NAME: Polyclonal Rabbit Anti-Neuron Specific Enolase (NSE)
SYNONYM: NSE
IMMUNOGEN: 90 kDa homodimer gamma/gamma from bovine brain

CODE NO.: N1516
Ready-to-Use DAKO® N-SERIES
Primary Antibody and Negative Control Reagent
For use with DAKO EnVision™, DAKO EnVision™ Doublestain and DAKO LSAB®2 Systems

INTENDED USE:
Polyclonal rabbit anti-Neuron Specific Enolase (NSE) (anti-NSE) is intended for laboratory use to identify qualitatively by light microscopy neuron specific enolase in normal and neoplastic tissues using immunohistochemical (IHC) test methods. Positive results aid in the differentiation of neural and neuroendocrine neoplasms. Differential identification is aided by the results from a panel of antibodies. Interpretation must be made within the context of the patient’s clinical history and other diagnostic tests by a qualified pathologist.

Refer to the General Instructions for immunohistochemistry (IHC) or the Detection System Instructions of IHC procedures for:

SUMMARY AND EXPLANATION:
Enolases are homo- or heterodimeric enzymes that catalyze the reaction pathway between 2-phosphoglycerate and phosphoenolpyruvate in the terminal step of anaerobic glycolysis. Dimers composed of two of three distinct subunits, alpha, beta and gamma. Neuron specific enolase, or gamma-gamma-enolase, is present in high concentrations in both neuronal and neuroendocrine cells and tumors derived from them. It is also present in non-nervous system tissues.1,2

Anti-NSE reacts with the human isoenzymes of neuron specific enolase which contain gamma–subunits. In Ouchterlony double diffusion between this antiserum and undiluted normal human serum no immune precipitate was seen.

REAGENTS PROVIDED:
The primary antibody is available in 7 mL and 22 mL volumes as rabbit antibody in 0.05 mol/L Tris-HCl, pH 7.6, containing stabilizing protein and 0.015 mol/L sodium azide.

The negative control reagent is available in 5 mL and 11 mL volumes as nonimmune rabbit serum in 0.05 mol/L Tris-HCl, pH 7.6, containing stabilizing protein and 0.015 mol/L sodium azide.

This product has been optimized for use in DAKO LSAB®2 Systems, DAKO EnVision™, or DAKO EnVision™ Doublestain detection systems.

The primary antibody and negative control reagents should be applied as directed in the Staining Procedure section of the Instructions included with each detection system. The recommended incubation time for this primary antibody is 10 minutes at room temperature.

MATERIALS REQUIRED, NOT SUPPLIED:
Refer to the General Instructions for immunohistochemistry and/or the Detection System Instructions.

PRECAUTIONS:
1. For In Vitro Diagnostic Use.
2. This product contains sodium azide (NaN₃), a chemical highly toxic in pure form. At product concentrations, though not classified as hazardous, build-ups of NaN₃ 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.³
3. Minimize microbial contamination of reagents or increase in nonspecific staining may occur.

STORAGE:
Store at 2-8°C

Do not use after expiration date stamped on vial. If reagents are stored under any conditions other than those specified, the conditions must be verified by the user.⁴

There are no obvious signs to indicate instability of this product. Therefore, positive and negative controls should be run simultaneously with patient specimens. If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem with the antibody is suspected, contact DAKO Technical Services.

SPECIMEN PREPARATION:
Paraffin Sections: Anti-NSE can be used on formalin-fixed, paraffin-embedded tissue sections. Pretreatment of tissue with proteolytic enzymes is not recommended.

Cryostat Sections And Cell Smears: Anti-NSE can be used for labelling acetone-fixed cryostat sections or fixed cell smears.

STAINING PROCEDURE:
Follow the procedure for the detection system selected. Use the recommended incubation time presented in the “Reagents Provided” section.

PRODUCT SPECIFIC LIMITATIONS:
As with all polyclonal and monoclonal antibodies to NSE, caution should be exercised in the interpretation of the staining results obtained with anti-NSE as gamma-subunits of enolases are not confined to neuronal tissue.⁵ The types of fixation⁵,⁷ or the use/absence of enzymatic pretreatment of sections further complicate the interpretation of staining.⁸
PERFORMANCE CHARACTERISTICS:
The cellular staining pattern for anti-NSE is cytoplasmic.

**Normal tissues:** Similar to the findings with monoclonal\textsuperscript{6,9} and other polyclonal antibodies\textsuperscript{13,17} to the gamma-subunit of enolase, anti-NSE is showing a complex distribution of this subunit in neuronal and nonneuronal tissues. In these reports, tissues found to be positive for the gamma-subunit include the brainstem, cerebral cortex, central and peripheral neurons, and their processes (except Purkinje’s cells) including myelinated and unmyelinated nerves, astrocytes and ganglions, adrenal medulla, megakaryocytes and platelets, pancreatic islets, pituitary adenohypophysis and smooth muscle.

No immunoreactivity was detected by use of polyclonal \(\gamma\)-subtype specific antiserum\textsuperscript{12} in the lung, heart, liver, kidney, spleen, lymph nodes and skeletal muscle.

**Abnormal tissues:** The inconsistency in the expression of NSE by use of both monoclonal\textsuperscript{6,9,12,14-16} and polyclonal antibodies\textsuperscript{6,7,10,13,16-18} to the gamma-isoenzyme requires caution in the interpretation of positive staining. The above authors have reported NSE in neuronal and neuroendocrine tumors including various neuroblastoma, parangangioma, melanoma, small cell carcinoma, islet cell tumor, Merkel cell and medullary carcinoma, gastroenteropancreatic tumors and carcinoid tumors. Nonneural and nonendocrine tumors included miscellaneous carcinomas, sarcomas, lymphomas and meningiomas.

Consistently negative when tested with specific polyclonal antibodies\textsuperscript{10-13,17} to \(\gamma\)-enolase were cases of meningioma,\textsuperscript{10} epithelioid sarcoma,\textsuperscript{2} squamous cell carcinoma,\textsuperscript{2} basal cell carcinoma,\textsuperscript{2} large bowel adenocarcinoma\textsuperscript{2} and intraductal carcinoma.\textsuperscript{2}

**REFERENCES:**