ONCORE ERO*

BIOCARE M E D I C A L

Mouse-on-Rat HRP-Polymer

Micro-Polymer Detection 902-OPRR621-111023

Available Product Formats		
Catalog Number	Volume	
OPRR621T60	60 tests	

Intended Use:

For Research Use Only. Not for use in diagnostic procedures.

Background Information:

The Mouse-on-Rat HRP-Polymer is designed using a one-step method for detecting mouse primary antibodies to form an antibody-enzyme complex on rat tissues. This complex is then visualized using an appropriate substrate/chromogen. In the one-step method a secondary antibody directly linked to the micro polymer is applied. Mouse-on-Rat HRP-Polymer is provided ready-to-use and is intended to be applied as defined by the staining protocols on the ONCORE Pro X Automated Slide Stainer.

Known Applications:

Immunohistochemistry (Formalin-fixed paraffin-embedded tissues). Other applications have not been tested.

Materials and Methods:

Reagents Provided:

Kit Catalog No.	Component Description	Quantity x Volume
OPRR621T60	Mouse-on-Rat HRP-Polymer	1 x 10.5 mL

* Refer to the Biocare Medical website located at http://biocare.net for information regarding catalog numbers and ordering.

Reconstitution, Mixing, Dilution, Titration:

The Mouse-on-Rat HRP-Polymer kit reagent(s) are optimized and ready to use with Biocare IHC antibodies and ancillary reagents. No reconstitution, mixing, dilution, or titration is required.

Species Reactivity:

Mouse IgG heavy and light chains with minimum cross-reactivity on rat tissues.

Supplied As:

Buffered saline solution, pH 7.6-7.8, containing a protein carrier and less than 0.01% ProClin 300 and/or less than 0.5% ProClin 950 as a preservative. See Safety Data Sheet for additional details.

Storage and Stability:

Store at 2°C to 8°C. The product is stable to the expiration date printed on the vial label when stored under these conditions. Do not use after expiration date. Storage under any condition other than those specified must be verified. The kit reagent(s) are ready-to-use and should not be diluted.

Staining Protocol Recommendations (ONCORE[™] Pro X Automated Slide Staining System):

The following programming and protocol recommendations are to assist the user when staining using Biocare's ONCORE Pro X Automated Staining Platform for research applications. The user is responsible for further optimizations of the protocol.

Mouse-on-Rat HRP-Polymer is provided in vials ready for use on the ONCORE Pro X Automated Slide Stainer. Uncap the vial and place in the ONCORE Pro X reagent tray. The ONCORE Pro X Automated Slide Stainer will apply reagent as required in the selected protocol. Refer to the appropriate antibody data sheet for the recommended staining protocol. Refer to the ONCORE Pro X Automated Slide Staining System User Manual for detailed instructions on instrument operation and additional protocol options.

Technical Notes:

1. Some primary mouse antibodies may not bind optimally with the secondary polymer; thus, a longer incubation time with the primary antibody may be required.

2. If endogenous rat IgG is observed in the negative control, add 1-2 drops of XR Factor (1 drop equals 33 $\mu L)$ to 2.5ml of Mouse-on-Rat HRP-Polymer and mix well.

3. Post-fixing tissue sections on slides for 15-30 minutes in 10% formalin reduces endogenous rat IgG and helps prevent tissues from falling off the slides.

4. Biocare's Background Punisher can be used as a blocker for nonspecific background staining.

Limitations:

This product is provided for Research Use Only (RUO) and is not for use in diagnostic procedures. Suitability for specific applications may vary and it is the responsibility of the end user to determine the appropriate application for its use.

Precautions:

1. Kit reagents contain less than 0.05% ProClin 300 and/or less than 1% ProClin 950. Wear gloves and protective clothing and take reasonable precautions when handling as ProClin is classified as an irritant and may cause skin contact sensitization. Avoid contact with eyes, skin, and mucous membranes.

2. Handle materials of human or animal origin as potentially biohazardous and dispose of such materials with proper precautions. In the event of exposure, follow the health directives of the responsible authorities where used.^{1,2}

3. 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 into contact with sensitive areas, wash with copious amounts of water.³

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

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

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

7. The reagent is optimized for use with Biocare antibodies and ancillary reagents. Refer to the primary antibody and other ancillary reagent instructions for use for recommended protocols and conditions for use.

Follow local and/or state authority requirements for method of disposal.
The SDS is available upon request and is located at http://biocare.net.

Technical Support:

Contact Biocare's Technical Support at 1-800-542-2002 for questions regarding this product.

Biocare Medical 60 Berry Drive Pacheco, CA 94553 USA

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Troubleshooting Guide:

- 1. Critical reagent (such as primary antibody) omitted.
- 2. Staining steps performed incorrectly or in the wrong order.
- 3. Heat-induced epitope retrieval (HIER) step was performed incorrectly using the wrong time, the wrong order, or the wrong pretreatment.
- 4. Insufficient amount of antigen.
- 5. Primary antibody incubation period too short.
- 6. Improperly mixed substrate and/or chromogen solution(s).
- Weak Staining
- 1. Tissue is either over-fixed or under-fixed.
- 2. Primary antibody incubation is too short.
- 3. Low expression of antigen.
- 4. Heat-induced epitope retrieval (HIER) steps performed incorrectly using
- wrong time, in the wrong order, or the wrong pretreatment.
- 5. Overdevelopment of substrate.
- 6. Excessive rinsing during wash steps.
- 7. Omission of critical reagent.
- 8. Incorrect procedure in reagent preparation.
- 9. Improper procedure in test steps.
- Non-specific or High Background Staining
- 1. Tissue is either over-fixed or under-fixed.
- 2. Incorrect blocking reagent used; blocker should be from same species in
- which the secondary antibody was raised.
- 3. Tissue may need a longer or a more specific protein block.
- 4. Substrate is overly developed.
- 5. Tissue was inadequately rinsed.
- 6. Deparaffinization incomplete.
- 7. Tissue damaged or necrotic.

Tissues Falling Off

- 1. Slides were not positively charged.
- 2. A slide adhesive was used in the water bath.
- 3. Tissue was not dried properly.
- 4. Tissue contained too much fat.
- Specific Staining Too Dark
- 1. Concentrated antibody not diluted out properly (being used at too high of a concentration).
- 2. Incubation of primary antibody or detection too long

References:

1. Occupational Safety and Health Standards: Occupational exposure to hazardous chemicals in laboratories. (29 CFR Part 1910.1450). Fed. Register. 2. Directive 2000/54/EC of the European Parliament and Council of 18 September 2000 on the protection of workers from risks related to exposure to biological agents at work.

3. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline-Fourth Edition CLSI document M29-A4 Wayne, PA 2014.