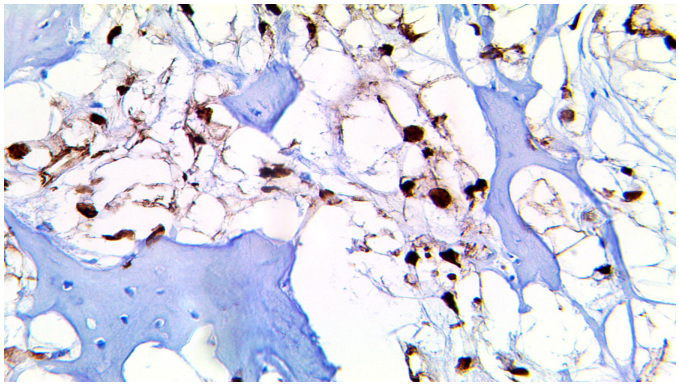
Brachyury

Clone: RBT-TBXT
Rabbit Monoclonal

CE IVD



Inset: IHC of Brachyury on a FFPE Chordoma Tissue

Antibody Type	Rabbit Monoclonal	**Clone**	RBT-TBXT
Isotype	IgG	**Reactivity**	Paraffin, Frozen
Localization	Nuclear	**Species Reactivity**	Human
Control	Testis, Chordoma		
Application	Neural & Neuroendocrine Cancer, Liver Cancer		

Intended Use
For In Vitro Diagnostic Use.

This antibody is intended for use in Immunohistochemical applications on formalin-fixed paraffin-embedded tissues (FFPE), frozen tissue sections and cell preparations. Interpretation of results should be performed by a qualified medical professional.

Immunogen
Synthetic peptide corresponding to the N-terminus of the human brachyury protein.

Summary and Explanation
Brachyury is a protein that in humans is encoded by the T gene. Brachyury is a transcription factor within the T-box complex of genes which appears to have a conserved role in defining the midline of a bilaterian organism and thus the establishment of the anterior-posterior axis; this function is apparent in chordates and molluscs. The number of cervical vertebrae is highly conserved among all mammals; however a spontaneous vertebral and spinal dysplasia (VSD) mutation in this gene has been associated with the development of six or fewer cervical vertebrae instead of the usual seven.

Expression of the brachyury gene has been identified as a definitive diagnostic marker of chordoma, a malignant tumor that arises from remnant notochordal cells lodged in the vertebrae. Furthermore, germ line duplication of brachyury confers major susceptibility to chordoma. The chromosomal region on 6q27 containing the brachyury gene was gained in 6 of 21 chordomas (29%), and none of the 21 chordomas analyzed showed deletions that could have affected this gene.

Brachyury is an important factor in promoting the epithelial-mesenchymal transition (EMT). Cells that over-express brachyury have down-regulated expression of the adhesion molecule E-cadherin, which allows them to undergo EMT. Overexpression of brachyury has been linked to Hepatocellular carcinoma. While brachyury is promoting EMT, it can also induce metastasis of HCC cells. Brachyury expression is a prognostic biomarker for HCC, and the gene may be a target for cancer treatments in the future. Additionally, overexpression of brachyury may play a part in EMT associated with benign disease such as renal fibrosis.

Presentation
Anti-Brachyury is a Rabbit Monoclonal antibody derived from cell culture supernatant that is concentrated, dialyzed, filter sterilized and diluted in buffer pH 7.5, containing BSA and sodium azide as a preservative.

Catalog No.	*Presentation*	*Dilution*	*Volume*
BSB 3490	Predilute	Ready-to-Use	3.0 mL
BSB 3491	Predilute	Ready-to-Use	7.0 mL
BSB 3492	Predilute	Ready-to-Use	15.0 mL
BSB 3493	Concentrate	1:25-1:100	0.1 mL
BSB 3494	Concentrate	1:25-1:100	0.5 mL
BSB 3495	Concentrate	1:25-1:100	1.0 mL

Control Slides Available

Catalog No.	*Quantity*
BSB-9036-CS	5 slides

Storage Store at 2-8°C (Control Slides: Store at 20-25°C)

Precautions
1. For professional users only. Results should be interpreted by a qualified medical professional.
2. This product contains <0.1% sodium azide (NaN_3) as a preservative. Ensure proper handling procedures are used with this reagent.
3. Always wear personal protective equipment such as a laboratory coat, goggles, and gloves when handling reagents.
4. Dispose of unused solution with copious amounts of water.
5. Do not ingest reagent. If reagent is ingested, seek medical advice immediately.
6. Avoid contact with eyes. If contact occurs, flush with large quantities of water.
7. Follow safety precautions of the heating device used for epitope retrieval (TintoRetriever Pressure Cooker or similar).
8. For additional safety information refer to the Safety Data Sheet for this product.
9. For complete recommendations for handling biological specimens, please refer to the CDC document, "Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories" (see References in this document).

Stability
This product is stable up to the expiration date on the product label.
Do not use the after expiration date listed on the package label. Temperature fluctuations should be avoided. Store appropriately when not in use and avoid prolonged exposure to room temperature conditions.

Specimen Preparation

Paraffin sections: The antibody can be used on formalin-fixed paraffin-embedded (FFPE) tissue sections. Ensure tissue undergoes appropriate fixation for best results. Pre-treatment of tissues with heat-induced epitope retrieval (HIER) is recommended using Bio SB ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023), ImmunoDNA Retriever with EDTA (BSB 0030-BSB 0033), or ImmunoDNA Digestor (BSB 0108-0112). See reverse side for complete protocol. Tissue should remain hydrated via use of Bio SB Immuno/DNA Washer solutions (BSB 0029 & BSB 0042).

