

Novocastra™ Lyophilized Mouse Monoclonal Antibody Thyroid Stimulating Hormone

Product Code: NCL-TSH

Intended Use	For In Vitro Diagnostic Use: This product is intended for qualitative immunohistochemistry with normal and neoplastic formalin-fixed, paraffin-embedded tissue sections, to be viewed by light microscopy.
Specificity	Thyroid stimulating hormone (TSH). No reactivity with luteinizing hormone, follicle stimulating hormone and human chorionic gonadotrophin.
Clone	QB2/6
Ig Class	IgG1
Antigen Used for Immunizations	TSH molecule.
Hybridoma Partner	Mouse myeloma (NS0).
Preparation	Lyophilized tissue culture supernatant 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 1:50–1:100. Trypsin digestion of paraffin sections is recommended. 60 minutes primary antibody incubation at 25 °C. Standard ABC technique. Western Blotting: Not evaluated.
Positive Controls	Immunohistochemistry: Normal human pituitary gland.
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	Thyroid stimulating hormone (TSH or thyrotropin) is a glycoprotein produced in the thyrotrophs of the anterior pituitary gland. TSH and TSH receptor (TSHR) are key proteins in the control of thyroid function. TSH synthesis in the anterior pituitary is stimulated by thyrotropin-releasing hormone (TRH) and inhibited by thyroid hormone in a classical endocrine negative-feedback loop. TSH controls thyroid function upon its interaction with the G protein-coupled TSHR. TSH binding to its receptor on thyroid cells lead to the stimulation of second messenger pathways involving predominantly cAMP, inositol 1,4,5-triphosphate (IP3) and diacylglycerol (DAG), ultimately resulting in the modulation of thyroidal gene expression. TSH also acts as a factor protecting thyroid cells from apoptosis and plays a critical role in ontogeny.
General References	Fares F A, Levi F, Reznick A Z, et al.. Journal of Biological Chemistry. 276: 4543–4548 (2001). Oda Y, Sanders J, Evans M, et al.. Thyroid. 10: 1051–1059 (2000). Haugen B R, Pacini F, Reiners C, et al.. Journal of Clinical Endocrinology and Metabolism. 84: 3877–3885 (1999). Beck-Peccoz P, Brucker-Davis F, Persani L, et al.. Endocrine Reviews. 17: 610–638 (1996). Meier C A, Braverman L E, Ebner S A, et al.. Journal of Clinical Endocrinology and Metabolism. 78: 188–196 (1994). Cole E S, Lee K, Lauziere K, et al.. Biotechnology. 11: 1014–1024 (1993).



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.*