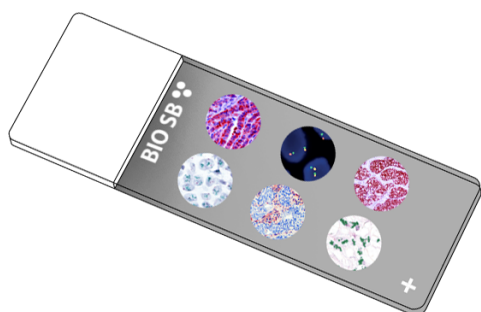


Factor H/Complement Factor H Control Slides



Intended Use

For In Vitro Diagnostic Use.

Summary and Explanation

Factor H or Complement Factor H is the major soluble inhibitor of complement, where its binding to self markers (i.e. particular glycan structures) prevents complement activation and amplification on host surfaces. Mutations and polymorphisms that affect recognition of self markers by Factor H are associated with diseases of complement dysregulation, such as age-related macular degeneration and atypical hemolytic uremic syndrome. In addition, pathogens and cancer cells can hijack Factor H to evade the immune response. Lung, Ovarian, Glial and Colon Cancer cells show enhanced expression and surface binding of soluble regulators, including Factor H. Factor H has been shown to be expressed by human Breast Cancer cells, which correlates with the presence of immunosuppressive macrophages, Breast Cancer recurrence and severity of the disease. Lung cancer cells may develop a protective mechanism against complement attack by expressing and binding Factor H to their cell membranes. Additionally, it has been demonstrated that Factor H is upregulated by constitutive activation of STAT4, which is accounted for by SOCS silencing in Lung Cancer cells. Several studies have also suggested the importance of Factor H in the protection of tumor cells against complement activation. The importance of Factor H expression for the protection of cancer cells in vivo will help to elucidate the mechanisms used by tumor cells to avoid complement activity and assist in the design of more efficient complement-mediated immunotherapies. It has been demonstrated that SARS-CoV-2 spike proteins activate complement by engaging the alternative pathway of complement, which may explain many of the clinical manifestations (microangiopathy, thrombocytopenia, renal injury, and thrombophilia) and that blocking this process by inhibiting factor D or C5 could mitigate SARS-CoV-2-induced immunopathology. Addition of Factor H mitigates this complement attack. It has been suggested that a subset of patients with COVID-19 may have a genetic predisposition associated with complement dysregulation.

Presentation

Five slides of Factor H/Complement Factor H positive tissues, each mounted on Hydrophilic Plus Slides, provided in a plastic mailer.

Catalog No.	Quantity
BSB-9174-CS	5 slides
BSB-3723-CS	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 PermaMunter (BSB 0169-0174) or organic solvent based resin such as PermaMunter (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. Parente R, Clark SJ, Inforzato A, Day AJ. Complement factor H in host defense and immune evasion. *Cell Mol Life Sci.* 2017;74(9):1605-1624. doi:10.1007/s00018-016-2418-42. Kolev M, Towner L, Donev R. Complement in Cancer and Cancer Immunotherapy. *Arch. Immunol. Ther. Exp.* 59, 407-419 (2011). <https://doi.org/10.1007/s00005-011-0146-x3>. Junnikkala S, Jokiranta TS, Friese MA, Jarva H, Zipfel PF, Meri S. Exceptional resistance of human H2 glioblastoma cells to complement-mediated killing by expression and utilization of factor H and factor H-like protein 1. *J Immunol.* 2000;164(11):6075-6081. doi:10.4049/jimmunol.164.11.60754. Smolag KI, Mueni CM, Leandersson K, et al. Complement inhibitor factor H expressed by breast cancer cells differentiates CD14+ human monocytes into immunosuppressive macrophages. *Oncoimmunology.* 2020;9(1):1731135. Published 2020 Mar 6. doi:10.1080/2162402X.2020.17311355. Yoon YH, Hwang HJ, Sung HJ, et al. Upregulation of Complement Factor H by SOCS-1/3-STAT4 in Lung Cancer. *Cancers (Basel).* 2019;11(4):471. Published 2019 Apr 3. doi:10.3390/cancers110404716. Yu J, Yuan X, Chen H, Chaturvedi S, Braunstein EM, Brodsky RA. Direct activation of the alternative complement pathway by SARS-CoV-2 spike proteins is blocked by factor D inhibition. *Blood.* 2020;136(18):2080-2089. doi:10.1182/blood.2020082487. U.S. Department of Health and Human Services: Centers for Disease Control and Prevention. Guidelines for Safe WorkPractices 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