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Datasheet for K915

Epitope Tag Antibody Sampler Kit

Overview

Description:	Epitope Tag Antibody Sampler Kit - K915
Item No.:	K915
Size:	1 Kit
Applications:	WB
Reactivity:	GFP, 6X His-Tag, FLAG-Tag, GST-Tag

Product Details

Background:	Epitope tags are useful for the labeling and detection of proteins using western blotting, immunoprecipitation, and immunostaining techniques. Epitope Tag Antibody Sampler Kit detects overexpressed fusion proteins containing the corresponding epitope tags: FLAG™ tag (DYKDDDDK Tag Antibody) recognizes the DYKDDDDK peptide recognized by Sigma's Anti-FLAG® antibodies, 6X HIS antibody (His-tag or hexahistidine tag), GFP Antibody, and GST Antibody.
Synonyms:	FLAG, Green Fluorescent Protein, GFP, GST, six histidine, 6xHis

Target Details

Reactivity:	GFP, 6X His-Tag, FLAG-Tag, GST-Tag
Immunogen Type:	Conjugated Peptide
Immunogen:	GFP, GST, DYKDDDDK peptide (the same epitope recognized by Sigma's Anti-FLAG® antibodies), 6X His peptide
Purity/Specificity:	6X HIS antibody, GFP Antibody, DYKDDDDK Antibody (Binds to same epitope as Sigma's Anti-FLAG® M2 Antibody), GST Antibody and Conjugated Secondary Antibodies are all affinity purified.

Application Details

Tested Applications:	WB

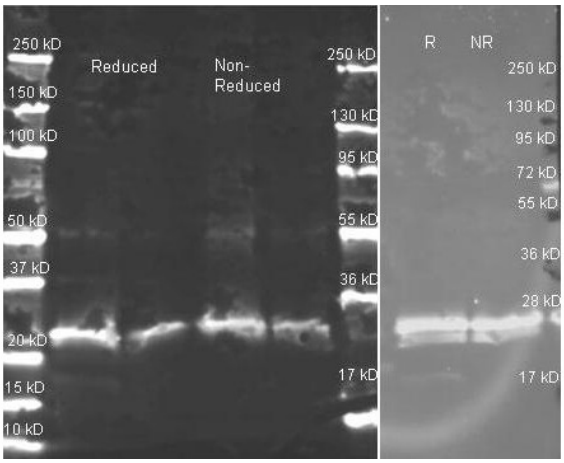
Application Note:	Epitope Tag kit contains: 6X HIS Antibody 100 µg DYKDDDDK Antibody (Binds to same epitope as Sigma's Anti-FLAG® M2 Antibody) 250 µg GFP Antibody 100 µg GST Antibody 1.0mg Anti-RABBIT IgG (H&L) (GOAT) Antibody Peroxidase Conjugated 100 µg Anti-GOAT IgG (H&L) (RABBIT) Antibody Peroxidase Conjugated 100 µg
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ChIP:	User Optimized
ELISA:	User Optimized
FC:	User Optimized
IF:	User Optimized
IHC:	User Optimized
IP:	User Optimized
WB:	User Optimized

Formulation

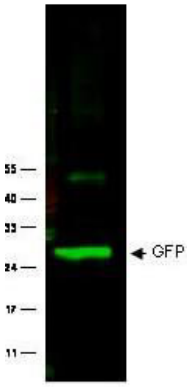
Shipping & Handling

Shipping Condition:	Dry Ice
Storage Condition:	Store Epitope Tag Kit at -20° C prior to opening. Aliquot antibodies and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.
Expiration:	Expiration date is one (1) year from date of receipt.

Images

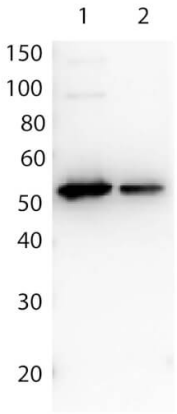


Western Blot
Rockland's anti-GST polyclonal antibody (600-101-200) in western blot shows detection of recombinant GST (indicated by band at ~ 28 kDa). The SDS-PAGE contained approximately 0.2 µg of rGST loaded on to a 4-20% gradient gel for separation. After electrophoresis, the gel was transferred to nitrocellulose and blocked with "Blocking Buffer for Fluorescent Western Blotting" p/n MB-070 in TBS for 1h at RT. The membrane was probed with anti-GST antibody at a 1:2,000 dilution in blocking reagent, overnight at 4° C. For detection DyLight™800 conjugated Donkey-a-Goat IgG (p/n 605-745-002) was used at a 1:20,000 dilution (in blocking reagent) for 30 min at 25° C. Fluorescent data was collected on a LICOR Odyssey instrument.



