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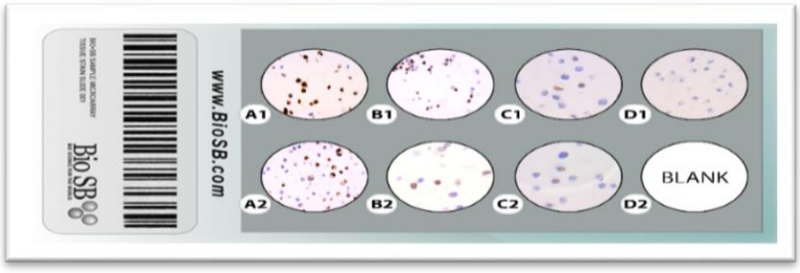
ER/PR PolyDetector HRP/DAB Detection System



Intended Use *For Research Use Only*

Summary And Explanation

The ER/PR PolyDetector HRP/DAB Detection System is a non-biotin, 2-step polymeric detection system that allows for the Immunohistochemical detection of Estrogen Receptor and Progesterone Receptor in tissues. The ER/PR PolyDetector HRP/DAB Detection System has been developed using a proprietary tandem hyperlabelling technology used to directly label immunoglobulins with enzymes. This ensures consistent and reproducible immunostaining for ER/PR. Included in the kit are Breast Tissue Microarray (TMA) slides of formalin-fixed paraffin-embedded tissues or cells that display 0, 1+, 2+ or 3+ positivity that enable the user to quantify the ER/PR expression in tested samples.

The Estrogen and Progesterone Receptor (ER/PR) proteins are nuclear hormone receptors overexpressed in various breast carcinomas. ER and PR strongly stain the nucleus of epithelial cells. The ER and PR are important regulators of growth and differentiation of the mammary gland. The progesterone receptor (PR) is an estrogen-regulated protein. It has been proposed that expression of PR determination indicates a responsive estrogen receptor (ER) pathway.

Presentation

The **ER/PR PolyDetector HRP/DAB Detection System** contains rabbit ER and PR prediluted antibodies clones RBT11 and RBT22, respectively, prediluted rabbit negative control, positive control slides with Breast Tissues, ImmunoRetriever with Citrate heat epitope retrieval solution, Peroxidase Blocker solution, an Anti-Rabbit Horseradish Peroxidase solution, a DAB Buffer and a DAB Chromogen solution. All the components are buffered and contain proteins, stabilizers and a non-azide anti-microbial.

Availability

Component Name	Catalog Number		
	BSB 0251 (70 Tests)	**BSB 0252 (150 Tests)**	**BSB 0253 (500 Tests)**
Rabbit Monoclonal ER Prediluted Antibody	7 mL	15 mL	50 mL
Rabbit Monoclonal PR Prediluted Antibody	7 mL	15 mL	50 mL
Rabbit Negative Control	7 mL	15 mL	50 mL
7-Core ER/PR Cell Line Microarray	5 Slides	10 Slides	30 Slides
20X ImmunoRetriever with Citrate	50 mL	100 mL	200 mL
PolyDetector Peroxidase Blocker	15 mL	30 mL	100 mL
PolyDetector HRP Label	15 mL	30 mL	100 mL
PolyDetector DAB Buffer	15 mL	30 mL	100 mL
PolyDetector DAB Chromogen	1 mL	3 mL	6 mL

Storage Store at 2-8^{0}C

Stability Stable up to the expiration date listed on the product label.
Do not use this product after the expiration date listed on the product label.

