CONFIRM anti-Vimentin (V9) Primary Antibody

REF 790-2917

05278139001

IVD Σ 50

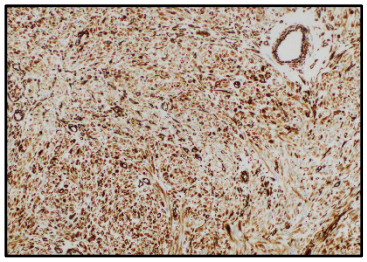

Figure 1. CONFIRM anti-Vimentin (V9) antibody staining in leiomyosarcoma.

INTENDED USE

CONFIRM anti-Vimentin (V9) Primary Antibody is intended for laboratory use in the qualitative immunohistochemical detection of vimentin 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

Vimentin, a 57 kDa type III intermediate filament protein encoded by the VIM gene, is located on chromosome 10 (10p13).[1] Vimentin is the primordial intermediate filament protein expressed during embryogenesis, throughout which it functions to modulate cell formation and mitochondrial movement, provide structure to signaling molecules, and mediate signaling pathways.[1,2,3] Vimentin is predominantly expressed in mesenchymal cells and the majority of tissues including, lymphoid tissues, astrocytes, adipocytes, epidermal cells, and glandular cells.[1,3,4] Under normal physiological conditions, the primary function of vimentin is to control cellular shape, architecture, adhesion, migration, signaling, and motility.[2,4,5] Vimentin also acts as a marker of epithelial-mesenchymal transition (EMT), a critical process for regulating tissue regeneration, inflammatory responses, and cellular differentiation.[2,4,5] Overexpression of vimentin during EMT is associated with malignant transformation, tumor dissemination, and metastases.[2,4,5]

Vimentin is a broad mesenchymal marker that is expressed by virtually all mesenchymal neoplasms, regardless of histologic subtype, although it is not considered to be a cell lineage specific marker.[3,6,7] Vimentin is widely expressed in sarcomas, lymphomas, melanoma, and some carcinomas.[8,9] Among carcinomas, vimentin is typically expressed in clear cell, papillary, collecting duct, and unclassified renal cell carcinoma and is generally absent in chromophobe renal cell carcinoma.[3,10] Furthermore, because vimentin expression is not lineage specific it is typically evaluated as part of a panel of immunohistochemistry (IHC) studies that include other lineage markers.

The detection of vimentin by IHC with the CONFIRM anti-Vimentin (V9) Primary Antibody (CONFIRM anti-Vimentin (V9) antibody) may be used as an aid in the identification of sarcomas and as an aid in the diagnosis and subclassification of renal cell carcinoma. The cellular staining pattern for CONFIRM anti-Vimentin (V9) antibody is cytoplasmic.

PRINCIPLE OF THE PROCEDURE

CONFIRM anti-Vimentin (V9) antibody binds to vimentin in formalin-fixed, paraffin-embedded (FFPE) tissue sections. This antibody can be visualized using OptiView DAB IHC Detection Kit (Cat. No. 760-700 / 06396500001) or *ultra*View Universal DAB Detection Kit (Cat. No. 760-500 / 05269806001). Refer to the respective method sheet for further information.

MATERIAL PROVIDED

CONFIRM anti-Vimentin (V9) antibody contains sufficient reagent for 50 tests.

One 5 mL dispenser of CONFIRM anti-Vimentin (V9) antibody contains approximately 12.5 µg of a mouse monoclonal antibody.

The antibody is diluted in a Tris buffer containing carrier protein and preservative.

Specific antibody concentration is approximately 2.5 µg/mL. There is no known non-specific antibody reactivity observed in this product.

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. Negative Control (Monoclonal) (Cat. No. 760-2014 / 05266670001)
4. *ultra*View Universal DAB Detection Kit (Cat. No. 760-500 / 05269806001)
5. OptiView DAB IHC Detection Kit (Cat. No. 760-700 / 06396500001)
6. EZ Prep Concentrate (10X) (Cat. No. 950-102 / 05279771001)
7. Reaction Buffer Concentrate (10X) (Cat. No. 950-300 / 05353955001)
8. LCS (Predilute) (Cat. No. 650-010 / 05264839001)
9. ULTRA LCS (Predilute) (Cat. No. 650-210 / 0542434001)
10. Cell Conditioning Solution (CC1) (Cat. No. 950-124 / 05279801001)
11. ULTRA Cell Conditioning (ULTRA CC1) (Cat. No. 950-224 / 05424569001)
12. Hematoxylin II (Cat. No. 790-2208 / 05277965001)
13. Bluing Reagent (Cat. No. 760-2037 / 05266769001)
14. General purpose laboratory equipment
15. 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 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 BenchMark IHC/ISH instruments. The recommended tissue fixative is 10% neutral buffered formalin.[11] 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.[12,13]
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 this could produce 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 a BenchMark IHC/ISH instruments in combination with VENTANA detection kits and accessories. Refer to the tables below for recommended staining protocol.

