

Super Sensitive[™] Polymer-HRP IHC Detection For Manual and Open* Systems

Catalog No.	Description		
QD400-60KE	Super Sensitive [™] Polymer-HRP		
60 slides	Detection Kit, HRP/DAB		
QD420-YIKE	Super Sensitive [™] Polymer-HRP		
500 slides	Detection Kit, HRP/DAB		
QD430-XAKE	Super Sensitive [™] Polymer-HRP		
1000 slides	Detection Kit, HRP/DAB		
QD440-XAKE	Super Sensitive [™] Polymer-HRP		
1000 slides	Detection Kit,HRP		
QD550-YCXE	Xviz [™] Detection Kit for Xmatrx		
200 slides	Infinity		
QD550-YCDE	Xviz [™] Detection Kit for Xmatrx		
200 slides	Elite		
QD410-YAXE	Super Sensitive [™] Polymer-HRP		
200 slides	Detection System for i6000		

*Can be used on other open systems.

Intended Use

For In Vitro Diagnostic Use. Super Sensitive[™] Polymer-HRP Detection Systems are designed for the chromogenic detection of antigen-antibody binding reactions with mouse and/or rabbit IgG and IgM <u>primary antibodies</u> to achieve highly sensitive and specific immunohistochemical staining.

Summary and Explanation

Antigen detection by immunohistochemistry (IHC) is a two-step process wherein the primary antibody binds to specific epitopes of the antigen of interest and that binding is detected by a chromogen. BioGenex Super Sensitive[™] Polymer-HRP Detection Systems use a non-streptavidin-biotin proprietary micropolymer-complex technology to minimize background staining wherein an antibody enhancer/amplifier and a polymer-HRP reagent are bound to the primary antibody and visualized by diaminobenzidine (DAB).

Storage and Handling

Store at 2-8°C. Do not use after the printed expiration date.

Pretreatment

Routine tissue fixation can have adverse effects on antigenicity, leading to false-negative staining. Recovery of antigens can often be accomplished using Antigen Retrieval pretreatment or proteolytic digestion. Antigen Retrieval pretreatment increases staining intensity and reduces background staining. BioGenex offers Antigen Retrieval solutions covering a wide pH range, as that is an important factor for some antigens. **To determine which solution is best for each antibody, refer to the antibody datasheet.**

Quality Control

Run positive and negative controls simultaneously with all patient specimens. Follow the staining protocol for the kit and primary antibody exactly. Use a consistent volume of reagent across all slides.

Staining Procedure

The following IHC protocol is applicable to all Super SensitiveTM Polymer-HRP Manual Two-Step Detection Systems. See the Quality Control section and the user manual for more details.

- Do not allow tissue sections to dry out at any point during the rehydration and staining procedures.
- Always use freshly prepared DAB working solution at a ratio of 1 drop (40 ul) of DAB chromogen per 1 ml of substrate buffer.

Step	Incubation	Rinse
Dewax	10 min at Room	Alcohol 2 min, DI
	Temperature (RT)	water 5 min
Antigen Retrieval	See primary antibody datasheet	DI water x3
Peroxide Block	5 min at RT	PBS wash buffer x3
Power Block TM	5 min at RT	N/A
Primary Antibody	See primary antibody datasheet	PBS wash buffer x3
Super Enhancer TM	20 min at RT	PBS wash buffer x3
Poly-HRP	30 min at RT	PBS wash buffer x3
Substrate Solution	5-10 min at RT	DI water 5 min
Counterstain	1 min at PT	DI water 5 min,
		alcohol 2 min
Mount and Coverslip	N/A	N/A

Note: For automation, please refer to the factory default protocol on instrument software.

Limitations

It is recommended that reagents not be substituted across kit lot numbers. Interpretation of the staining result is solely the responsibility of the user. Experimental results should be confirmed by a medically-established diagnostic product or procedure. Evaluation must be performed by a qualified pathologist.

