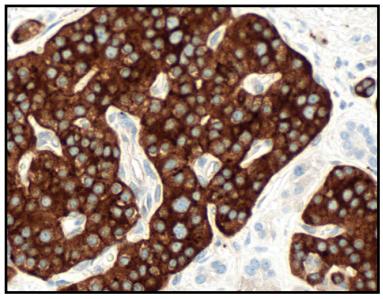
CONFIRM anti-Synaptophysin (SP11) Rabbit Monoclonal Primary Antibody

REF 790-4407



INTENDED USE

This antibody is intended for *in vitro* diagnostic (IVD) use. Ventana Medical Systems' (Ventana) CONFIRM anti-Synaptophysin (SP11) Rabbit Monoclonal Primary Antibody is designed to qualitatively detect the presence of synaptophysin expressing cells via light microscopy in formalin fixed, paraffin embedded tissue. Positive staining results may aid in the classification of neuroendocrine tumors. The clinical interpretation of any staining, or the absence of staining, must be complemented by morphological studies and evaluation of proper controls. Evaluation must be made by a qualified pathologist within the context of the patient's clinical history and other diagnostic tests.

SUMMARY AND EXPLANATION

Synaptophysin is a membrane glycoprotein present in neuroendocrine cells. In normal tissues, neuroendocrine cells of the human adrenal medulla, carotid body, skin, pituitary, thyroid, lung, pancreas, and gastrointestinal mucosa are labeled with this antibody. [1,2]

Anti-Synaptophysin is useful in the detection of normal neuroendocrine cells and neuroendocrine neoplasms. Reactive neoplasms of neural type include neuroblastomas, ganglio neuroblastomas, ganglioneuromas, pheochromocytomas, chromaffin and non-chromaffin paragangliomas. The antibody also labels neuroendocrine neoplasms of epithelial type including: pituitary adenomas, islet cell neoplasms, medullary thyroid carcinomas, parathyroid adenomas, carcinoids of the bronchopulmonary and gastrointestinal tracts, neuroendocrine carcinomas of the bronchopulmonary and gastrointestinal tracts, and neuroendocrine carcinomas of the skin.[3,4,5,6,7,8,9,10]

REAGENT PROVIDED

CONFIRM anti-Synaptophysin (SP11) contains sufficient reagent for staining 50 slides.

One 5 mL dispenser of CONFIRM anti-Synaptophysin (SP11) contains approximately 2.6µg of a rabbit monoclonal antibody produced as supernatant.

The antibody is diluted in Phosphate buffer with carrier protein and 0.05% ProClin 300, a preservative.

Total protein concentration of the reagent is approximately 19 mg/mL. Specific antibody concentration is approximately 0.5 µg/mL. There is no known irrelevant antibody reactivity observed in this product.

Refer to the appropriate Ventana detection kit package insert for detailed descriptions of: (1) Principles of the Procedure, (2) Materials and Reagents Needed but Not Provided, (3) Specimen Preparation, (4) Quality Control, (5) Troubleshooting, (6) Interpretation of Staining, and (7) General Limitations.

MATERIALS REQUIRED BUT NOT PROVIDED

Staining reagents such as Ventana detection kits (for example, *ultra*View Universal DAB detection kit), and ancillary components, including negative and positive tissue control slides, are not provided.

STORAGE

Store at 2-8ºC. Do not freeze.

To ensure proper reagent delivery and stability of the antibody, after every use the cap must be replaced and the dispenser must be immediately placed 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, formalin fixed, paraffin embedded tissues are suitable for use with this primary antibody when used with Ventana detection kits and a Ventana automated slide stainer. The recommended tissue fixative is 10% neutral buffered formalin.[11] Heat induced epitope retrieval with an EDTA based basic pH (~8.0) buffer is recommended. Slides should be stained immediately, as antigenicity of cut tissue sections may diminish over time.

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

WARNINGS AND PRECAUTIONS

1. For *in vitro* diagnostic use.
2. This product contains 2% or less bovine serum which is used in the manufacture of the antibody.
3. Avoid contact of reagents with eyes and mucous membranes. If reagents come in contact with sensitive areas, wash with copious amounts of water.
4. Avoid microbial contamination of reagents.
5. Consult local or state authorities with regard to recommended method of disposal.
6. The preservative in the reagent is ProClin 300. Symptoms of overexposure to ProClin 300 include skin and eye irritation, and irritation of mucous membranes and upper respiratory tract. The concentration of ProClin 300 in this product is less than or equal to 0.10% and does not meet the OSHA criteria for a hazardous substance. Systemic allergic reactions are possible in sensitive individuals.

STAINING PROCEDURE

Ventana primary antibodies have been developed for use on a Ventana automated slide stainer in combination with Ventana detection kits and accessories. A recommended staining protocol for a BenchMark XT instrument with *ultra*View Universal DAB detection kit (Cat. No. 760-500) is listed below in Table 1. The parameters for the automated procedures can be displayed, printed, and edited according to the procedure in the instrument's Operator's Manual. Refer to the appropriate Ventana detection package insert for more details regarding immunohistochemistry staining procedures.

