

SensiTek Horseradish Peroxidase

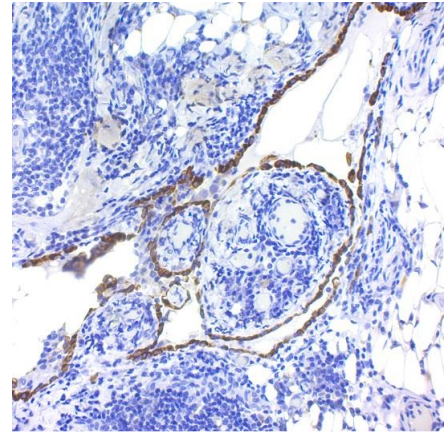
Uses/Limitations: A Ready-to-use streptavidin-horseradish peroxidase enzyme label designed for 3-step immunohistochemistry protocols. Formulated to provide optimal staining with an incubation for 15-20 minutes. May be used with automated systems, reagent jars, and manual dropping/pipetting.

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>Item #</u>	<u>Volume</u>
ABG008	8 ml
ABG015	15 ml
ABG125	125 ml
ABG500	500 ml
ABG999	1000 ml




Human ovary stained with DAB and SensiTek Horseradish Peroxidase within an IHC protocol.


Storage: Store at 2-8°C 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 needed, incubate slide in hydrogen peroxide for 10-15 minutes to reduce nonspecific background staining due to endogenous peroxidase.
3. Wash 2 times in buffer.
4. If required, incubate tissue in digestive enzyme.
5. Wash 4 times in buffer.
6. 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.

Storage: 2° C  8° C

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7. Wash 1 time in buffer.
8. Apply primary antibody and incubate according to manufacturer's protocol.
9. Wash 4 times in buffer.
10. Place slide in biotinylated link secondary antibody, and incubate per instructions.
11. Wash 4 times in buffer.
12. Place slide in Sensitek Horseradish Peroxidase, and incubate for 15-20 minutes.
13. Rinse 4 times in buffer.
14. Place slide in appropriate chromogenic substrate and incubate until desired reaction is achieved.
15. 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.

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EC REP


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References:


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