CDX2 (M) + CDH17 (RM)
Prediluted Multiplex Antibody Reagent
902-3135DS-090117

Catalog Number: APR 3135DS AA
Description: 6.0 ml, prediluted
Dilution: Ready-to-use
Diluent: N/A

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

Summary and Explanation:
CDX2 is a homeobox gene that encodes an intestine-specific transcription factor (1). CDX2 has been useful to establish gastrointestinal origin of metastatic adenocarcinomas and carcinoids and can be especially useful in distinguishing metastatic colorectal adenocarcinoma from tumors of unknown origin (1-7). CDX2 has been shown to be more specific and more sensitive than villin or CK20 (1,4,6). CDX2 has also been shown to be expressed in mucinous ovarian cancer, bladder adenocarcinoma, cholangiocarcinoma and malignant germ cell tumors of the testes (1,2,6-8). Only very rare examples of carcinomas of the genitourinary and gynecologic tracts or breast, lung, and head and neck cancers showed elevated levels of CDX2 expression (1).

CDH17 (Cadherin 17 or Li-cadherin) is a novel oncogene expressed in intestinal epithelium which is involved in tumor invasion and metastasis (9-10). CDH17 is a highly specific marker in colon cancer (99/99, 100%) and is a more sensitive marker than CDX2 (93/99, 94%) and CK20 (91/99, 92%) (11). Overexpression of CDH17 (and conversely, under expression of CDX2) correlates to poor prognosis in patients with epithelial ovarian cancer (1). CDH17 may be helpful for early diagnosis of Barrett’s esophagus (12). CDH17 has been shown to be a useful marker for distinguishing between primary urinary bladder adenocarcinoma and urothelial carcinoma with glandular differentiation (13). Note that CDH17 does not distinguish primary urinary bladder adenocarcinoma from colorectal adenocarcinoma secondarily involving the bladder (13). In addition, 89% of medullary carcinomas of the colon and rectum were recognized by CDH17 (14). Compared to CDX2 or CK20 alone, the combination of CDX2 and CDH17 is highly sensitive and somewhat specific for colorectal and stomach adenocarcinoma in routine immunohistochemistry, especially in cases with a CK7-/CDX2-/CK20- carcinoma (6,10,14). Data suggests that the combination of CDX2 and CDH17 along with CK7 may improve specificity compared to the panel consisting of CDX2, CDX2, villin and CK7 (11-13).

Principle of Procedure:
This product is a primary antibody cocktail of mouse and rabbit antibodies, which may be used in a Multiplex IHC staining procedure to produce a two-color stain. Following application of the primary antibody cocktail to the tissue sample, detection is performed by separate secondary antibodies specific for each species (i.e. mouse or rabbit) of the primary antibody cocktail, which are conjugated to horseradish peroxidase (HRP) or alkaline phosphatase (AP) enzymes. Visualization is accomplished by the application of chromogenic substrates (DAB and Warp Red), which are enzymatically activated (by HRP or AP, respectively) to produce a colored reaction product at the antigen site. The specimen may be counterstained and coverslipped. Results are interpreted using a light microscope.

Reagent Provided:
CDX2 (M) + CDH17 (RM) is provided as a prediluted antibody cocktail of anti-CDX2 and anti-CDH17 antibodies, in buffer with carrier protein and preservative.

<table>
<thead>
<tr>
<th>Antibody</th>
<th>Anti-CDX2</th>
<th>Anti-CDH17</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clone</td>
<td>CDX-88</td>
<td>EP96</td>
</tr>
<tr>
<td>Source</td>
<td>Mouse monoclonal</td>
<td>Rabbit monoclonal</td>
</tr>
<tr>
<td>Isotype</td>
<td>IgG1</td>
<td>IgG</td>
</tr>
<tr>
<td>Epitope/Antigen</td>
<td>CDX2</td>
<td>CDH17</td>
</tr>
<tr>
<td>Cellular Localization</td>
<td>Nuclear</td>
<td>Cytoplasmic/Cell membrane</td>
</tr>
<tr>
<td>Staining</td>
<td>Brown (DAB)</td>
<td>Red (Warp Red)</td>
</tr>
</tbody>
</table>

Storage and Stability:
Store at 2°C to 8°C. Do not use reagent after the expiration date printed on the vial. If reagents are stored under conditions other than those specified in the package insert, they must be verified by the user.

Known Applications:
Immunohistochemistry (formalin-fixed paraffin-embedded tissues)

Species Reactivity:
Human; others not tested

Positive Tissue Control:
Normal colon or colon cancer

Staining Protocol Recommendations:
Peroxide Block: Block for 5 minutes with Biocare’s Peroxidized 1.

Protein Block: Incubate for 10 minutes at RT with Biocare’s Background Purisher.

Primary Antibody: Incubate for 30 minutes at RT.

Double Stain Detection: Incubate for 30 minutes at RT using Biocare’s MACH 2 Double Stain 2.

Chromogen (1): Incubate for 5 minutes at RT with Biocare’s Betazoid DAB.

Chromogen (2): Incubate for 5-7 minutes at RT with Biocare’s Warp Red. Rinse in deionized water.

Counterstain: Counterstain with hematoxylin. Rinse with deionized water. Apply Tacha’s Bluing Solution for 1 minute. Rinse with deionized water.

Technical Notes:
This antibody has been standardized with Biocare’s MACH 2 Double Stain 2. Use TBS for washing steps.

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. This antibody contains less than 0.1% sodium azide. Concentrations less than 0.1% are not reportable hazardous materials according to U.S. 29 CFR 1910.1200, OSHA Hazard communication and EC Directive 91/155/EC. Sodium azide (NaN3) used as a preservative is toxic if ingested. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. Upon disposal, flush with large volumes of water to prevent azide build-up in plumbing. (Center for Disease Control, 1976, National Institute of Occupational Safety and Health, 1976) (15)
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. (16)
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.

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

References:
References Cont’d:
11. Tacha D, Zhou D. CDH17 is a highly specific marker and is a more sensitive marker than CDX2 and CK20 in colon cancers. Poster session presented at: CAP’14 The Pathologists’ Meeting; 2014 Sep 7-10; Chicago, IL.

Produced using Abcam’s RabMAB® technology. RabMAB® technology is covered by the following U.S. Patents, No. 5,675,063 and/or 7,429,487.