



DATA SHEET

Code **M363501**

Intended use

For In Vitro Diagnostic Use.

Monoclonal Rabbit Anti-Human Cyclin D1, Clone SP4, is intended for laboratory use in immunohistochemistry. This antibody is useful for the identification of mantle cell lymphoma (1-3). The clinical interpretation of any staining or its absence should be complemented by morphological studies using proper controls and should be evaluated within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist.

Synonyms for antigen

PRAD-1, CCND1, Bcl-1 (4)

Summary and explanation

Cyclin D1 is a 36 kD protein encoded by the CCND1 (bcl-1) gene located on chromosome 11q13. Cyclin D1 associates with and activates cyclin-dependent kinases (CDKs) CDK4 and CDK6 (4-6). The protein functions as a CDK-dependent regulator of the cell cycle by phosphorylating and inactivating the retinoblastoma protein, thereby allowing for progression through the G1-S phase of the cell cycle (4,6). Cyclin D1 is also involved in cell cycle or CDK-independent functions including associating with and regulation of a variety of transcription factors and transcriptional coregulators. Cyclin D1 has also been shown to be involved in the regulation of cellular metabolism, fat cell differentiation and cell migration (4,6). Cyclin D1 overexpression is associated with tumorigenesis, and Cyclin D1 amplification and/or overexpression have been demonstrated in a variety of human tumors, including mantle cell lymphomas, breast carcinomas, head and neck squamous cell carcinomas and esophageal cancers (4).

Reagent provided

Monoclonal Rabbit antibody provided in liquid form as cell culture supernatant and containing 0.015 mol/L sodium azide. This product contains stabilizing protein.

Clone: SP4 Rabbit Ig concentration mg/L: See label on vial.

Immunogen A synthetic peptide from C-terminus of human Cyclin D1

Specificity

In Western blotting of EL4 cell lysates the antibody labels a major band of approximately 36 kD, corresponding to the expected molecular weight of Cyclin D1 (5,7).

Precautions

1. For professional users.
2. This product contains sodium azide (NaN₃), a chemical highly toxic in pure form. At product concentrations, though not classified as hazardous, sodium azide may react with lead and copper plumbing to form highly explosive build-ups of metal azides. Upon disposal, flush with large volumes of water to prevent metal azide build-up in plumbing.
3. As with any product derived from biological sources, proper handling procedures should be used.
4. Wear appropriate Personal Protective Equipment to avoid contact with eyes and skin.
5. Unused solution should be disposed of according to local, State and Federal regulations.

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. 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 suspect contact us.

Specimen preparation including materials required but not supplied

Paraffin sections:

The antibody can be used for labeling paraffin-embedded tissue sections fixed in formalin or B5 (2).

Pre-treatment of deparaffinized tissue sections with heat-induced epitope retrieval (HIER) is required. Optimal results are obtained by pretreating tissues with HIER using Dako Target Retrieval Solution, pH 9 (Codes S2368/S2367).

Waterbath HIER: 20 minutes at 95–99 °C. Preheat target retrieval solution prior to immersing slides

After thermal treatment, allow the jar with buffer and slides to cool for 20 minutes at room temperature. Rinse sections gently with buffer or deionized water. The tissue sections should not dry out during the treatment or during the following immunohistochemical staining procedure. For greater adherence of tissue sections to glass slides.

Frozen sections and cell smears:

The antibody can be used for labeling acetone-fixed, frozen sections or fixed cell smears.

Staining procedure including materials required but not supplied

Dilution: Monoclonal Rabbit Anti-Human Cyclin D1, Code M3635, may be used at a dilution range of 1:50-1:100 when applied on pretreated, formalin-fixed, paraffin-embedded sections using a 30 minute incubation at room temperature. It is recommended that the antibody is diluted. These are

guidelines only. Optimal conditions may vary depending on specimen and preparation method, and should be determined individually by each laboratory. The recommended negative control Rabbit Immunoglobulin Fraction (Solid-Phase Absorbed) (Code X0936), diluted to the same rabbit IgG concentration as the primary antibody. Unless the stability of the diluted antibody and negative control has been established in the actual staining procedure, it is recommended to dilute these reagents immediately prior to use. Positive and negative controls should be run simultaneously with patient specimens.

Staining interpretation

The cellular staining pattern is predominantly nuclear.

Performance characteristics

Normal tissues (8):

Tissue Type (# tested) Positively Staining Tissue Elements

Bone Marrow (2) 0/2

Breast (2) 2/2 Ductal epithelial cells, (1%) nuclear and (25%) nuclear and cytoplasmic

Cervix (2) 1/2 Basal epithelial cells (30%), nuclear

Mesothelial Cells (3) 0/3

Nerve (2) 0/2

Ovary (3) 0/3

Parathyroid (3) 1/3 Glandular epithelial cells (<1%), nuclear

Prostate (2) 1/2 Glandular epithelial cells (10%), nuclear

Salivary gland (3) 1/3 Glandular epithelial cells, (50%) nuclear and (5%) nuclear and cytoplasmic

Small intestine (3) 1/3 Glandular epithelial cells and macrophages (1%), nuclear Thyroid (2) 0/2

Tonsil (3) 2/3 Basal epithelial cells (80%), nuclear

Uterus (2) 1/2 Stromal cells (1%), nuclear

1/2 Surface epithelial cells (5%), nuclear

Abnormal tissues (1,3):

Tissue Type (# tested) Positively Staining Tumors

Mantle cell lymphoma (42) 1,3 42/42

Hairy cell leukemia (5) 3 2/5

Chronic lymphocytic leukemia/small lymphocytic lymphoma (27) 1,3, 2/27

Diffuse large B-cell lymphoma (25),3, 0/25

Extranodal marginal zone lymphoma of MALT(14),3,0/14

Follicular lymphoma (16),1,3, 0/16

Marginal zone lymphoma (9),1,3, 0/9

Lymphoplasmacytic lymphoma (1),3, 0/1

Lymphoblastic lymphoma (1 T and 1 B lineage) (2),3,0/2

Extranodal NK/T-cell lymphoma (13),3, 0/13

Peripheral T-cell lymphoma, NOS (6),3, 0/6

Anaplastic large cell lymphoma (1),3, 0/1

Angioimmunoblastic T-cell lymphoma (1),3, 0/1

Multiple Myeloma (14),1,3,5/14

Hodgkin lymphoma (4),3, 0/4

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