Table 1. Recommended Staining Protocol for CONFIRM anti-Synaptophysin (SP11) with *ultra*View Universal DAB Detection Kit on BenchMark XT Instrument.

Procedure Type	Method
Deparaffinization	Selected
Cell Conditioning (Antigen Unmasking)	Mild Cell Conditioning 1
Enzyme (Protease)	None Required
Antibody (Primary)	Approximately 32 Minutes, 37°C
Counterstain	Hematoxylin II, 4 Minutes
Post Counterstain	Bluing Reagent, 4 Minutes

Due to variation in tissue fixation and processing, it may be necessary to increase or decrease the primary antibody incubation, cell conditioning or protease pretreatment based on individual specimens and detection used. For further information on fixation variables, refer to "Immunohistochemistry Principles and Advances".[12]

POSITIVE TISSUE CONTROL

An example of positive control tissue for CONFIRM anti-Synaptophysin (SP11) is normal pancreas (as depicted in the above image). Islet cells in normal pancreas should stain positively.

STAINING INTERPRETATION

The cellular staining pattern for CONFIRM anti-Synaptophysin (SP11) is cytoplasmic stain.

SPECIFIC LIMITATIONS

This antibody has been optimized for a 32 minute incubation time on a BenchMark XT automated slide stainer in combination with *ultra*View Universal DAB detection kit (Cat. No. 760-500). The user must validate results obtained with this reagent in their own lab. Clone SP11 was found to occasionally exhibit some diffuse granular cytoplasmic background staining in the exocrine pancreas.

PERFORMANCE CHARACTERISTICS

1. Immunoreactivity of CONFIRM anti-Synaptophysin (SP11) was determined by testing formalin fixed, paraffin embedded normal and neoplastic tissues.

   For normal tissues, results are as follows: adrenal gland (2/3), bone marrow (0/3), brain cerebrum (3/3), brain cerebellum (2/3), breast (0/3), cervix 0/3), colon (1/3), esophagus (0/3), heart (0/3), hypophysis (3/3), intestine (0/3), kidney (0/3), liver (0/3), lung (0/3), mesothelium (0/3), nerve (0/3), ovary (0/3), pancreas (2/3), parathyroid (0/3), prostate (0/3), salivary gland (0/3), skin (0/3), spleen (0/3), stomach (0/3), striated muscle (0/3), testis (0/3), thymus (0/3), thyroid (0/3), tonsil (0/3), and uterus (0/3).

   For neoplastic tissues, results are as follows: atypical meningioma (0/1), glioblastoma (1/1), ependymoma (0/1), oligodendroglioma (1/1), ovarian serous papillary adenocarcinoma (0/1), ovarian mucous papillary adenocarcinoma (0/1), islet cell carcinoma (1/1), pancreatic adenocarcinoma (0/1), testicular seminoma and embryonal carcinoma (0/2), medullary thyroid carcinoma (1/1), papillary thyroid carcinoma (0/1), intraductal, lobular, and infiltrating breast carcinoma (0/3), diffuse B-cell lymphoma in spleen (0/1), small cell lung carcinoma (1/1), squamous cell lung carcinoma (0/1), lung adenocarcinoma (0/1), esophageal squamous cell and adenocarcinoma (0/2), adenocarcinoma in stomach (0/1), intestinal adenocarcinoma and mesenchymoma (0/2), colorectal adenocarcinoma and mesenchymoma (0/4), hepatocellular carcinoma (0/1), hepatoblastoma (0/1), clear cell carcinoma (0/1), adenocarcinoma in prostate (0/1), transitional cell carcinoma in prostate and bladder (0/2), uterine leiomyoma (0/1), endometrial carcinoma (0/1), uterine clear cell and squamous carcinomas (0/3), embryonal rhabdomyosarcoma (0/1), rectal melanoma (0/1), basal cell carcinoma in skin (0/1), squamous cell carcinoma in skin (0/1), neurofibroma and neuroblastoma (1/2), mesothelioma (0/1), Hodgkin's lymphoma (0/1), diffuse type lymphoma (0/3), transitional cell carcinoma and leiomyosarcoma in smooth muscle (0/3), osteosarcoma (0/1), and spindle cell rhabdomyosarcoma (0/1).
2. Immunoreactivity of CONFIRM anti-Synaptophysin (SP11) was also evaluated by testing a variety of formalin fixed, parrafin embedded neoplastic neuroendocrine tissues. For neuroendocrine tissue, 288 samples were stained and evaluated for positivity. 129/288 neuroendocrine cases demonstrated positive staining with the antibody.
3. Inter-run reproducibility was determined by staining 5 replicate slides containing the same 3 tissues from duplicate sample types across a dynamic staining range (high expressing tumor, low expressing tumor and normal tissue) over 5 days on a BenchMark XT instrument. 148 of 150 tissues tested scored equivalently.
4. Intra-run reproducibility was determined by staining 14 replicate slides containing the same 3 tissues from duplicate sample types across a dynamic staining range on a BenchMark XT instrument. 83 of 84 tissues tested scored equivalently.
5. Intra-platform reproducibility was determined by staining 5 replicate slides containing the same 3 tissues from duplicate sample types across a dynamic staining range over 3 BenchMark XT instruments. 89 of 90 tissues tested scored equivalently.

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