## **EDTA Decloaker**, 5X

Pretreatment Reagent 901-CB917-073117

## Catalog Number: CB917 L, M

Description: 100, 500 ml, concentrate

### Intended Use:

For In Vitro Diagnostic Use

EDTA Decloaker, 5X is a buffered solution that is intended for laboratory use in heat-induced antigen retrieval of formalin-fixed paraffin-embedded (FFPE) tissues for immunohistochemistry (IHC) procedures. The clinical interpretation of any staining or its absence should be complemented by morphological studies using proper controls and should be evaluated within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist.

## Summary & Explanation:

EDTA Decloaker is a heat retrieval solution, pH 8.4-8.7. It is recommended for use with antibodies with low antigen expression (cyclin D1, CD1a, CD3, CD4, CD8, CD23, bcl-6, CD61, CD79a and TdT). EDTA Decloaker is a potent agent for epitope retrieval and tissues may be 5-10 times more sensitive to immunohistochemical staining. EDTA Decloaker incorporates Assure™ technology, a color-coded high temperature pH indicator solution. The end-user is assured by visual inspection that the solution is at the correct dilution and pH. This product is specially formulated for superior pH stability at high temperatures and will help prevent the possibility of losing pH sensitive antigens. EDTA Decloaker is non-toxic, non-flammable, odorless and sodium azide and thimerosal free.

## **Known Applications:**

Immunohistochemistry (formalin-fixed paraffin-embedded tissues) **Supplied As:** 

100ml

EDTA Decloaker, 5X concentrate (CB917L) 500ml

EDTA Decloaker, 5X concentrate (CB917M)

## Materials and Reagents (Needed But Not Provided):

Microscope slides, positively charged Desert Chamber\* (Drying oven) Positive and negative tissue controls Xylene (Could be substituted with xylene substitute\*) Ethanol or reagent alcohol Decloaking Chamber\* (Pressure cooker) Deionized or distilled water Wash buffer\*(TBS/PBS) Avidin-Biotin Blocking Kit\*(Labeled Streptavidin Kits Only) Peroxidase block\* Protein block\* Primary antibody\* Negative control reagents\* Detection kits\* Detection components\* Chromogens\* Hematoxylin\* Bluing reagent\* Mounting medium\*

\* Biocare Medical Products: Refer to a Biocare Medical catalog for further information regarding catalog numbers and ordering information. Certain reagents listed above are based on specific application and detection system used.



### Storage and Stability:

Store at room temperature. Do not use after expiration date printed on vial. If reagents are stored under conditions other than those specified in the package insert, they must be verified by the user. Diluted reagents should be used promptly; any remaining reagent should be stored at room temperature.

#### **Protocol Recommendations:**

1. Deparaffinize tissues and hydrate to water. If necessary, block for endogenous peroxidase and wash in DI water.

2. Dilute concentrate EDTA at a ratio of 1:5 (1 ml EDTA to 4 ml deionized water)

3. Place slides into 1X retrieval solution in a slide container (e.g. Coplin jar, Tissue-Tek<sup>™</sup>, staining dish or metal slide canister).

4. Retrieve sections under pressure using Biocare's Decloaking Chamber. Follow the recommendations in the antibody data sheet and Decloaking Chamber User Manual.

5. Check solution for appropriate color change. (See Technical Note #1)

6. Gently rinse by gradually adding DI water to the solution, then remove slides and rinse with DI water.

## **Technical Notes:**

1. Concentrated EDTA Decloaker is a blue color. RTU or 1X solution is a clear blue color. When heating, if the solution turns greenish blue, green or yellow, the pH of the solution is not optimum and tissue staining may be significantly reduced. The pH of the 1X solution should range from 8.0 to 8.7 for optimum staining. The grade of deionized water is very important. If high-grade water is difficult to obtain, sterile water may be used.

2. If using Biocare's Desert Chamber Pro (a programmable turboaction drying oven), dry sections at  $25^{\circ}$ C overnight or at  $37^{\circ}$ C for 30-60 minutes and then dry slides at  $60^{\circ}$ C for 30 minutes.

3. Use positive charged slides (use Biocare's Kling-On HIER Slides) and cut tissues at 4-5 microns. Do not use any adhesives in the water bath. Poor fixation and processing of tissues will cause tissue sections to fall off the slides, especially fatty tissues such as breast. Tissues should be fixed a minimum of 6-12 hours.

4. Protocol time and temperatures for HIER can vary depending on the Decloaking Chamber model used. Please refer to the relevant Decloaking Chamber manual for appropriate protocol times and temperatures.

## Limitations:

The protocols for a specific application can vary. These include, but are not limited to: fixation, heat-retrieval method, incubation times, tissue section thickness and detection kit used. Due to the superior sensitivity of these unique reagents, the recommended incubation times and titers listed are not applicable to other detection systems, as results may vary. The data sheet recommendations and protocols are based on exclusive use of Biocare products. Ultimately, it is the responsibility of the investigator to determine optimal conditions. The clinical interpretation of any positive or negative staining should be evaluated within the context of clinical presentation, morphology and other histopathological criteria by a qualified pathologist. The clinical interpretation of any positive or negative staining should be complemented by morphological studies using proper positive and negative internal and external controls as well as other diagnostic tests.





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## **Quality Control:**

Refer to CLSI Quality Standards for Design and Implementation of Immunohistochemistry Assays; Approved Guideline-Second edition (I/LA28-A2). CLSI Wayne, PA, USA (www.clsi.org). 2011

## Precautions:

1. This product is not classified as hazardous. The preservative used in this reagent is Proclin 300 and the concentration is less than 0.25%. Overexposure to Proclin 300 can cause skin and eye irritation and irritation to mucous membranes and upper respiratory tract. The concentration of Proclin 300 in this product does not meet the OSHA criteria for a hazardous substance. Wear disposable gloves when handling reagents.

2. Specimens, before and after fixation, and all materials exposed to them should be handled as if capable of transmitting infection and disposed of with proper precautions.

Never pipette reagents by mouth and avoid contacting the skin and mucous membranes with reagents and specimens. If reagents or specimens come in contact with sensitive areas, wash with copious amounts of water.

3. Microbial contamination of reagents may result in an increase in nonspecific staining.

4. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change.

5. Do not use reagent after the expiration date printed on the vial.

6. The SDS is available upon request and is located at http://biocare.net/.

7. Consult OSHA, federal, state or local regulations for disposal of any toxic substances. Proclin is a trademark of Rohm and Haas Company, or of its subsidiaries or affiliates.

#### Troubleshooting:

Follow the antibody specific protocol recommendations according to data sheet provided. If atypical results occur, contact Biocare's Technical Support at 1-800-542-2002.





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