Kit Includes	Quantity	Applications	Reactivity	MW (kDa)	Isotype
GST (91G1) Rabbit mAb #2625	40 µl	W IP IF-IC	All		Rabbit IgG
Myc-Tag (71D10) Rabbit mAb #2278	40 µl	W IP IF-IC F	All		Rabbit IgG
HA-Tag (C29F4) Rabbit mAb #3724	40 µl	W IP IHC-P IF-IC F ChIP	All		Rabbit IgG
His-Tag Antibody #2365	40 µl	W IP IF-IC	All		Rabbit
DYKDDDDK Tag (9A3) Mouse mAb (Binds to same epitope as Sigma's Anti-FLAG®M2 Antibody) #8146	40 µl	W IP IHC-P IF-IC F	All		Mouse IgG1
Anti-rabbit IgG, HRP-linked Antibody #7074	100 µl				Goat
Anti-mouse IgG, HRP-linked Antibody #7076	100 µl				Horse

Applications Key: W=Western Blotting IP=Immunoprecipitation IHC-P=Immunohistochemistry (Paraffin) IF-IC=Immunofluorescence (Immunocytochemistry) F=Flow Cytometry ChIP=Chromatin IP

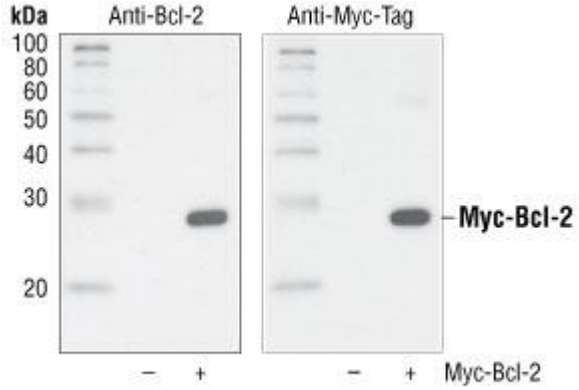
Reactivity Key: H=Human All=All Species Expected

Species enclosed in parentheses are predicted to react based on 100% sequence homology.

Specificity / Sensitivity

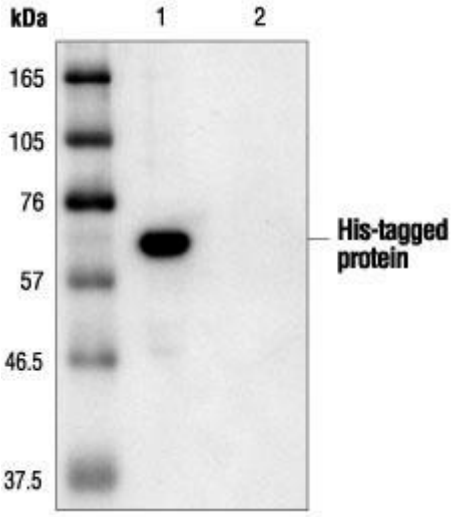
All antibodies in the Epitope Tag Antibody Sampler Kit detect overexpressed fusion proteins containing the corresponding epitope tags. DYKDDDDK Tag Antibody recognizes the DYKDDDDK peptide (the same epitope recognized by Sigma's Anti-FLAG® antibodies), and its binding specificity is NOT dependent on the presence of divalent metal cations.

Western Blotting



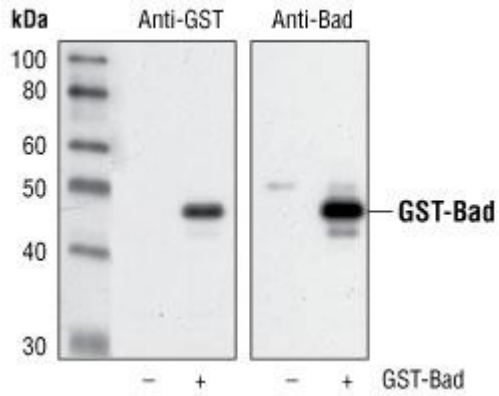
Western blot analysis of extracts from COS cells, untransfected or transfected with a construct overexpressing Myc-Bcl-2, using Bcl-2 Antibody #2872 (left) and Myc-Tag (71D10) Rabbit mAb #2278 (right).

Western Blotting



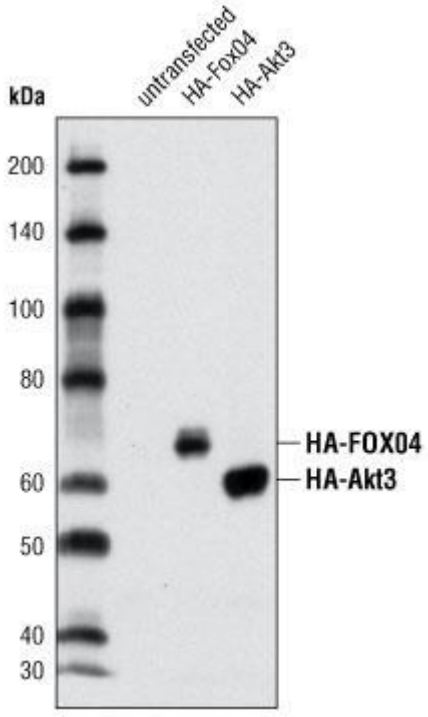
Western blot analysis of extracts from cells expressing C-terminal His-tagged protein (lane 1) or control extract (lane 2), using His-Tag Antibody #2365.

