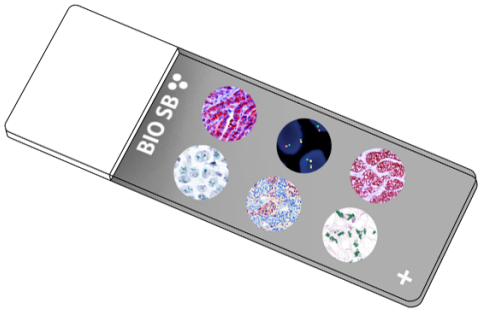
FOXO1
Control Slides

CE IVD



Intended Use
For In Vitro Diagnostic Use.

Summary and Explanation
The Forkhead transcription factor FOXO1, an important downstream target of phosphatidylinositol-3-kinase (PI3K)/AKT signaling pathway, regulates cellular homeostasis by maintaining cell proliferation, apoptosis and viability in normal cells. FOXO1 is a transcription factor that plays important roles in regulation of gluconeogenesis and glycogenolysis by insulin signaling, and is also central to the decision for a preadipocyte to commit to adipogenesis.

FOXO1 is broadly expressed in different types of cells with high levels of expression in lymphoid cells and non-Hodgkin's lymphomas. In contrast, in most classical Hodgkin lymphoma, Reed-Sternberg cells are FOXO1 negative. A study investigated the role of PI3K inhibition on FOXO1 regulation in Cervical Cancer. Expression profiling of primary tumors and cell lines show over expression of PIK3CA and AKT1; and down regulation of FOXO1. It was concluded that activation of FOXO1 and its nuclear sequestration is critical in the regulation of cell proliferation, cell viability and apoptosis in Cervical Cancer. Hence, PI3K/AKT pathway may be a potential molecular target for cervical cancer therapy. Androgens and the androgen receptor are essential for growth and differentiation of the normal prostate gland as well as proliferation and survival of Prostate Cancer. FOXA1, functions as a pioneer factor to facilitate AR transactivation and Prostate Cancer growth. In contrast, the O-class of FOX proteins such as FOXO1 and FOXO3, which are downstream effectors of the PTEN tumor suppressor, inhibits the transcriptional activity of either full-length androgen receptors or constitutively active splice variants of androgen receptors in a direct or indirect manner in Prostate Cancer. Translocation of FOXO1 gene with PAX3 has been associated with alveolar rhabdomyosarcoma.

Presentation
Five slides of FOXO1 positive tissues, each mounted on Hydrophilic Plus Slides, provided in a plastic mailer.

Catalog No.	*Quantity*
BSB-9184-CS	5 slides
BSB 2921	5 slides

Storage Store at 20-25°C

Precautions
1. For professional users only. Results should be interpreted by a qualified medical professional.
2. 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. Follow safety precautions of the heating device used for epitope retrieval (TintoRetriever Pressure Cooker or similar).
8. For additional safety information, refer to 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 after expiration date listed on package label.

IHC Protocol
1. 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).
2. 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.
3. After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15-20 minutes.
4. For manual staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to instrument manufacturer's instructions.
5. Wash slides with ImmunoDNA washer or DI water.
6. Continue IHC staining 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

Abbreviated IF Protocol

Step	Incubation Time
Rinse slides in IF wash buffer	5 minutes
Drain and wipe excess IF wash buffer off slide	
Conduct remaining steps in the dark	
Apply Antibody	30-60 minutes
Rinse with 3 changes of IF wash buffer	3x15 minutes each
Coverslip with IF mounting medium	

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. Galili N, et al. Fusion of a fork head domain gene to PAX3 in the solid tumour alveolar rhabdomyosarcoma. Nat. Genet. 1993; 5 (3): 230–5.
2. Nakae J, et al. The forkhead transcription factor Foxo1 regulates adipocyte differentiation. Dev. Cell. 2003; 4 (1): 119–29.
3. Rena G, et al. Phosphorylation of the transcription factor forkhead family member FKHR by protein kinase B. J. Biol. Chem. 1999; 274(24): 17179–83.
4. Prasad SB, et al. Down Regulation of FOXO1 Promotes Cell Proliferation in Cervical Cancer. J Cancer. 2014 Aug; 5(8):655-62.
5. Zhao Y, et al. Modulation of Androgen Receptor by FOXA1 and FOXO1 Factors in Prostate Cancer. Int J Biol Sci 2014; 10(6):614-619.
6. Linardic CM. PAX3-FOXO1 fusion gene in rhabdomyosarcoma. Cancer Lett. 2008; 270 (1): 10–8.
7. 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

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