Novocastra™ Liquid Mouse Monoclonal Antibody Calcitonin

Product Code: NCL-L-CALCITONIN

Intended Use	FOR RESEARCH USE ONLY
Specificity	Human calcitonin. Does not cross react with calcitonin gene related peptide (CGRP).
Clone	CL1948
Ig Class	IgG2b
Antigen Used for Immunizations	Prokaryotic recombinant fusion protein containing 32 amino acids corresponding to the mature human calcitonin molecule.
Hybridoma Partner	Mouse myeloma (p3-NS1-Ag4.1).
Preparation	Liquid tissue culture supernatant containing 15 mM sodium azide. Volume as indicated on vial label.
Effective on Frozen Tissue	Not evaluated
Effective on Paraffin Wax Embedded Tissue	Yes (using 5 minutes proteinase K enzyme digestion: see overleaf).
Recommendations on Use	Immunohistochemistry: Typical working dilution 1:200. 5 minutes proteinase K digestion. 30 minutes primary antibody incubation at 25 °C. Polymer detection recommended. Western Blotting: Not recommended.
Positive Controls	Immunohistochemistry: Thyroid gland (C cells).
Staining Pattern	Cytoplasmic
Storage and Stability	Store liquid antibody at 4 °C. Under these conditions, there is no significant loss in product performance up to the expiry date indicated on the vial label. Prepare working dilutions on the day of use.
General Overview	Calcitonin (CT) is a 32 amino acid peptide synthesized by the parafollicular C-cells of the thyroid. It acts through its receptors to inhibit osteoclast mediated bone resorption, decrease calcium resorption by the kidney and decrease calcium absorption by the intestines. The action of calcitonin is therefore to cause a reduction in serum calcium, an effect opposite to that of parathyroid hormone. The calcitonin gene transcript also encodes the calcitonin gene-related peptide (CGRP), which is thought to be a potent vasodilator. The tissue specificity of the transcript produced depends on alternative splicing of the CT/CGRP gene transcript. In the parafollicular cells of the thyroid 95 % of the CT/CGRP RNA is processed and translated to produce CT, however, in neuronal cells 99 % of the CT/CGRP RNA is translated into CGRP. The C cells of the thyroid give rise to an endocrine tumor, medullary thyroid carcinoma (MTC), which occurs in a sporadic (75 % of cases) and hereditary form (25 % of cases). Familial MTC is associated with C cell hyperplasia (CCH), whereas sporadic MTC is thought not to be. However, in the general population CCH is present in 20–30 % of thyroid glands, either with normal histology, thyroiditis or follicular tumors.
General References	Leboulleux S, Baudin E, Travagli J et al. Clinical Endocrinology. 2004; 61:299–310. Hirsch P, Lester G, Talmage R. Journal of Musculoskeletal & Neuronal Interactions. 2001; 1(4):299–305. Pondel M. International Journal of Experimental Pathology. 2000; 81:405–422.

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Instructions for Use

Enzyme Digestion for Immunohistochemical Demonstration on Paraffin Sections

Prior to undertaking this methodology, users must be trained in immunohistochemical techniques.

Customers should determine optimal dilutions for antibodies. Unless indicated, all steps are performed at room temperature (25 °C)

1. Cut and mount sections on slides coated with a suitable tissue adhesive.
2. De-paraffinize sections in xylene or xylene substitutes.
3. Re-hydrate through graded alcohols.
4. Wash slides in running tap water.

Pretreat the sections as follows:

5. Wash slides in d H_2O
6. Incubate in Enzyme Proteinase K (RE7330-K) at 25 °C for 5 minutes (or alternative time if this is indicated in the Recommendations on Use).
7. Wash in TBS.
8. Proceed with IHC protocol according to manufacturer's Instructions for Use for the primary antibody and detection system.

Reagents required but not supplied

1. Standard solvents used in immunohistochemistry.
2. 50 mM Tris-buffered saline (TBS) pH 7.6
3. Antibody diluent, Novocastra IHC Diluent, RE7189
4. Visualization system, Novolink™ Polymer Detection Systems.
5. Mounting medium - use as recommended by manufacturer.

E. Amendments to Previous Issue

Not applicable.

F. Date of Issue

13 November 2009 (RUOprotocol/Proteinase K).