Western Blotting



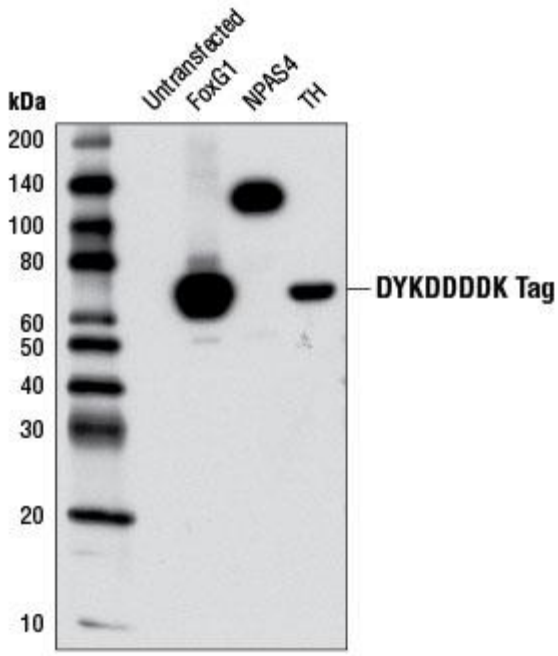
Western blot analysis of extracts from COS cells, untransfected (-) or transfected with a construct overexpressing GST-Bad (+), using GST (91G1) Rabbit mAb #2625 (left) and Bad Antibody #9292 (right).

Western Blotting



Western blot analysis of extracts from HeLa cells, untransfected or transfected with either HA-FoxO4 or HA-Akt3, using HA-Tag (C29F4) Rabbit mAb #3724.

Western Blotting



Western blot analysis of extracts from 293T cells, untransfected or transfected with either DYKDDDDK-tagged FoxG1, NPAS4, or Tyrosine Hydroxylase (TH), using DYKDDDDK Tag (9A3) Mouse mAb #8146.

Description

The Epitope Tag Antibody Sampler Kit provides an economical means to analyze the expression of a variety of epitope tagged proteins. The kit contains enough primary and secondary antibodies to perform four Western blots per primary antibody.

Source / Purification

Rabbit monoclonal antibodies are producted by immunizing rabbits with a synthetic peptide corresponding to residues 410-419 of human c-Myc (EQKLISEEDL), with a GST fusion protein or with a synthetic peptide containing the influenza hemagglutinin epitope (YPYDVPDYA). Mouse monoclonal antibodies are produced by immunizing animals with a synthetic DYKDDDDK peptide. Polyclonal antibodies are produced by immunizing rabbits with a synthetic DYKDDDDK peptide or with a 6xHis synthetic peptide. Polyclonal antibodies are purified by protein A and peptide affinity chromatography.

Background

Epitope tags are useful for the labeling and detection of proteins using immunoblotting, immunoprecipitation, and immunostaining techniques. Because of their small size, they are unlikely to affect the tagged protein's biochemical properties.

Several different epitope tags are now commonly utilized and readily available. For instance, a variety of plasmids contain DNA that encodes an amino-terminal tag consisting of six histidine (6xHis) residues followed by an extended multiple cloning site. The 6xHis tag on the expressed recombinant proteins allows for efficient coupling to Ni2+ affinity resins and purification by single step chromatography (1). As is the case with other protein tag systems (2), this polyhistidine tag can often be cleaved at sites recognized by proteases such as thrombin and enterokinases to isolate the protein of interest (1). Glutathione S-transferase (GST) is another widely used fusion partner, since it provides both an easily detectable Tag and a simple purification process with little effect on the biological function of the protein of interest. Numerous vectors containing GST-Tag have been developed for both prokaryotic and eukaryotic systems over the past decade (3-5). The HA tag, derived from an epitope of the influenza hemagglutinin protein, has also been extensively used as a general epitope tag in expression vectors (6), while the Myc epitope tag is routinely used to detect expression of recombinant proteins in bacteria, yeast, insect and mammalian cell systems (7). Finally, the DYKDDDDK peptide has been used extensively as a general epitope tag in expression vectors and consists of only eight amino acids. This peptide can be expressed and detected with the protein of interested as an amino-terminal or carboxy-terminal fusion (8).

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2. di Guan, C. et al. (1988) *Gene* 67, 21-30.
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