylin II, 4 minutes	
Post Counterstain	Bluing, 4 minutes	

[a] Concordance was demonstrated between BenchMark ULTRA and BenchMark ULTRA PLUS instruments using representative assays.

Due to variation in tissue fixation and processing, as well as general lab instrument and environmental conditions, it may be necessary to increase or decrease the primary antibody incubation, cell conditioning or protease pretreatment based on individual specimens, detection used, and reader preference. For further information on fixation variables, refer to "Immunohistochemistry Principles and Advances."[19]

NEGATIVE REAGENT CONTROL

In addition to staining with the anti-c-MYC (Y69) antibody, a second slide should be stained with the appropriate negative control reagent.

POSITIVE TISSUE CONTROL

Optimal laboratory practice is to include a positive control section on the same slide as the test tissue. This helps identify any failures applying reagents to the slide. Tissue with weak positive staining is best suited for quality control. Control tissue may contain both positive and negative staining elements and serve as both the positive and negative control. Control tissue should be fresh autopsy, biopsy, or surgical specimen, prepared or fixed as soon as possible in a manner identical to test sections.

Known positive tissue controls should be utilized only for monitoring performance of reagents and instruments, not as an aid in determining specific diagnosis of test samples. If the positive tissue controls fail to demonstrate positive staining, results of the test specimen should be considered invalid.

An example of positive control tissue for this antibody is skin.

STAINING INTERPRETATION / EXPECTED RESULTS

The cellular staining pattern for anti-c-MYC (Y69) antibody is nuclear.

SPECIFIC LIMITATIONS

This antibody may demonstrate cross-reactivity to mucin in the small intestine. This non-specific staining does not interfere with staining interpretation.

OptiView detection system is generally more sensitive than the *ultra*View Universal DAB detection system. The user must validate the results obtained with this reagent and detection systems.

All assays might not be registered on every instrument. Please contact your local Roche representative for more information.

PERFORMANCE CHARACTERISTICS

ANALYTICAL PERFORMANCE

Staining tests for specificity, sensitivity, and precision were conducted and the results are listed below.

Sensitivity and Specificity

Table 3. Sensitivity/Specificity of anti-c-MYC (Y69) antibody was determined by testing FFPE normal tissues.

Tissue	# positive / total cases	Tissue	# positive / total cases
Cerebrum	0/6	Stomach	0/3

Tissue	# positive / total cases	Tissue	# positive / total cases
Cerebellum	0/3	Small intestine	2/3
Adrenal gland	0/3	Colon	1/3
Ovary	3/3	Liver	0/3
Pancreas	0/3	Salivary glands	1/3
Lymph node	8/14	Larynx	2/3
Pituitary gland	0/3	Kidney	1/3
Testis	0/3	Prostate	0/3
Thyroid	0/4	Bladder	2/3
Breast	1/3	Endometrium	1/6
Spleen	1/3	Cervix	1/5
Tonsil	6/9	Skeletal muscle	0/6
Thymus	0/3	Skin	4/7
Bone marrow	0/3	Nerve	1/6
Lung	0/4	Mesothelium	0/5
Heart	0/3	Eye	0/2
Esophagus	1/3		

Table 4. Sensitivity/Specificity of anti-c-MYC (Y69) antibody was determined by testing a variety of FFPE neoplastic tissues.

