

Affinity Purified Antibody to FITC

Produced in Goat



Catalog No.
01-40-01

Size
1.0 mg

DESCRIPTION

Affinity purified goat-derived antibody to Fluorescein (FITC). This antibody has specifically been raised against FITC isomer 1. It is designed for the detection of FITC in immunohistochemistry (IHC) applications.

FORM/STORAGE

Lyophilized. Store at 2-8 °C. Stable for a minimum of 1 year from date of receipt when stored at 2-8 °C.

STABILIZER AND PRESERVATIVE

No stabilizers or preservatives added. Non-sterile.

ANTIBODY CONCENTRATION

The amount of affinity purified antibody is 1.0 mg as determined by UV absorbance at 280 nm.

REHYDRATION AND STORAGE

Note: Rehydration of antibodies in TBS or buffers other than those listed here is not recommended.

Procedure A: 50% Glycerol

At a working dilution, the concentration of glycerol is too small to affect most assays. The use of glycerol is not recommended when the antibody is used in live cell work.

Rehydration: Add 1 mL of 50% glycerol in water to the vial. Pipette up and down several times to ensure proper mixing.

Storage: This product may be stored either refrigerated or at 20 °C. Stable for a minimum of 1 year.

Procedure B: Reagent Quality Water

Rehydration: Add 1 mL of reagent quality water to the vial. Pipette up and down several times to ensure proper mixing.

Storage: This product may be stored aliquotted at -20 °C. Care should be taken to minimize multiple freeze/thaw cycles.

NOTES

1. This antibody is intended to be used for the detection of FITC-labeled antibodies in IHC assays.
2. Minimize procedures that may destroy epitopes.
 - a. Universal Block is not recommended for inactivation of endogenous enzyme activity, as it may destroy epitopes.
3. Secondary antibodies should be titrated to determine proper working dilutions.
4. Always incorporate a positive control, negative control and reagent control.
5. Do not allow sections to dry out during incubations.
6. Remove as much buffer as possible after washes.
7. Reagent quality water is recommended for use.

PROCEDURES

The protocol below is designed for use on frozen sections with colorimetric detection. Assay optimization will need to occur when using this antibody on other specimen types.

FIXATION

1. Fix the section (ex. – acetone, methanol), and block for endogenous enzyme activity, using a block that will not destroy epitopes (ex. – 1% H₂O₂).

APPLY SERUM BLOCK

1. Shake off buffer and wipe off excess buffer surrounding section.
2. Completely cover section with Normal Rabbit Serum.
3. Incubate 15 minutes at room temperature in a humidified chamber.
4. Soak in TBS for 5 minutes.

APPLY PRIMARY ANTIBODY

1. Shake off TBS and wipe off any excess surrounding the section.
2. Dilute antibody to requisite concentration in TBS containing 1% Normal Rabbit Serum.
3. Completely cover section with diluted primary antibody.
4. Incubate 10 - 30 minutes at room temperature in a humidified chamber.
5. Rinse off primary antibody with TBS. Soak 5 minutes in same buffer.

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APPLY ANTI-FITC ANTIBODY

1. Shake off TBS and wipe off any excess surrounding the section.
2. Completely cover section with anti-FITC antibody (diluted 1:100 – 1:1,000) in TBS containing 1% Normal Rabbit Serum.
3. Incubate 10 - 30 minutes at room temperature in a humidified chamber.
4. Rinse off antibody with TBS.
5. Soak 5 minutes in same buffer.

APPLY BIOTINYLATED ANTIBODY

1. Shake off TBS and wipe off any excess surrounding the section.
2. Completely cover section with biotinylated Rabbit anti-Goat IgG antibody diluted in TBS containing 1% Normal Rabbit Serum.
3. Incubate 10 - 30 minutes at room temperature in a humidified chamber.
4. Rinse off antibody with TBS.
5. Soak 5 minutes in same buffer.

APPLY ENZYME-LABELED STREPTAVIDIN

1. Shake off TBS and wipe off any excess surrounding the section.
2. Completely cover section with enzyme-labeled streptavidin diluted in 1% BSA.
3. Incubate 10 - 30 minutes at room temperature in a humidified chamber.
4. Rinse off streptavidin with TBS. Rinse 5 minutes in same buffer.
5. Repeat Step 4.

COLOR DEVELOPMENT

Develop color using one of KPL's HistoMark® substrates (See RELATED PRODUCTS).

NOTE: If color develops too rapidly for your staining conditions, (i.e. less than one minute), further dilution of the primary antibody is recommended. An estimation of appropriate primary antibody dilution may be obtained by applying 1/50, 1/100, 1/200, 1/400 and 1/800 dilutions to tissue sections. The optimal dilution is the one that results in appropriate color development within 10 minutes without background staining.

RELATED PRODUCTS

HistoMark TrueBlue	Cat. No. 50-78-02
HistoMark Black	Cat. No. 54-75-00
HistoMark Red	Cat. No. 55-69-00
HistoMark Blue	Cat. No. 55-70-00
Normal Rabbit Serum	Cat. No. 71-00-28
10% BSA	Cat. No. 50-61-00
Biotin, Rabbit anti-Goat IgG	Cat. No. 71-00-37
10X TBS	Cat. No. 51-17-01

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