P.O. Box 3286 - Logan, Utah 84323, U.S.A. - Tel. (800) 729-8350 - Fax (435) 755-0015 - www.scytek.com

Human to Human Blocking Reagent

1. Description: ScyTek's Human to Human reagent has been formulated to provide the researcher with a staining system capable of visualizing human monoclonal antibodies on human tissue. In most cases a 30-minute incubation with Human to Human block will virtually eliminate background staining that is caused by endogenous immunoglobulins.

2. Uses/Limitations: Not to be taken internally.
For In-Vitro Diagnostic use only.
Histological applications.
Do not use if reagent becomes cloudy.
Do not use past expiration date.
Use caution when handling reagent.
Non-Sterile.

3. Availability:

Item #	Volume
HTH008	8ml
HTH015	15ml
HTH100	100ml

4. Storage:
Store at 2-8°C. Solution is stable for 18 months after date of manufacture.

5. Procedure:

1. Deparaffinize and rehydrate tissue section.
2. To reduce nonspecific background staining due to endogenous peroxidase, incubate slide in a Hydrogen Peroxide Block (ScyTek Item#: ACA) for 10 minutes.
3. Wash 2 times in buffer.
4. If required, incubate tissue in digestive enzyme.
5. Wash 4 times in buffer.
6. Apply a Protein Block (Item#: AAA), and incubate for 5 minutes at room temperature to block nonspecific background staining. **Note:** Do not exceed 10 minutes or there may be a reduction in desired stain.
7. Wash 1 time in buffer.
8. Apply Human-To-Human Blocking Reagent and incubate 10-60 minutes. Incubation time is dependent on the amount of endogenous Ig found in the tissue type.
9. Rinse 4 times in buffer.
10. Apply primary antibody and incubate according to manufacturer's protocol.
12. Wash 4 times in buffer.
13. Apply link antibody and incubate according to manufacturer's protocol.
14. Wash 4 times in buffer.
15. Apply enzyme label and incubate according to manufacturer's protocol.

Storage: 2° C  8° C


ScyTek Laboratories, Inc.
205 South 600 West
Logan, UT 84321
435-755-9848
U.S.A.

CE IVD

EC REP
Emergo Europe
Prinsessegracht 20
2514 AP The Hague, The Netherlands

P.O. Box 3286 - Logan, Utah 84323, U.S.A. - Tel. (800) 729-8350 - Fax (435) 755-0015 - www.scytek.com

16. Rinse 4 times in buffer.
17. Apply chromogen/substrate and incubate according to manufacturer's protocol.
18. Counterstain and coverslip.

Troubleshooting Guide

Overstaining:

1. Concentration of the primary antibody was too high or the incubation time was too long.
2. Temperature during incubation was too high.
3. Incubation time with detection reagents was too long.

Nonspecific Background Staining:

1. Rinsing between steps was inadequate.
2. Tissue was allowed to dry with reagents on.
3. Folds in tissue trapped reagents.
4. Inadequate blocking with Human-To-Human Blocking Reagent.
5. Tissue contains endogenous biotin.
6. Antigen migrated in tissue.
7. Excessive tissue adhesive on slides.
8. Inadequate blocking with Super block.

Weak Staining:

1. Primary antibody concentration was too low or incubation time was too short.
2. Reagents are past their expiration date.
3. Inadequate removal of wash water between steps, resulting in dilution of reagents.
4. Counterstain or mounting media were incompatible and dissolved the chromogen reaction product.
5. Room temperature was excessively cool.
6. The primary antibody does not recognize an antigen that survives fixation and embedding.
7. Excessive incubation with Super Block.

No Staining:

1. Steps were inadvertently left out.
2. There is no antigen in the tissue.
3. The primary antibody is not of mouse, rat, rabbit or guinea pig origin.

Product Specific Literature References:

1. Lee, Eun Sook, Keon Wook Kang, Byong Chul Yoo, Ho-Young Lee, Sun Young Kong, Se Hun Kang, Nam Suk Baek, Bu-Mi Kwon, and Young Mi Kwon. Kit for diagnosis of breast cancer using herceptin, a composition comprising herceptin and a method for detecting herceptin-sensitive her2 overexpressed cell using the same. United States US20100316635A1, filed October 16, 2008, and issued December 16, 2010. https://patents.google.com/patent/US20100316635A1/en.

Storage: 2° C 8° C

ScyTek Laboratories, Inc.
205 South 600 West
Logan, UT 84321
435-755-9848
U.S.A.

CE IVD

EC REP

Emergo Europe
Prinsessegracht 20
2514 AP The Hague, The Netherlands