Pathology	# positive / total cases
Glioblastoma (Cerebrum)	1/1
Meningioma (Cerebrum)	0/1
Ependymoma (Cerebrum)	0/1
Oligodendroglioma (Cerebrum)	0/1
Serous adenocarcinoma (Ovary)	1/1
Adenocarcinoma (Ovary)	1/1
Neuroendocrine neoplasm (Pancreas)	0/1
Adenocarcinoma (Pancreas)	0/1
Seminoma (Testis)	1/2
Medullary carcinoma (Thyroid)	0/1
Papillary carcinoma (Thyroid)	0/1
Microinvasive ductal carcinoma (Breast)	0/1
Invasive ductal carcinoma (Breast)	2/2
Small cell carcinoma (Lung)	1/1
Squamous cell carcinoma (Lung)	1/1
Adenocarcinoma (Lung)	0/1
Neuroendocrine carcinoma (Esophagus)	0/1
Adenocarcinoma (Esophagus)	1/1
Signet ring cell carcinoma (Stomach)	0/1
Adenocarcinoma (Small Intestine)	0/1
Stromal sarcoma (Small Intestine)	1/1
Adenocarcinoma (Colon)	1/1
Gastrointestinal stromal tumor (Colon)	0/1
Adenocarcinoma (Rectum)	0/1
Gastrointestinal stromal tumor (Rectum)	1/1
Melanoma (Rectum)	0/1
Hepatocellular carcinoma (Liver)	0/1
Hepatoblastoma (Liver)	0/1
Clear cell carcinoma (Kidney)	0/1
Adenocarcinoma (Prostate)	0/2
Leiomyoma (Uterus)	0/1
Adenocarcinoma (Uterus)	0/1
Clear cell carcinoma (Uterus)	0/1
Squamous cell carcinoma (Cervix)	1/2
Embryonal rhabdomyosarcoma (Striated muscle)	0/1
Basal cell carcinoma (Skin)	0/1
Squamous cell carcinoma (Skin)	1/1
Neurofibroma (Lumbar)	0/1
Neuroblastoma (Retroperitoneal)	0/1
Spindle cell rhabdomyosarcoma (Peritoneum)	0/1
Mesothelioma (Peritoneum)	1/1
Diffuse Large B-cell lymphoma	48/108
Burkitt lymphoma	3/3
Mantle cell lymphoma	0/1
B-cell chronic lymphocytic leukemia	0/2
Nodal marginal zone B-cell lymphoma	0/1
MALT lymphoma	3/7
Follicular lymphoma	1/4
Plasma cell myeloma	1/8
B-cell lymphoma, NOS	23/63
Hodgkin Lymphoma	4/11
Angioimmunoblastic T-cell lymphoma	1/2
Anaplastic large cell lymphoma	0/1
Peripheral T-cell lymphoma, NOS	1/2

Pathology	# positive / total cases
T-cell lymphoma, NOS	9/17
Urothelial carcinoma (Bladder)	0/1
Leiomyosarcoma (Bladder)	0/1
Osteosarcoma (Bone)	0/1
Leiomyosarcoma (Smooth muscle)	0/1

Repeatability

Precision

Precision studies for anti-c-MYC (Y69) antibody were completed to demonstrate:

- Between lot precision of the antibody.
- Within run and between day precision on a BenchMark XT instrument.
- Between instrument precision on the BenchMark XT and BenchMark ULTRA instrument.
- Between platform precision between the BenchMark XT and BenchMark ULTRA instrument.

All studies met their acceptance criteria.

Precision on the BenchMark ULTRA PLUS instrument was demonstrated using representative assays. Studies included Within-run Repeatability, Between-day and Between-run Intermediate Precision. All studies met their acceptance criteria.

CLINICAL PERFORMANCE

Clinical performance data relevant to the intended purpose of anti-c-MYC (Y69) antibody were assessed by systematic review of the literature. The data gathered support the use of the device in accordance with its intended purpose.

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NOTE: A point (period/stop) is always used in this document as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

The summary of safety and performance can be found here:

https://ec.europa.eu/tools/eudamed

Symbols

Ventana uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see dialog.roche.com for definition of symbols used):

GTIN	Global Trade Item Number
UDI	Unique Device Identification
	Indicates the entity importing the medical device into the European Union

REVISION HISTORY

Rev	Updates
F	Updates to Specimen Preparation, Staining Procedure, Analytical Performance, Clinical Performance and Symbols sections. Added BenchMark ULTRA PLUS instrument.

INTELLECTUAL PROPERTY

CONTACT INFORMATION

Ventana Medical Systems, Inc.
1910 E. Innovation Park Drive
Tucson, Arizona 85755
USA
+1 520 887 2155
+1 800 227 2155 (USA)

www.roche.com

EC REP

Roche Diagnostics GmbH
Sandhofer Strasse 116
D-68305 Mannheim
Germany
+800 5505 6606

CE 0123