

# Hot Rinse, 25X

## Ancillary Reagent

Control Number: 901-HTR1001-090617

**Catalog Number:** HTR1001 M

**Description:** 500 ml; concentrate

**Intended Use:**

For In Vitro Diagnostic Use

Hot Rinse, 25X is intended for laboratory use in immunohistochemistry procedures on formalin-fixed paraffin-embedded (FFPE) tissues to ensure complete deparaffinization. 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:**

Hot Rinse is sodium azide and thimerosal free, non-toxic, non-flammable and odorless. After deparaffinizing tissues in Reveal, Universal or Borg Decloaker, a subsequent hot rinse ensures complete deparaffinization. It is specially designed for use with Biocare's Decloaking Chamber and is also compatible with other commercial heat retrieval deparaffinization solutions.

**Known Applications:**

Immunohistochemistry (formalin-fixed paraffin-embedded tissues)

**Supplied As:**

**500ml**

Hot Rinse, 25X concentrate (HTR1001M)

**Materials and Reagents Needed But Not Provided:**

- Microscope slides, positively charged
- Desert Chamber\* (Drying oven)
- Positive and negative tissue controls
- Xylene (Could be replaced with a xylene substitute\*)
- Ethanol or reagent alcohol
- Decloaking Chamber\* (Pressure cooker)
- Deionized or distilled water
- Wash buffer\*(TBS/PBS)
- Pretreatment Reagents\*
- Enzyme digestion\*
- 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 media\*

\* 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. Dry tissue sections for 1 hour at 37°C and then dry slides for 10-30 minutes at 60°C.
2. Dilute concentrated Hot Rinse at a ratio of 1:25 (1 ml Hot Rinse to 24 ml of deionized water).
3. Fill a Coplin Jar, Tissue-Tek™ staining dish or metal slide canister with 1X Hot Rinse.
4. Fill a second container with 1X of Biocare's Reveal, Borg or Universal Decloaker HIER solution.
5. Place both solutions into the Decloaking Chamber.
6. Place the slides into the slide container with the 1X HIER solution.
7. Retrieve sections under pressure using Biocare's Decloaking Chamber. Follow the recommendations on the antibody data sheet and Table 1 (below).

**Protocol Recommendations Cont'd:**

8. Check solution for appropriate color change (refer to appropriate HIER solution data sheet).
9. Transfer slides to the slide container with 1X Hot Rinse and agitate 20 dips.
10. Gently rinse by gradually adding DI water to the solution, then remove slides and rinse with DI water.

**Technical Notes:**

1. Hot Rinse may be used with various heat retrieval methods including a microwave oven, pressure cooker, water bath or steamer.
2. If using Biocare's Desert Chamber Pro (a programmable turbo-action drying oven), dry sections at 25°C overnight or at 37°C for 30-60 minutes then dry slides at 60°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.

Table 1. Biocare Decloaking Chamber Protocol Equivalence

| Protocol     | DC2002/DC2008 | DC NxGen           |
|--------------|---------------|--------------------|
| 125°C/30 sec | 125°C/30 sec  | 110°C/10 to 15 min |
| 95°C/40 min  | 95°C/40 min   | 95°C/40 min        |
| 90°C/15 min  | 90°C/15 min   | 90°C/15 min        |
| 80°C/60 min  | 80°C/60 min   | 80°C/60 min        |
| 60°C/15 hrs  | 60°C/15 hrs   | 60°C/15 hrs        |

**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.

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