

BenchMark[™] His-tagged Protein Standard

Cat. no. LC5606	Size 125 μL	Store at -30°C to -10°C
Pub. Part no. LC5606.pps	MAN0001519	Rev. Date: 12 January 2012

Description

The BenchMark[™] His-tagged Protein Standard allows easy visualization of protein molecular weight ranges of His-tagged fusion proteins after electrophoresis or western blotting. Each standard protein contains a 6X His-tag enabling the detection of standard proteins with the InVision[™] His-tag In-gel Stain (Cat. no. LC6030) or with Anti-His(C-term) Antibody (Cat. no. R930-25) using western detection. The BenchMark[™] His-tagged Protein Standard is ideal for use as a positive control for InVision[™] His-tag In-gel Stain. The standard can also be detected with Coomassie staining.

The important features of the standard are listed below:

- Consists of 10 distinct protein bands in the range of 10-160 kDa
- Visualized with InVision[™] His-tag In-gel Stain (Cat. no. LC6030) or Coomassie stains on SDS-PAGE gels
- Visualized also with Anti-His(C-term) Antibody (Cat. no. R930-25) using chromogenic or chemiluminescent western detection systems
- Suitable for use with NuPAGE® Novex Gels or Tris-Glycine Gels
- Supplied in a ready-to-use format

Specifications

Contents:	125 μL of BenchMark [™] His-tagged Protein Standard
Storage	0.45 M Tris-HCl, pH 8.5; 50 mM DTT; 2% SDS; 12% glycerol;
Buffer:	0.0025% Coomassie G-250
Storage:	Store at -30°C to $-10^\circ\text{C}.$ Avoid repeated freezing and thawing.
Stability:	6 months at -20°C

For research use only. Not for human or animal therapeutic or diagnostic use.

Directions

The BenchMark[™] His-tagged Protein Standard is supplied in a ready-to-use format. There is no need to heat or add reducing agent.

- 1. Thaw the ladder at room temperature. Vortex gently to ensure the solution is homogenous.
- 2. Load the ladder onto a SDS gel using the loading volumes listed below to obtain the best results:

Gel	Electrophoresis	Blotting	
Mini-Gel, 1.0-mm	5 µL	2.5–5 μL*	
Mini-Gel, 1.5-mm	7 μL	5–10 µL*	

*The amount of standard will depend on the sensitivity of your western detection system and exposure time.

3. After electrophoresis, stain the standard proteins using the InVision[™] His-tag In-gel Stain (for staining protocol, refer to the manual supplied with the stain). After staining, the standard proteins are visualized using a UV transilluminator or a visible light laser-based scanner. The protein bands appear fluorescent as shown on the next page (lane A). The standard can also be stained with SimplyBlue[™] SafeStain (lane C, next page).

For western blotting with the Life Technologies Anti-His(C-Term) Antibody, use 1:5000 dilution and perform detection using the WesternBreeze® Chemiluminescent Kit-Anti-Mouse (lane B, page 3).

Note: The standard cannot be visualized using Anti-HisG Antibodies available from Life Technologies.

Product Qualification

The Certificate of Analysis (CofA) provides detailed quality control information for each product. The CofA is available on our website at **www.lifetechnologies.com/support**, and is searchable by product lot number, which is printed on each box.

Example

The apparent molecular weights of the protein bands in BenchMark[™] His-tagged Protein Standard are shown below. Five microliters of BenchMark[™] His-tagged Protein Standard was separated on a NuPAGE[®] Novex 4–12% Bis-Tris Gel with MES buffer. The standards were visualized after staining with InVision[™] His-tag In-gel Stain as described in the manual supplied with the stain (lane A), after western detection with 1:5000 dilution of Anti-His(C-Term) Antibody using the WesternBreeze[®] Chemiluminescent Kit-Anti-Mouse (lane B), or with SimplyBlue[™] SafeStain (lane C).



Note: If you do not observe any standard protein bands after staining with InVision[™] His-tag In-gel Stain, refer to the troubleshooting section of the manual supplied with the stain. For troubleshooting western detection with the antibody, refer to Anti-His(C-Term) Antibody manual.

Accessory Products

The following products are available separately. For more information, visit **www.lifetechnologies.com/support** or contact Technical Support.

Product	Quantity	Cat. no.
InVision [™] His-tag In-gel Stain	500 mL	LC6030
Anti-His(C-term) Antibody	50 µL	R930-25
WesternBreeze® Chromogenic Kit, Anti-Mouse	1 kit	WB7103
WesternBreeze [®] Chemiluminescent Kit, Anti-Mouse	1 kit	WB7104
SimplyBlue [™] SafeStain	1 L	LC6060

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