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 790-2917.

Table 1. Recommended staining protocol for CONFIRM anti-Vimentin (V9) antibody with *ultra*View Universal DAB Detection Kit on BenchMark IHC/ISH instruments.

Procedure Type	Method		
	GX	XT	ULTRA or ULTRA PLUS [a]
Deparaffinization	Selected	Selected	Selected
Cell Conditioning (Antigen Unmasking)	CC1, Standard	CC1, Standard	ULTRA CC1 64 minutes, 95 °C (Standard)
Antibody (Primary)	16 minutes, 37 °C	16 minutes, 37 °C	16 minutes, 36 °C
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 CONFIRM anti-Vimentin (V9) antibody with OptiView DAB IHC Detection Kit on BenchMark IHC/ISH instruments.

Procedure Type	Method		
	GX	XT	ULTRA or ULTRA PLUS [a]
Deparaffinization	Selected	Selected	Selected
Cell Conditioning (Antigen Unmasking)	CC1, 24 minutes	CC1, 24 minutes	ULTRA CC1 24 minutes, 100 °C
Pre-Primary Peroxidase Inhibitor	Selected	Selected	Selected
Antibody (Primary)	16 minutes, 37 °C	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."[14]

NEGATIVE REAGENT CONTROL

In addition to staining with CONFIRM anti-Vimentin (V9) 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 a specific diagnosis of patient samples. If the positive tissue controls fail to demonstrate positive staining, results with the test specimens should be considered invalid.

An example of a positive control tissue for this antibody is tonsil.

STAINING INTERPRETATION / EXPECTED RESULTS

The cellular staining pattern for CONFIRM anti-Vimentin (V9) antibody is cytoplasmic.

SPECIFIC LIMITATIONS

OptiView detection system is generally more sensitive than *ultra*View 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 sensitivity, specificity, and precision were conducted and the results are listed below.

Sensitivity and Specificity

Table 3. Sensitivity/Specificity of CONFIRM anti-Vimentin (V9) antibody was determined by testing FFPE normal tissues.

Tissue [a]	# positive / total cases	Tissue	# positive / total cases
Cerebrum	0/3	Stomach	0/3
Cerebellum	0/3	Small intestine	0/3
Adrenal gland	2/3	Colon	0/3
Ovary	3/3	Appendix	0/3
Pancreas	3/3	Liver	0/3
Lymph node	3/3	Salivary gland	0/3
Parathyroid gland	0/3	Pharynx, oral cavity	1/3
Pituitary gland	0/3	Kidney	6/6
Testis	20/20	Prostate	3/3
Thyroid	3/3	Bladder	0/3
Breast	3/3	Endometrium	3/3
Spleen	3/3	Cervix	0/3
Tonsil	6/6	Skeletal muscle	0/3

Tissue [a]	# positive / total cases	Tissue	# positive / total cases
Thymus	3/3	Soft Tissue	3/3
Bone marrow	3/3	Skin	0/3
Lung	3/3	Nerve	3/3
Heart	0/3	Mesothelium	3/3
Esophagus	0/3		

[a] Vimentin is expressed in many normal structures such as fibroblasts, endothelial cells, smooth muscle cells, myoepithelial cells, lymphocytes and macrophages, along with other structures of mesenchymal origin. When evaluating the above normal tissues, a positive or negative status was derived based on assessment of the site specific normal cells, therefore tissues deemed as negative may have staining in some or all of the aforementioned structures.

Table 4. Sensitivity/Specificity of CONFIRM anti-Vimentin (V9) antibody was determined by testing a variety of FFPE neoplastic tissues.

