

Instructions For Use ABH-IFU

Rev. Date: August 1, 2019

Revision: 3

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

SensiTek Alkaline Phosphatase

Uses/Limitations: A Ready-to-use streptavidin-Alkaline Phosphatase enzyme label designed for 3-step

immunohistochemistry protocols. Formulated to provide optimal staining with an incubation for 15-20 minutes. May be used with automated systems, reagent jars, and

manual dropping/pipetting.

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.

Control Tissue: Any well-fixed tissue.

Availability/Contents:

<u>Volume</u>
8 ml
15 ml
125 ml
500 ml
1000 ml



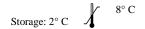
Human prostate stained with Permanent Red and SensiTek Alkaline Phosphatase within an IHC protocol.

Storage: Store at 2-8°Centigrade. Product is stable for 18 months from date of manufacture.

Procedure:

Allow reagents to come to room temperature before use.

- 1. Deparaffinize and rehydrate tissue section.
- 2. If required, incubate tissue in digestive enzyme.
- 3. Wash 4 times in buffer.
- 4. Place slide in protein block (ScyTek's "Superblock") and incubate 5-10 minutes at room temperature to block nonspecific background staining. Note: Do not exceed 10 minutes or there may be a reduction in desired stain.
- 5. Wash 1 time in buffer.
- Apply primary antibody and incubate according to manufacturer's protocol.
- 7. Wash 4 times in buffer.









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- 8. Place slide in biotinylated link secondary antibody, and incubate per instructions.
- 9. Wash 4 times in buffer.
- 10. Place slide in Sensitek Alkaline Phosphatase, and incubate for 15-20 minutes.
- 11. Rinse 4 times in buffer.
- 12. Place slide in appropriate chromogenic substrate and incubate until desired reaction is achieved.
- 13. 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 link antibody or streptavidin/enzyme label 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. Tissue contains endogenous enzyme.
- 5. Tissue contains endogenous biotin.
- 6. Antigen migrated in tissue.
- 7. Excessive tissue adhesive on slides.
- 8. Inadequate blocking with protein block.

Weak Staining:

- 1. Primary antibody concentration was too low or incubation time was too short.
- 2. Reagents are past their expiration date.
- 3. Reagent is reaching the end of its useful life.
- 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 in high enough amounts.
- 7. Excessive incubation with protein block (Super Block or normal serum).

No Staining:

- 1. Steps were inadvertently left out.
- 2. There is no relevant antigen in the tissue.
- 3. Chromogenic substrate does not match enzyme label.
- 4. One or more components have been inactivated.

