

## **BrightVision, two steps colored detection system Goat Anti-Mouse/Rabbit IgG HRP BGX**

### **Instruction For Use**

**These instructions apply to the WellMed BrightVision; two steps colored detection system Goat Anti-Mouse/Rabbit HRP BGX.**

1. Intended use
2. Summary and explanation
3. Kit components
4. Availability
5. Recommended staining protocol
6. Control slides
7. Storage
8. Warnings and precautions
9. Troubleshooting
10. Reference

#### **1: Intended use**

For in-vitro diagnostic use.

WellMed BrightVision two steps colored detection system, peroxidase Goat Anti-Mouse/Rabbit IgG HRP BGX, is intended for use in immunohistochemistry for the detection of mouse or rabbit antibodies.

#### **2: Summary and explanation**

The WellMed BrightVision colored detection system, peroxidase Goat Anti-Mouse/Rabbit HRP BGX, is a ready-to-use system that has been manufactured to give an optimal staining, when using the protocol advised in this IFU.

Prior to staining some routine fixed, paraffin-embedded tissue sections should be subjected to pre-treatment (HIER or digestive enzyme).

The BrightVision detection system detects mouse or rabbit antibodies bound to an antigen in tissue sections. The antibodies are not provided but it is recommended to use the WellMed-antibodies. This polymer-complex is then visualized with a suitable substrate/chromogen. The substrate is not provided but it is recommended to use the WellMed-substrate. The clinical interpretation of any staining or its absence should be determined by a qualified pathologist and complemented by morphologic studies; controls should be evaluated within the context of the patient's clinical history and/or other diagnostic tests.

#### **3: Kit components**

WellMed BrightVision, two steps colored detection system, peroxidase Goat Anti-Mouse/Rabbit HRP BGX.

## 4: Availability

Catalog Number / Reference	Contents	Volume / Size
c-B55HRP-BGX	BrightVision, two steps colored detection system, Goat Anti-Mouse/Rabbit HRP BGX (ready-to-use) 1. Post-blocking (ready-to-use) (gold) 2. Polymer Goat Anti-Mouse/Rabbit HRP (ready-to-use) (ruby)	2x 55 ml 55 ml 55 ml
c-B110HRP-BGX	BrightVision, two steps colored detection system, Goat Anti-Mouse/Rabbit HRP BGX (ready-to-use) 1. Post-blocking (ready-to-use) (gold) 2. Polymer Goat Anti-Mouse/Rabbit HRP (ready-to-use) (ruby)	2x 110 ml 110 ml 110 ml
c-B500HRP-BGX	BrightVision, two steps colored detection system, Goat Anti-Mouse/Rabbit HRP BGX (ready-to-use) 1. Post-blocking (ready-to-use) (gold) 2. Polymer Goat Anti-Mouse/Rabbit HRP (ready-to-use) (ruby)	2x 500 ml 500 ml 500 ml
c-B1000HRP-BGX	BrightVision, two steps colored detection system, Goat Anti-Mouse/Rabbit HRP BGX (ready-to-use) 1. Post-blocking (ready-to-use) (gold) 2. Polymer Goat Anti-Mouse/Rabbit HRP (ready-to-use) (ruby)	2x 1000 ml 1000 ml 1000 ml

## 5: Recommended staining protocol

Step	Reagent	Template step	Incubation time
1	Deparaffinize and rehydrate tissue section	Slide/tissue preparing	-
2	Wash aqua dest	Wash	2x 5 min
3	If applicable; HIER or digestive enzyme	Pre-treatment	*
4	Wash buffer	PBS or TBS buffer	2x 5 min
5	H <sub>2</sub> O <sub>2</sub> (concentration 3%)	Tissue preparing	10 min
6	Wash buffer	PBS or TBS buffer	2x 5 min
7	Primary mouse or rabbit antibody	Antibody	30 min
8	Wash buffer	PBS or TBS buffer	2x 5 min
9	<b>Detection system, step 1, post-blocking</b>	Post-blocking	10 min
10	Wash buffer	PBS or TBS buffer	2x 5 min
11	<b>Detection system, step 2, polymer Mouse/Rabbit HRP</b>	Labeled polymer	10 min
12	Wash buffer	PBS or TBS buffer	2x 5 min
13	Substrate	DAB	*
14	Wash aqua dest	Wash	2x 2 min
15	Counterstain, dehydrate and coverslip	Auxiliary	-

\* See applicable IFU

## 6: Control slides

A positive control, negative control and reagent control are needed and processed in the same way as the unknown specimen slide to interpret staining results.

## 7: Storage

Store at 2-8 °C and in the dark. Do not use after expiration date.

## 8: Warnings and precautions

Refer to safety data sheet (SDS).

## 9: Troubleshooting

Please contact WellMed by phone or by email.

## 10: Reference

- 1) Shi ZR, Itzkowitz SH, Kim YS. A comparison of three immunoperoxidase techniques for antigen detection in colorectal carcinoma Tissues. *J Histochem Cytochem* 36: 317-322, 1988
- 2) Loos, C. M. van der. (2007). Multiple Immunoenzyme Staining: Methods and Visualizations for the Observation With Spectral Imaging. *Journal of Histochemistry & Cytochemistry*, 56(4), 313–328. doi:10.1369/jhc.2007.950170
- 3) NCCLS. Quality Assurance for Immunocytochemistry; Approved Guideline. NCCLS document MM4-A [1- 56238-396-5]. NCCLS, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 1999
- 4) Shi ZR, Au A, Soriano R et al: Non-Biotin Amplication (NBA) kit prevents nonspecific background staining of endogenous biotin induced by Heat Induced Epitope Retrieval (HIER) procedure. *The J Histotechnol* 23:327, 2000
- 5) Mount SL, Cooper K. Beware of biotin: a source of false-positive immunohistochemistry. *Current Diagnostic Pathology*. 2001; 7:161–167
- 6) Shi SR, Key ME, Kalra KL: Antigen retrieval in formalin-fixed paraffin embedded tissues: An enhanced method for immunohisto-chemical staining based on microwave oven heating of tissue sections. *J Histochem Cytochem* 39: 741-748, 1991
- 7) Shan-Rong Shi, James Guo, Richard J. Cote, Lillian Young, Debra Hawes, Yan Shi, Sandra Thu and Clive R. Taylor, *Applied Immunohistochemistry & Molecular Morphology*, vol 7, 201-208, 1999
- 8) Nakane PK and Pierce GB. Enzyme labeled antibodies: Preparations and applications for the localization of antigens. *Journal of Histochemistry and Cytochemistry*. 1967; 14:929–931
- 9) Battifora, H. Diagnostic Uses of Antibodies to Keratins: A Review and Immunohistochemical Comparison of Seven Monoclonal and Three Polyclonal Antibodies. In: Fenoglio-Preiser, C.M., Wolff, M., Rilke, F. (eds) *Progress in Surgical Pathology*. Springer, Berlin, Heidelberg. (1988)
- 10) Omata M, Liew CT, Ashcava M, Peters RL. Nonimmunologic binding of horseradish peroxidase to hepatitis B surface antigen: a possible source of error in immunohistochemistry. *American Journal of Clinical Pathology*. 1980; 73:626
- 11) Nadjji, M., & Morales, A. R. (1983). Immunoperoxidase: Part I. The Technique and Its Pitfalls. *Laboratory Medicine*, 14(12), 767–771. doi:10.1093/labmed/14.12.767