Pathology [a]	# positive / total cases
Glioblastoma (Cerebrum)	1/1
Meningioma (Cerebrum)	1/1
Ependymoma (Cerebrum)	1/1
Oligodendroglioma (Cerebellum)	0/1
Adenocarcinoma (Head, neck)	0/1
Squamous Cell Carcinoma (Head, neck)	0/1
Granulosa cell tumor (Ovary)	1/1
Serous carcinoma (Ovary)	0/1
Dermoid cyst (Ovary)	1/1
Neuroendocrine neoplasm (Pancreas)	1/1
Adenocarcinoma (Pancreas)	1/1
Embryonal carcinoma (Testis)	0/7
Seminoma (Testis)	13/47
Endodermal sinus tumor (Testis)	1/9
Teratoma (Testis)	4/4
NK/T-cell lymphoma (Testis)	1/1
B cell lymphoma, NOS (Testis)	2/3
Non-Hodgkin lymphoma, NOS (Testis)	4/4
Follicular carcinoma (Thyroid)	1/1
Papillary carcinoma (Thyroid)	1/1
Ductal carcinoma in situ (DCIS) (Breast)	0/1
Invasive ductal carcinoma (Breast)	0/1
Invasive lobular carcinoma (Breast)	0/1
Adenoma (Adrenal gland)	1/1
Pheochromocytoma (Adrenal gland)	1/1
Adenocarcinoma (Lung)	0/1
Squamous cell carcinoma (Lung)	0/1
Small cell lung carcinoma (Lung)	0/1
Adenocarcinoma (Esophagus)	0/1
Squamous cell carcinoma (Esophagus)	0/1
Adenocarcinoma (Stomach)	0/1
Gastrointestinal stromal tumor (GIST) (Gastrointestinal)	2/2
Adenocarcinoma (Small intestine)	0/1
Osteosarcoma (Small intestine)	1/1
Adenocarcinoma (Colon)	0/1
Adenosquamous carcinoma (Colon)	0/1
Carcinoid tumor (Appendix)	0/1
Cholangiocarcinoma (Liver)	0/1
Hepatocellular carcinoma (Liver)	0/1
Pleomorphic Adenoma (Salivary gland)	1/1
Warthin's Tumor (Salivary gland)	1/1
Papillary Renal Adenoma (Kidney)	1/1
Renal cell carcinoma (Kidney)	1/1
Clear cell carcinoma (Kidney)	43/52
Oncocytoma (Kidney)	0/1
Papillary cell carcinoma (Kidney)	6/7
Chromophobe carcinoma (Kidney)	0/3
Medullary carcinoma (Kidney)	0/1
Renal cell carcinoma, unclassified (Kidney)	5/9
Urothelial carcinoma (Kidney)	2/11
Carcinosarcoma (Kidney)	1/1
Adenocarcinoma (Kidney)	0/1
Squamous cell carcinoma (Kidney)	0/4
Undifferentiated carcinoma (Kidney)	1/1
Wilm's tumor (Kidney)	1/1
Nephroblastoma (Kidney)	2/2
Adenocarcinoma (Prostate)	0/2
Clear cell carcinoma (Uterus)	0/1
Endometrioid carcinoma (Uterus)	1/1
Leiomyoma (Uterus)	1/1
Leiomyosarcoma (Uterus)	1/1
Squamous cell carcinoma (Cervix)	0/1

Pathology [a]	# positive / total cases
Adenocarcinoma (Cervix)	0/1
Basal cell carcinoma (Skin)	0/1
Melanoma (Skin)	1/1
Squamous cell carcinoma (Skin)	0/1
Neurofibrosarcoma (Nerve)	1/1
Schwannoma (Nerve)	1/1
Diffuse large B-cell lymphoma (DLBCL)	5/10
Anaplastic large cell lymphoma (Lymph node)	1/1
Follicular lymphoma (Lymph node)	1/1
Hodgkin lymphoma (Lymph node)	1/1
Squamous cell carcinoma (Bladder)	0/1
Urothelial carcinoma (Bladder)	0/1
Giant cell tumor (Bone)	11/11
Osteosarcoma (Bone)	4/4
Ameloblastoma (Bone)	0/2
Chondrosarcoma (Bone)	3/3
Malignant fibrous histiocytoma (Bone)	2/2
Fibrosarcoma (Bone)	1/1
Metastatic adenocarcinoma (Bone)	0/1
Metastatic carcinoma, NOS (Bone)	0/3
Angiosarcoma (Soft tissue)	1/1
Liposarcoma (Soft tissue)	18/18
Chondroma (Soft tissue)	1/1
Chondrosarcoma (Soft tissue)	1/1
Lipoma (Soft tissue)	1/1
Fibroma (Soft tissue)	2/2
Fibrosarcoma (Soft tissue)	19/19
Dermatofibrosarcoma protuberans (DFSP) (Soft tissue)	3/3
Malignant fibrous histiocytoma (Soft tissue)	2/2
Rhabdomyosarcoma (Soft tissue)	1/1
Embryonal rhabdomyosarcoma (Soft tissue)	3/3
Alveolar rhabdomyosarcoma (Soft tissue)	2/4
Polymorphic rhabdomyosarcoma (Soft tissue)	3/3
Leiomyosarcoma (Soft tissue)	9/9
Synovial sarcoma (Soft tissue)	2/3
Epithelioid sarcoma (Soft tissue)	2/2
Spindle cell sarcoma (Soft tissue)	1/1
Clear cell sarcoma (Soft tissue)	1/1
Carcinosarcoma (Soft tissue)	1/1
Myxoma (Muscle)	1/1
Plasmacytoma (Extramedullary)	1/1
Mesothelioma (Mesothelium)	1/1
Pleural Solitary Fibrous Tumor (Mesothelium)	1/1