Frozen sections and cell preparations: The antibody can be used on acetone-fixed frozen sections and acetone-fixed cell preparations.

IHC Protocol

1. Cut and mount 3-5 micron formalin-fixed paraffin-embedded tissues on positively charged slides such as Bio SB Hydrophilic Plus Slides (BSB 7028).
2. Air dry for 2 hours at 58° C.
3. Deparaffinize, dehydrate, and rehydrate tissues.
4. Subject tissues to heat induced epitope retrieval (HIER) using a suitable retrieval solution such as ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023) or EDTA (BSB 0030-BSB 0033).
5. Any of three heating methods may be used:

a. TintoRetriever Pressure Cooker or Equivalent

Place tissues/slides in a staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA and place on trivet in the pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high. Incubate for 15 minutes. Open and immediately transfer slides to room temperature.

b. TintoRetriever PT Module or Water Bath Method

Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA at 95°-99° C. Incubate for 30-60 minutes.

c. Conventional Steamer Method

Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA in a steamer, cover and steam for 30-60 minutes.

6. After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15-20 minutes.
7. For manual IHC, perform antibody incubation at ambient temperature. For automated IHC methods, perform antibody incubation according to instrument manufacturer's instructions.
8. Wash slides with ImmunoDNA washer or DI water.
9. Continue IHC protocol. Wash slides between each step with ImmunoDNA washer solution.

Abbreviated Immunohistochemical Protocol

Step	ImmunoDetector AP/HRP	PolyDetector AP/HRP	PolyDetector Plus HRP
Peroxidase/AP Blocker	5 min.	5 min.	5 min
Primary Antibody	30-60 min.	30-60 min.	30-60 min.
1st Step Detection	10 min.	30-45 min.	15 min.
2nd Step Detection	10 min.	Not Applicable	15 min.
Substrate- Chromogen	5-10 min.	5-10 min.	5-10 min.
Counterstain / Coverslip	Varies	Varies	Varies

Mounting Protocols

For detailed instructions using biodegradable permanent mounting media such as XyGreen PermaMounter (BSB 0169-0174) or organic solvent based resin such as PermaMounter (BSB 0094-0097), refer to PI0174 or PI0097.

Product Limitations

Due to inherent variability present in immunohistochemical procedures (including fixation time of tissues, dilution factor of antibody, retrieval method utilized, and incubation time), optimal performance should be established through the use of positive and negative controls. Results should be interpreted by a qualified medical professional.

References

1. Entrez Gene: T T brachyury transcription factor [Homo sapiens (human)] https://www.ncbi.nlm.nih.gov/gene?Db=gene&Cmd=ShowDetailView&TermToSearch=6862
2. Edwards YH, et al. The human homolog T of the mouse T (Brachyury) gene; gene structure, cDNA sequence, and assignment to chromosome 6q27". Genome Res. 1996; 6 (3): 226–33.
3. Scholz CB and Technau U. The ancestral role of Brachyury: expression of NemBra1 in the basal cnidarian Nematostella vectensis (Anthozoa). Dev. Genes Evol. 2003; 212 (12): 563–70.
4. Le Gouar, M, et al. Expression of a SoxB and a Wnt2/13 gene during the development of the mollusc Patella vulgata. Development genes and evolution. 2004; 214(5): 250–256.
5. Lartillot, N; at al. Expression pattern of Brachyury in the mollusc Patella vulgata suggests a conserved role in the establishment of the AP axis in Bilateria.". Development. 2002; 129 (6): 1411–1421.
6. Kromik A, et al. The mammalian cervical vertebrae blueprint depends on the T (brachyury) Gene. Genetics. 2015; 199 (3):183+.
7. Vujovic S, et al. Brachyury, a crucial regulator of notochordal development, is a novel biomarker for chordomas. J. Pathol. 2006; 209 (2): 157–65.
8. Sun SR, et al. The T-box transcription factor Brachyury promotes renal interstitial fibrosis by repressing E-cadherin expression. J Cell Commun Signal. 2014; 12:76.
9. U.S. Department of Health and Human Services: Centers for Disease Control and Prevention. Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories. Supplement / Vol. 61, January 6, 2012. https://www.cdc.gov/mmwr/pdf/other/su6101.pdf

Symbol Key / Légende des symboles/Erläuterung der Symbole

EC REP QAdvis EAR AB Ideon Science Park Scheelevägen 17 SE-223 70 Lund, Sweden	Storage Temperature Limites de température Zulässiger Temperaturbereich	Manufacturer Fabricant Hersteller	REF Catalog Number Référence du catalogue Bestellnummer
IVD In Vitro Diagnostic Medical Device Dispositif médical de diagnostic in vitro In-Vitro-Diagnostikum	Read Instructions for Use Consulter les instructions d'utilisation Gebrauchsanweisung beachten	Expiration Date Utiliser jusque Verwendbar bis	LOT Lot Number Code du lot Chargenbezeichnung

Bio SB BIOSCIENCE FOR THE WORLD
5385 Hollister Avenue, Bldg. 8, Ste. 108 Santa Barbara, CA 93111, USA
Tel. (805) 692-2768 | Tel. (800) 561-1145 | Fax. (805) 692-2769
E-mail: sales@biosb.com | Website: www.biosb.com