

AEC Substrate Chromogen

For In Vitro Diagnostic Use (IVD)
Instructions for use

INTENDED USE

The reagent is designated to detect target antigens in formalin-fixed, paraffin-embedded tissue sections when used in conjunction with antibodies and appropriate chromogen in the IHC staining process.

SUMMARY AND EXPLANATION

When in the presence of HRP, AEC produces a rust red precipitate that can be readily visualized using light microscopy.

PRINCIPLES AND PROCEDURES

The chromogen is the final step in the detection portion of the IHC process; it enables the antibody-antigen complex to be viewed under the light microscope. AEC acts as an electron donor in the presence of enzyme HRP. As a result, AEC gets reduced and the color change occurs.

MATERIALS AND METHODS

Reagents Supplied As:

| Reagent Cat. No. | Contents | Vol. (ml) |
|------------------|-------------------------|-----------|
| 958D-20 | AEC Substrate Chromogen | 50.0 mL |
| 958D-30 | AEC Substrate Chromogen | 200.0 mL |

Materials and Reagents Needed But Not Provided

- | | |
|--|--------------------------------|
| 1. TBS or PBS wash buffer* | 8. Slide rack* |
| 2. Volumetric flask/graduated cylinder | 9. Staining dishes* |
| 3. Microscope slides, positively charged | 10. Pressure cooker* |
| 4. Drying oven | 11. Pretreatment reagents* |
| 5. Positive and negative controls | 12. Proteolytic enzyme |
| 6. Clearing agent (xylene, Clearene, etc.) | 13. Avidin-Biotin block* |
| 7. Ethanol or reagent alcohol | 14. Peroxide block |
| | 15. Negative control reagents* |
| | 16. Hematoxylin* |
| | 17. Mounting medium |

*See Cell Marque Catalog for product numbers. Some of the reagents listed are based on specific applications and detection system used.

Storage and Stability

Store at 2-8°C up to a maximum of 24 month(s) from the date of manufacture (see product label for expiration date). Keep protected from light.

Recommended Protocol Instructions

1. Cover tissue specimen completely with AEC ready-to-use.
2. Incubate at room temperature for 30 seconds to 30 minutes, as needed for full color development.
3. Rinse slides in distilled water and counterstain.
4. Dehydrate and coverslip with an aqueous based mounting medium.

Protocol Notes

1. Product is photosensitive and temperature sensitive. Replace cap on vial and return product to refrigerator when not in use.
2. It is recommended to use a non-alcoholic counterstain in combination with an aqueous mounting media. Optimal results are achieved when tissues are mounted in an aqueous mounting media.

INTERPRETATION OF RESULTS

The Cell Marque AEC Substrate Chromogen causes a rust red reaction product to precipitate at the antigen sites localized by the primary antibody. A qualified pathologist must evaluate controls and qualify the stained product before interpreting results.

QUALITY CONTROL PROCEDURES

Refer to NCCLS Quality Assurance for Immunocytochemistry approved guidelines, December 1999 MM4-A Vol.19 No.26 for more information on tissue controls.

WARNINGS AND PRECAUTIONS

1. This product is for *in vitro* diagnostic use by professionals only.
2. Do not use after expiration date printed on product labels. The user must validate any storage conditions other than those specified in the package insert.
3. Bring all reagents, slides, and specimens to room temperature (18-24°C) prior to use.
4. Cross contamination of reagents or samples may give false results.
5. Avoid microbial contamination of reagents, as this could produce incorrect results.
6. Avoid contact of reagents with eyes and mucous membranes. If reagents come in contact with sensitive areas, wash with copious amounts of water.
7. Do not smoke, eat, or drink in areas where specimens or reagents are handled.
8. Avoid splashing or generation of aerosols at all times.
9. Reusable glassware must be washed and thoroughly rinsed free of detergent prior to use. All glassware must be clean and dry before use.

10. Never pipette by mouth and avoid contact of reagents and specimens with skin and mucous membranes. If contact occurs, wash with germicidal soap and copious amounts of water.
11. Refer to product SDS.
12. AEC end product is soluble in organic compounds and may result in stain fading over time if exposed to organic compounds.
13. Do not put slides through alcohols or xylenes during coverslipping.
14. Do not store or use in strong light as product is photosensitive.
15. Return product to 2-8°C when not in use as product is temperature sensitive.

LIMITATIONS

Immunohistochemistry is a multiple step diagnostic process that requires specialized training and selection of appropriate reagents and controls. The protocols for a specific application can vary. It is the responsibility of the end user to determine optimal conditions.

TROUBLESHOOTING

Refer to reagent-specific protocol recommendation according to data sheet provided.

For further help, feel free to contact Cell Marque's Technical Support at +1-800-665-7284.

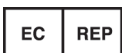
REFERENCES

1. NCCLS Quality Assurance for Immunocytochemistry approved guideline, December 1999 MM4-A Vol. 19 No.26 for more information on tissue controls.

DISCLAIMERS



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