[a] Vimentin is expressed in many normal structures such as fibroblasts, endothelial cells, smooth muscle cells, myoepithelial cells, lymphocytes and macrophages, along with other structures of mesenchymal origin. When evaluating the above tumors, a positive or negative status was derived based on assessment of the tumor cells, therefore tumors deemed as negative may have staining in some or all of the aforementioned structures.

Precision

Precision studies for CONFIRM anti-Vimentin (V9) antibody were completed to demonstrate:

- Between lot precision of the antibody.
- Within run and between day precision on a BenchMark ULTRA instrument.
- Between instrument precision on the BenchMark GX, BenchMark XT, and BenchMark ULTRA instrument.
- Between platform precision between the BenchMark GX, 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 CONFIRM anti-Vimentin (V9) antibody were assessed by systematic review of the literature. The data gathered support the use of the device in accordance with its intended purpose.

REFERENCES

1. Goldblum JR, Weiss SW, Folpe AL. Enzinger and Weiss's Soft Tissue Tumors E-Book. Elsevier Health Sciences; 2013.
2. Ridge KM, Shumaker D, Robert A, et al. Methods for Determining the Cellular Functions of Vimentin Intermediate Filaments. Methods Enzymol. 2016;568:389-426.
3. Dabbs DJ. Diagnostic Immunohistochemistry Theranostic and Genomic Applications, 5th Edition. Vol 5. Amsterdam, Netherlands: Elsevier; 2019.
4. Danielsson F, Peterson MK, Caldeira Araújo H, et al. Vimentin Diversity in Health and Disease. Cells. 2018;7(10).
5. Sharma P, Alsharif S, Fallatah A, et al. Intermediate Filaments as Effectors of Cancer Development and Metastasis: A Focus on Keratins, Vimentin, and Nestin. Cells. 2019;8(5):497.
6. International Agency for Research on Cancer. WHO Classification of Tumours of Soft Tissue and Bone. 5th ed (Fletcher CDM ed.). IARC; 2020.
7. Heim-Hall J, Yohe SL. Application of Immunohistochemistry to Soft Tissue Neoplasms. Arch Pathol Lab Med. 2008;132(3):476-489.
8. Lin F, Liu H. Immunohistochemistry in Undifferentiated Neoplasm/Tumor of Uncertain Origin. Arch Pathol Lab Med. 2014;138(12):1583-1610.
9. Miettinen M, Lehto VP, Badley R, et al. Expression of Intermediate Filaments in Soft-Tissue Sarcomas. Int J Cancer. 1982;30(5):541-546.
10. Akgul M, Cheng L. Immunophenotypic and Pathologic Heterogeneity of Unclassified Renal Cell Carcinoma: A Study of 300 Cases. Hum Pathol. 2020;102:70-78.
11. Carson F, Hladik C. Histotechnology: A Self Instructional Text, 3rd edition. Hong Kong: American Society for Clinical Pathology Press; 2009.

12. Occupational Safety and Health Standards: Occupational exposure to hazardous chemicals in laboratories. (29 CFR Part 1910.1450). Fed. Register.
13. 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.
14. Roche PC, Hsi ED. Immunohistochemistry-Principles and Advances. Manual of Clinical Laboratory Immunology, 6th edition. (NR Rose Ed.) ASM Press, 2002.

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
D	Updates to Intended Use, Summary and Explanation, Principle of the Procedure, Material Provided, Materials Required but not Provided, Storage and Stability, Specimen Preparation, Warnings and Precautions, Staining Procedure, Negative Reagent Control, Positive Tissue Control, Staining Interpretation / Expected Results, Specific Limitations, Analytical Performance, Clinical Performance, References, Symbols, Intellectual Property, and Contact Information sections. Added BenchMark GX, XT, ULTRA, and ULTRA PLUS instruments. Added recommended protocols for OptiView DAB IHC Detection Kit and *ultra*View Universal DAB Detection Kit. Removed recommended protocols for ES and NexES IHC instruments. Removed recommended protocols for *i*VIEW DAB, AEC, Alkaline Phosphatase Red and Enhanced Alkaline Phosphatase Red detection kits.

INTELLECTUAL PROPERTY

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