IsoStrip™ Mouse Monoclonal Antibody Isotyping Kit: *sc-24958*

BACKGROUND

The IsoStrip™ Mouse Monoclonal Antibody Isotyping Kit utilizes two main components for isotype characterization. Each development tube supplied with the kit contains latex beads bearing anti-mouse kappa and anti-mouse lambda antibodies, which will react with any mouse monoclonal antibody regardless of its isotype. The basis of the kit is the isotyping strip itself, which bears immobilized bands of goat anti-mouse antibodies corresponding to each of the common mouse antibody isotypes (IgG_1, IgG_{2a}, IgG_{2b}, IgG_3, IgM and IgA) and to the kappa and lambda light chains. Both sides of the strip also bear a positive control band, which indicates that the antibody-coated beads have traveled up the strip.

PROTOCOL AND USAGE

Remove the desired number of isotyping strips from the canister. Remove the caps from an equal number of development tubes. The tubes may be labeled with a pencil or lab marker for identification.

Dilute a sample containing the mouse monoclonal antibody in PBS (phosphate buffered saline), pH 7.2-7.6.

Culture supernatant samples should be diluted 1:10 to 1:100. Ascites samples should be diluted 1:20,000. These dilutions may vary depending on the concentration of antibody in your sample. We recommend using a monoclonal antibody concentration of 0.1-1 µg/ml of diluted sample for optimum results. 150 µl of this diluted sample will be added to the development tube.

Pipette 150 µl of the freshly diluted sample into each development tube. Incubate at room temperature for 30 seconds and briefly agitate (vortex) the tube so that the colored latex is completely resuspended.

Place one isotyping strip, with the black end at the bottom, in each development tube.

Interpret results at 5-10 minutes (once the positive control bands have appeared). Do not wash the strip to stop the reaction.

Within 5-10 minutes, a blue band will appear in either the kappa or lambda section of the strip, as well as in one of the class or subclass sections, indicating the light chain composition and class or subclass, respectively, of the monoclonal antibody. These blue bands will intensify as the sample moves up the strip. The positive control bands on each side of the isotyping strip should also appear, indicating that the antibody-coated latex beads are functional and have traveled up the strip. In some cases, such as testing very dilute samples, the test may take up to 10 minutes. If your sample contains a different number of bands than indicated above, the less intense bands may be due to heavy and light chain(s) from a small amount of contaminating antibody (if using ascites or an antibody-secreting myeloma line during hybridoma production) or a sample that was too concentrated. NOTE: For a permanent experimental record, or for an easier interpretation of results when testing samples containing host immunoglobulins, the black area may be cut off the bottom of the strip to prevent further band development once the positive control bands are visible.

PRODUCT

The IsoStrip™ Mouse Monoclonal Antibody Isotyping Kit is a three-step kit for the rapid characterization of mouse monoclonal antibodies. Each kit is suitable for 10 tests and contains 10 capped development tubes, each containing lyophilized latex beads, and 10 isotyping strips in a canister. IsoStrip is a trademark of Roche Applied Science.

STORAGE

Store at 4° C, do not freeze; stable for one year from the date of shipment.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PRODUCT CITATIONS

Product Citations not yet available. If you have published a paper using sc-24958, please contact Technical Service. 427: 349-352.

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