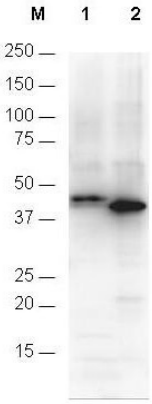
Western Blot
Rockland anti GFP polyclonal antibody (600-401-215) was used to detect GFP protein. Wild type GFP (0.1 µg) was used to spike 30 µg of a HeLa whole cell lysate. This antibody detects a 27 kDa band corresponding to the epitope tag GFP. A 4-20% Tris-Glycine gradient gel was used for SDS-PAGE. The protein was transferred to nitrocellulose using standard methods. After blocking with 5% BLOTTO in PBS, the membrane was probed overnight at 4° C with the primary antibody diluted in 5% BLOTTO to 1:1,000, followed by washes and reaction with a 1:10,000 dilution of IRDye® 800 conjugated Goat-a-Rabbit IgG [H&L] MX10 (611-132-122). IRDye® 800 fluorescence image was captured using the Odyssey® Infrared Imaging System developed by LI-COR. IRDye is a trademark of LI-COR, Inc. Other detection systems will yield similar results.

Kit Box
This product is assembled as a kit. See attached protocol or CofA for further details.



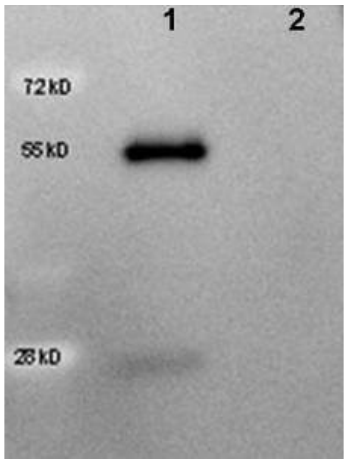
Western Blot

Rockland Affinity Purified anti FLAG™ Polyclonal Antibody (600-401-383) detects both C terminal linked and N terminal linked FLAG™ tagged recombinant proteins by western blot. This antibody was used at a dilution of 1:2,500 to detect 1.0 ?g of recombinant protein containing either the FLAG™ epitope tag linked at the carboxy (C) or the amino (N) terminus of the recombinant protein. A 4-20% gradient gel was used to resolve the protein by SDS-PAGE. The protein was transferred to nitrocellulose using standard methods. After blocking, the membrane was probed with the primary antibody for 1 h at room temperature followed by washes and reaction with a 1:10,000 dilution of IRDye® 800 conjugated Gt-a-Rabbit IgG (H&L) MX10 (code 611-132-122) for 30 min at room temperature. LICOR's Odyssey® Infrared Imaging System was used to scan and process the image. Other detection systems will yield similar results.



Western Blot

Anti-6X His epitope tag polyclonal antibody (600-401-382) detects His-tagged recombinant proteins by western blot. The blot was blocked with 3% BSA in TBST for 45 min at RT. Antibody was incubated with blot at a 1:1,000 dilution in TBST with 3% BSA for 1 hour at RT. Detection occurred using HRP Gt-a-Rabbit IgG (p/n 611-103-127) diluted 1:80,000 in blocking buffer (p/n MB-070) for 30 min at RT. Lane 1 was loaded with 12-Epitope Tag Protein Marker Lysate (p/n MB-301-0100) which has the His epitope tag incorporated through a C-terminal linkage (~42 kDa). Lane 2 was loaded with His-SUMO-GFP recombinant protein which has the His epitope tag incorporated through an N-terminal linkage (~40 kDa). A 4-20% gradient gel was used to resolve the protein by SDS-PAGE. Proteins were transferred to nitrocellulose using standard methods. Molecular weights were estimated by comparison to standards (lane M).



Western Blot
Western Blot of HRP anti-Goat IgG antibody (605-4313) showing detection of 50ng of goat IgG (lane 1) but not human IgG (lane 2). Samples were separated by 4-20% SDS-PAGE under reducing conditions and transferred to nitrocellulose membrane. The blot was blocked overnight at 4° C in 5% BSA in TBS. A 1:5,000 dilution of antibody in Blocking Buffer for Fluorescent Western Blotting (p/n MB-070) was used to probe the membrane at room temperature for 1 h. The image was developed using Chemiluminescent FemtoMax™ Super Sensitive HRP Substrate (p/n Femtomax-020) for one minute.

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