Improper tissue handling and processing prior to immunostaining can lead to inconsistent results. Variations in embedding and fixation or the nature of the tissue may lead to variations in results. Endogenous peroxidase activity or pseudo peroxidase activity in erythrocytes and tissue biotin may result in non-specific staining based on the detection system employed. Tissues containing Hepatitis B Surface Antigen (HBsAg) may give a false positive with horseradish peroxidase systems. Improper counterstaining and mounting may compromise the interpretation of results.

Troubleshooting

Refer to the troubleshooting section in the BioGenex Detection Kits user manual for remedial actions on detection system related issues, or contact BioGenex Technical Support Department at 1-800-421-4149 or support@biogenex.com or your local distributor to report unusual staining.

Category	Detection Systems	Revision No.	Н
Document No.	932-QD400-60KE.doc	Release Date	25-May-2022

48810 Kato Road, Suite 100E & 200E, Fremont, CA 94538, Tel: +1 (800) 421-4149, Fax: +1 (510) 824-1490, support@biogenex.com, www.biogenex.com



Precautions

Specimens and all materials exposed to them should be handled as if capable of transmitting infection and disposed of with proper precautions. Avoid microbial contamination of reagents to minimize non-specific staining. Wear suitable Personal Protective Equipment. Never pipette reagents by mouth. Avoid contact of reagents and specimens with skin and mucous membranes. If reagents or specimens come into contact with sensitive area, wash with copious amounts of water.

Some reagents in this kit contain sodium azide at concentrations of less than 0.1%. Sodium azide is not classified as a hazardous chemical at these concentrations, but proper handling protocols should be observed. DAB is classified as a possible carcinogen and can cause skin irritation upon contact. For more information on product hazards, precautions and waste disposal, Material Safety Data Sheets are available upon request. Dispose of unused reagents according to Local, State and Federal Regulations.

Reagents Available but Not Supplied

This section lists our most popular ancillary reagents and supplies, including reagents contained in some but not all of these kits, as laid out in the Materials and Reagents Provided table on p3. See the BioGenex Catalog for details and a complete listing of the reagents and sizes available.

Primary Antibodies	• Please refer to the BioGenex	
	Catalog for details.	
Dinco Buffor	• Phosphate Buffered Saline (PBS),	
Killse Dullei	pH 7.6 (HK091)	
Diluents for Primary Antibodies	 Common Antibody Diluent 	
	(HK156)	
	Enhanced Common Antibody	
	Diluent (HK941)	
Protein Blocks	• Peroxide Block (HK111)	
	• Power Block (HK083)	
Character Calastian	• Liquid DAB Chromogen (HK124)	
Chromogen Solution	Stable DAB Substrate Buffer	
Components	(HK520)	
E	• Pepsin (EK000)	
Enzymes for Tissue Digestion	• Trypsin (EK001)	
	• Protease XXIV (EK002)	
Antigen Retrieval Solutions	Please refer to the BioGenex	
	Catalog for details.	
Nagatina Control	Mouse Negative Control (HK119)	
Negative Controls	• Rabbit Negative Control (HK408)	
Counterstain	• Hematoxylin (HK100)	
	• Aqueous Mounting Media (HK099	
	SuperMount® Permanent Aqueous	
Mounting Media	Mounting Medium (HK079)	
	• XMount Permanent Mounting	
	Medium (HX035)	
Other Aneillery	 OptiPlus[™] Positive-Charged 	
Sunnlies	Microscope Slides (XT002)	
Supplies	• Micro chamber slides XT012,	

XT013, XT014

- Bibliography
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- 2. Centers for Disease Control. Decontamination of Laboratory Sink Drains to Remove Azide Salts. Center for Disease Control Manual Guide -- Safety Management, No. CDC-22, Atlanta, Georgia. April 30, 1976.
- 3. U.S. Department of Health and Human Services (NIOSH), Rockville, MD. Explosive azide hazard, Publication No. 78-127, 1976.
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- 5. Tacha, DE., et al., Modified antigen retrieval procedure: calibration technique for microwave ovens. J. Histotechnol. 17:365, (1994).
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