

# CD25 (4C9)

## Mouse Anti-Human CD25 Monoclonal Antibody (4C9)

**References and presentations<sup>1</sup>** 

- ready-to-use (manual or LabVision AutoStainer)
  MAD-000155QD-3
  MAD-000155QD-7
  MAD-000155QD-12
- Ready-to-use (MD-Stainer)<sup>2</sup> MAD-000155QD-3/V MAD-000155QD/V
- concentrated MAD-000155Q - 1:50 recommended dilution

**Composition**: anti-CD25 mouse monoclonal antibody obtained from supernatant culture and prediluted in a tris buffered solution pH 7.4 containing 0.375mM sodium azide solution as bacteriostatic and bactericidal. **Intended use D**: Immunohistochemistry (IHC) on paraffin embedded tissues. Not tested on frozen tissues or Western-Blotting

**Clone:** 4C9

**Immunogen:** Recombinant protein corresponding to the external domain of the human IL-2R

**Ig isotype:** Mouse IgG<sub>2b</sub>

**Species reactivity:** In vitro diagnostics in humans. Not tested in other species

**Description and aplications:** The CD25 molecule, also known as IK-2 receptor alpha, IL2RA, p55, T-cell growth factor receptor (TCGFR) or TAC antigen is an activation antigen present, along with CD4, in regulatory T cells. The gene that controls its expression, with 8 exons and over 25 kb, codes the alpha subunit of the cell surface receptor IL-2 and is located in the chromosome region 10p15.1.

The regulatory T cells, since they suppress the activation of autoreactive T cells controlling the immune tolerance, prevent autoimmune diseases and, as negative collateral effects, avoid the destruction of tumor cells by cytotoxic T lymphocytes and act as suppressors of the NK cells. Partial deletions of the CD25 gene are responsible for immunodeficiency 41 (characterized by the association of various lymphoproliferative syndromes with autoimmune diseases) and insulin-dependent diabetes mellitus type 10, a variant of diabetes mellitus type I associated with autoimmune diseases and with typical familial aggregation.

In normal tissues, CD25 can be expressed by activated B and T lymphocytes, macrophages and osteoblasts. Some thymocytes, myeloid precursors and oligodendrocytes can also show immunostaining. This molecule is not expressed in normal mastocytes.

According to the classification system of the World Health Organization, the main diagnostic criterion for the involvement of bone marrow by systemic mastocytosis (SM) is the presence of dense aggregates (more than 15 cells) of mastocytes. For this reason, the aberrant expression of CD25 as a low affinity receptor for interleukin-2 (IL-2) by neoplastic mast cells is a good diagnostic tool to distinguish them from reactive proliferations of mast cells, and for this reason it has recently become a minor criterion for the diagnosis of SM, where aberrant staining of mastocytes aggregates by anti-CD25 antibody is diagnostic of SM.

The anti-CD25 antibody has also been useful for the identification of mastocytes on skin biopsies in the context of urticaria pigmentosa as a predictor of systemic mastocytosis.

Additionally, the quantification of regulatory T-cells (Treg) expressing CD25 in the context of hepatocellular carcinoma has been used as an independent predictor of tumor recurrence following liver resection of a previous hepatocellular carcinoma. In addition, the percentage of regulatory T cells FOXP3+ CD25+ infiltrating between melanoma tumor cells and in their periphery is significantly higher in melanomas with recurrent capacity than in their non-recurrent forms.

Finally, CD25 together with CD103 and CD123 is useful for completing the panel of markers of hairy cell leukemia, although the latter two antibodies have greater specificity and sensitivity for this diagnosis than CD25 itself.

Similarly, due to its general low specificity, the CD25 antibody's positive staining should be evaluated within an antibody panel, not in isolation, and in correlation with the remaining morphological aspects of the lesions analyzed since many B lymphomas, T lymphomas, or even anaplastic large cell lymphoma may present staining against this marker.





<sup>&</sup>lt;sup>1</sup> These references are for presentation in vials of Low Density Polyethylene (LDPE) dropper. In case the products are used in automated stainers, a special reference is assigned as follows:

 <sup>/</sup> L: Cylindrical screw-cap vials (QD-3 / L, QD-7 / L, QD-12 / L).
/ N: Polygonal screw-cap vials (QD-3 / N, QD-7 / N, QD-12 / N).

For different presentations (references / volumes) please contact the supplier.

 $<sup>^2</sup>$  For Technical specifications for MD-Stainer, please contact your distributor.



**IHC positive control**: Tissue section from hairy cell leukemia.

Visualization: Membrane and cytoplasm

#### IHC recommended procedure:

- 4µm thick section should be taken on charged slides; dry overnight at 60<sup>o</sup>
- Deparaffinise, rehydrate and HIER (heat induced epitope retrieval) boil tissue in the Pt Module using Vitro S.A EDTA buffer pH8<sup>3</sup> for 20 min at 95°C. Upon completion rinse with 3-5 changes of distilled or deionised water followed by cooling at RT for 20 min
- Endogenous peroxidase block Blocking for 10 minutes at room temperature using peroxidase solution (ref. MAD-021540Q-125)
- Primary antibody: incubate for 20 minutes [The antibody dilution (when concentrated) and protocol may vary depending on the specimen preparation and specific application. Optimal conditions should be determined by the individual laboratory]
- For detection use Master Polymer Plus Detection System (HRP) (DAB included; ref. MAD-000237QK)
- Counterstaining with haematoxylin and final mounting of the slide

**Storage and stability:** If Stored at 2-8°C. Do not freeze. Once the packaging has been opened it can be stored until the expiration date of the reagent indicated on the label. If the reagent has been stored under other conditions to those indicated in this document, the user must first check its correct performance taking into account the product warranty is no longer valid.

#### Warnings and precautions:

1. Avoid contact of reagents with eyes and mucous membranes. If reagents come into contact with sensitive areas, wash with copious amounts of water.

2. This product is harmful if swallowed.

3. Consult local or state authorities with regard to recommended method of disposal.

4. Avoid microbial contamination of reagents.

#### SAFETY RECOMMENDATIONS

This product is intended for laboratory professional use only. The product is NOT intended to be used as a drug or for domestic purposes. The current version of the Safety Data Sheet for this product can be downloaded by searching the reference number at <u>www.vitro.bio</u> or can be requested at <u>regulatory@vitro.bio</u>.

#### BIBLIOGRAPHY

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<sup>3</sup> Ref: MAD-004072R/D







### LABEL AND BOX SYMBOLS

Explanation of the symbols of the product label and box:

$\Box$	Expiration date
Ĵ.	Temperature limit
***	Manufacturer
X	Sufficient content for <n></n>
	assays
REF	Catalog number
LOT	Lot code
Ĩ	Refer to the instructions of
	use
IVD	Medical product for in
	<i>vitro</i> diagnosis.
e-SDS	Material safety data sheet



