

Novocastra™ Lyophilized Rabbit Polyclonal Antibody Calcitonin

Product Code: NCL-CALp

Intended Use	FOR RESEARCH USE ONLY.
Specificity	Human calcitonin. Also reacts with mouse, rat, dog, horse, sheep and swine calcitonin.
Antigen Used for Immunizations	Synthetic human calcitonin 1-32 amino acid peptide coupled to bovine serum albumin.
Preparation	Lyophilized immunoglobulin fraction purified from rabbit serum diluted in PBS with 1% BSA containing 15 mM sodium azide. Reconstitute with the volume of sterile distilled water indicated on the vial label.
Effective on Frozen Tissue	Yes
Effective on Paraffin Wax Embedded Tissue	Yes
Recommendations on Use	Immunohistochemistry: Typical working dilution 1:100–1:200. Trypsin digestion of paraffin sections may be required in some cases. 60 minutes primary antibody incubation at 25 °C. Standard ABC technique. Western Blotting: Not evaluated.
Positive Controls	Immunohistochemistry: C cells of thyroid.
Staining Pattern	Cytoplasmic
Storage and Stability	Store unopened lyophilized 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. The reconstituted antibody is stable for at least two months when stored at 4 °C. For long term storage, it is recommended that aliquots of the antibody are frozen at -20 °C (frost-free freezers are not recommended). Repeated freezing and thawing must be avoided. Prepare working dilutions on the day of use.
General Overview	Calcitonin is a peptide hormone synthesized by the parafollicular cells of the thyroid. It causes reduction in serum calcium, an effect opposite to that of parathyroid hormone (PTH). Human calcitonin contains 32 amino acids and has a molecular weight of 3,421 kD. Multiple calcitonin polypeptides are encoded in a single messenger RNA. Evidence exists that indicates alternative RNA splicing of the transcripts of the calcitonin gene is responsible for the production of different polypeptide products. Genomic mapping results are consistent with the existence of a single calcitonin gene.
General References	Holm R, Nesland J M, Attramadal A, et al.. Journal of Pathology. 158: 213–217 (1989). Rosenfeld M G, Lin C R, Amara S G, et al.. Proceedings of the National Academy of Sciences USA. 79 (6): 1717–1721 (1982). Jacobs J W, Goodman R H, Chin W W, et al.. Science. 213 (4506): 457–459 (1981). Talerman A, Lindeman J, Kievit-Tyson P A, et al.. Histopathology. 3: 503–510 (1979).



Instructions for Use

Trypsin Digestion for Immunohistochemical Demonstration on Paraffin Sections

1. Preheat the following to 37 °C using a water bath:
 - (i) 200 mL of TBS
 - (ii) 200 mL of distilled water.
2. Dissolve 0.2 g Trypsin 250 and 0.2 g Calcium chloride in the 200 mL of TBS.
3. Once the Trypsin solution is at 37 °C, pH to 7.8 with 1 M sodium hydroxide.
4. Place rehydrated paraffin sections in the distilled water to preheat the sections to 37 °C for a minimum of 5 minutes.
5. Incubate sections in Trypsin solution at 37 °C. The time required will depend on the antibody and tissue, however, 30 minutes is usually sufficient.
6. Rinse sections in running tap water.
7. Proceed with immunohistochemistry protocol.

Reagents Required but not Supplied

50 mM Tris-buffered saline

Trypsin 250: Difco order code 0152-13 (available from Becton Dickinson).

Calcium chloride

1 M Sodium Hydroxide

** Trypsin containing chymotrypsin should always be used. The enzyme activities can vary from a supplier and between batches. Such variations may affect the incubation time required.*