ALPHA DIAGNOSTIC INTERNATIONAL

LIFE TECH Life Technologies™ Your Molecular & Cell Technology Partner

Product Specification Sheet

HA-tag Antibodies and protein controls

Cat. HA12-P	HA tag control peptide	**SIZE**: 100 ug
Cat. HA12-A	Goat Anti-HA tag IgG (aff pure)	**SIZE**: 100 ug
Cat. HA12-HRP	Goat Anti-Ha tag IgG-HRP Conjugate	**SIZE**: 100 ul
Cat. HA12-FITC	Goat Anti-HA tag IgG-FITC conjugate	**SIZE**: 100 ul
Cat. HA12-BTN	Goat Anti-HA tag IgG-Biotin conjugate	**SIZE**: 100 ul

Expression of genes in E. coli or yeast or baculovirus offers a convenient system to produce large amounts of recombinant proteins that may otherwise be difficult to isolate from natural cells and tissues. Very often antibodies to these newly identified proteins are not available to study its biochemical properties, monitor protein expression, and purification. In order to circumvent this problem, short pieces of well-defined peptides (Poly-His, Flag-epitope or c-myc epitope or HA-tag) or small proteins (bacterial GST, MBP, Thioredoxin, b-Galactosidase, VSV-Glycoprotein etc) are often cloned along with the target gene. Proteins are expressed as fusion proteins. Antibodies to these fusion-tags are already available to monitor fusion protein expression and purification. Therefore, fusion-tags serve as universal tags much like secondary antibodies. Many tags have their own characteristics. Poly-His-fusion proteins (6 x His) can bind to Nickel-Sepharose or Nickel-HRP. GST-fusion proteins can bind to glutathione-Sepharose. Therefore, a high degree of purification of fusion protein can be achieved in just one affinity purification step. Purity of fusion proteins can be followed by Tag-antibodies. Very often, fusion proteins are directly injected into animals to generate antibodies. Some fusion tags can be removed later by treatment with enzymes to generate tag-free recombinant proteins.

Source of Antigen and Antibodies

A 9-aa peptide sequence (**designated cat # HA12-P; control peptide**) from hemagglutinin influenza virus (aa 114-122; YPY DVP DYA) (1) was synthesized and purifed by HPLC. It is supplied in PBS pH 7.4 at 100 ug/100 ul (1 mg/ml) in solution or powder form.

HA12-P was coupled to KLH and polyclonal antibodies were generated in goats. Control peptide was used for affinity purification of antibodies (**Cat # HA12-A**). it is supplied as 1 mg/ml solution in PBS 7.4 as preservative or as powder.

Recommended 2-ab

Rabbit Anti-goat IgG-HRP conjugate Cat # 30220 (AP, biotin, FITC conjugates also available)

Affinity pure Anti-HA tag IgG was coupled to peroxidase (HRP) using periodate method at the IgG:HRP ratio of 1:2. Antibody concn in the purified conjugate (**Ca # HA12-HRP**) is ~1 mg/ml. It is supplied in PBS pH 7.4 containing 0.05% thimerosal. Store at 4oc for short term (2-4 weeks). Store frozen in suitable aliquots. Avoid repeated freeze and thaw.

Purified anti-HA tag IgG was covalently coupled to FITC (Fluorescein isothiocyanate) (**Cat # HA12-FITC**). It is supplied in PBS pH 7.4, 0.2% BSA and 0.05% azide. Antibody concentration is ~ 1mg/ml. FITC:IgG (F/P) ratio is 3 to 7. Store at 4oC.

Cat# HA12-BTN, Biotin-conjugate

Purified goat anti-HA-tag antibody was coupled to Biotin using Biotinamidocaproate N-Hydroxysuccinimide Ester (BAC**)** at F/P ratio ~10-20:1. The antibody is supplied in PBS, pH 7.4, 0.2% BSA and 0.05% azide in either **lyophilized** (0.05 mg) or **liquid** form (0.05 mg/0.1 ml). Reconstitute powder in PBS in 0.1 ml to prepare 1 mg/ml solution. Store at -20oC in suitable aliquots. Stability is ~6-12 months. Do not freeze and thaw.

Suggested conjugate dilutions are 1:5,000-1:30,000 ELISA, 1:2K-1:10K for western.

Recommend reagent
cat # 20365 Streptavidin-Peroxidase (HRP) conjugate

Storage

Short-term: unopened, undiluted liquid vials at -20OC and powder at 4oC or -20oC..

Long-term: at –20C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

Stability: 6-12 months at –20oC or below.
Shipping: 4oC for solutions and room temp for powder.

Recommended Usage

Western Blotting (1-2 ug/ml using Chemiluminescence technique). Antibodies react with native and denatured myc-tag containing proteins. The antibodies detect the HA-tagged fusion proteins contain the tag at either the N or C-terminus.

ELISA : 01-1 ug/ml using 50-100 ng control antigen/well).

Histochemistry & Immunofluorescence: not tested. We recommend the use of affinity pure antibody at 2-5 ug/ml.

Antibody concentration must be optimized for each application under defined experimental conditions.

Control peptide (cat #HA12-P) , because of its small size, is not suitable for Western. It can be used to coat ELISA plates, dot blots, or used as antibody blocking peptide (use 10 ug peptide per ul/ug of antibody) to show antibody specificity. Recombinant purified HA-tag protein control for Western #HA11-C is available for control studies. Recombinant purified HA-tag protein #HA15-R can be used for ELISA etc.

General References: Gazin C et al (1984) EMBO J 3, 383-387; Tachibana K et al (1992) Gene, in press.

*This product is for In vitro research use only.

HA12-A-P-HRP-FITC-BTN 70405A

India Contact:
Life Technologies (India) Pvt. Ltd.
306, Aggarwal City Mall, Opposite M2K Pitampura, Delhi – 110034 (INDIA). Ph: +91-11-42208000, 42208111, 42208222, Mobile: +91-9810521400, Fax: +91-11-42208444
Email: customerservice@lifetechindia.com Website: www.lifetechindia.com