ScyTek Laboratories

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

UltraTek Alk-Phos (Anti-Mouse) Ready-To-Use (500 slide)

Species of Origin: Goat
Antigen Specificity: Anti-Mouse IgG (H+L)
Preabsorbed Against: Human, Bovine, Horse, Rabbit, Swine
Enzyme Conjugate: Alkaline Phosphatase
Chromogen Substrate: None provided

Procedure:

1. Deparaffinize and rehydrate tissue section.
2. If required, incubate tissue in digestive enzyme.
3. Wash 4 times in buffer.
4. Apply Super Block (blue cap), and incubate for 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.
6. Apply primary antibody and incubate according to manufacturer's protocol.
9. Wash 4 times in buffer.
10. Apply UltraTek Anti-Mouse (yellow cap), and incubate for 10 minutes at room temperature.
11. Wash 4 times in buffer.
12. Apply UltraTek Alk-Phos Label (red cap), and incubate for 10 minutes at room temperature.
13. Rinse 4 times in buffer.
14. Apply chromogen intended for use with Alkaline Phosphatase.
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.

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2. Tissue was allowed to dry with reagents on.
3. Folds in tissue trapped reagents.
4. Tissue contains endogenous biotin.
5. Antigen migrated in tissue.
6. Excessive tissue adhesive on slides.
7. 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. 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 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 antigen in the tissue.
3. The primary antibody is not of mouse origin.
4. Chromogenic substrate has been replaced with another that is not intended for use with alkaline phosphatase.
5. One or more components of the kit have been inactivated.