Protocol

Preparation of Working Solutions

The **Prediluted ER and PR Rabbit Antibodies, Prediluted Rabbit Negative Control, PolyDetector Peroxidase Blocker,** and **Anti-Rabbit Horseradish Peroxidase Label** are ready-to-use working solutions and require no further preparation. The **20X ImmunoRetriever with Citrate** needs to be diluted with distilled water to a 1X concentration before use by adding 50 mL of 20X ImmunoRetriever with Citrate to 950 mL of distilled water and mixing. **The DAB Chromogen** is concentrated and needs to be diluted and mixed into the **DAB Buffer** to produce the working DAB substrate-chromogen solution. For each 1 mL of working DAB substrate-chromogen solution required for the experiment, 1 drop of **DAB Chromogen** should be added and mixed into 1 mL of **DAB Buffer**.

Working DAB Substrate-Chromogen Required	1 mL	2 mL	3 mL
DAB Buffer	1 mL	2 mL	3 mL
DAB Chromogen	1 drop	2 drops	3 drops

Recommended Immunohistochemical Protocol

1. Deparaffinize and rehydrate tissues if necessary.
2. Place cut and dried slides in the prepared **1X ImmunoRetriever Citrate** and heat treat for 15 minutes in a pressure cooker or 45 minutes in a water bath or steamer at 95°C - 98°C.
3. Wash with 5 changes of IHC Wash buffer.
4. Cover tissue with the **PolyDetector Peroxidase Blocker** for 5 min.
5. Wash with 3 changes of IHC wash buffer.
6. Cover tissue with the ER or PR Primary Antibody or Negative Control and incubate for 45 minutes.
7. Wash with 3 changes of IHC wash buffer.
8. Cover tissue with the PolyDetector HRP Label, incubate for 45 min.
9. Rinse with 3 changes of IHC wash buffer.
10. Prepare DAB by adding one drop of **PolyDetector DAB Chromogen** per 1 mL of **PolyDetector DAB Buffer** and mix.
11. Cover tissue with the prepared DAB substrate-chromogen solution, incubate for 10 min.
12. Rinse with 5 changes of DI water.
13. Counterstain and then dehydrate.
14. Coverslip.

Abbreviated Immunohistochemical Protocol

Step	PolyDetector HRP
Peroxidase Blocker	5 minutes
Primary Antibody	45 minutes
HRP Label	45 minutes
DAB Substrate-Chromogen	10 minutes
Counterstaining	Time varies with counterstain

Interpretation of Expected Results with 7-Core ER/PR Cell Line Microarray

A1 **ER/PR +** (Metastatic Breast Cancer from pleural effusion)	**B1** **ER/PR +** (Ductal Breast Cancer)	**C1** **ER/PR +** (Metastatic Breast Cancer from pleural effusion)	**D1** **ER/PR Negative Control** (Metastatic Breast Cancer from pleural effusion)
A2 **ER/PR +** (Metastatic Breast Cancer from pleural effusion)	**B2** **ER/PR +** (Ductal Breast Cancer)	**C2** **ER/PR +** (Metastatic Breast Cancer from pleural effusion)	**D2** BLANK

Precautions

1. For professional users only. Results should be interpreted by a medical professional.
2. Always wear proper personal protective equipment such as laboratory coat, goggles and gloves when handling reagents.
3. Minimize microbial contamination of reagents.
4. Dispose of unused solution with copious amount of water.
5. Do not ingest reagent. If reagent is ingested, contact a poison control center immediately.
6. Avoid contact with eyes. If contact occurs, flush with large quantities of water.
7. For complete recommendations for handling biological specimens please refer to the CDC document, "Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories" (1).

References

1. U.S. Department of Health and Human Services: Centers for Disease Control and Prevention. Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories. Supplement / Vol. 61, January 6, 2012.

Symbol Key / Légende des symboles/Erläuterung der Symbole

 Storage Temperature Limites de température Zulassiger temperaturbereich	 Manufacturer Fabricant Hersteller	REF Catalog Number Référence du catalogue Bestellnummer
 Read Instructions for Use Consulter les instructions d'utilisation Gebrauchsanweisung beachten	 Expiration Date Utiliser jusque Verwendbar bis	LOT Lot Number Code du lot Chargenbezeichnung

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