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Datasheet for ABIN3045984
anti-GST-Tag antibody

1 Validation

Overview

Quantity:	100 µg
Target:	GST-Tag
Reactivity:	Schistosoma japonicum
Host:	Mouse
Clonality:	Monoclonal
Application:	Western Blotting (WB), Immunoprecipitation (IP), Immunohistochemistry (IHC), Chromatin Immunoprecipitation (ChIP)

Product Details

Immunogen:	GST
Clone:	5C12
Isotype:	IgG1
Characteristics:	The GST tag issued in many protocols
Purification:	purified form hybridoma supernatant

Target Details

Target:	GST-Tag
Alternative Name:	GST Tag (GST-Tag Products)
Target Type:	Tag
Molecular Weight:	26 kDa

Target Details

UniProt: [P08515](#)

Application Details

Application Notes: WB 0.1-1.0ug/ml

Restrictions: For Research Use only

Handling

Format: Liquid

Concentration: 1 mg/mL

Buffer: PBS pH7.2

Storage: -20 °C



Successfully validated (Western Blotting (WB))

by [Celplor LLC](#)

Report Number: 029860

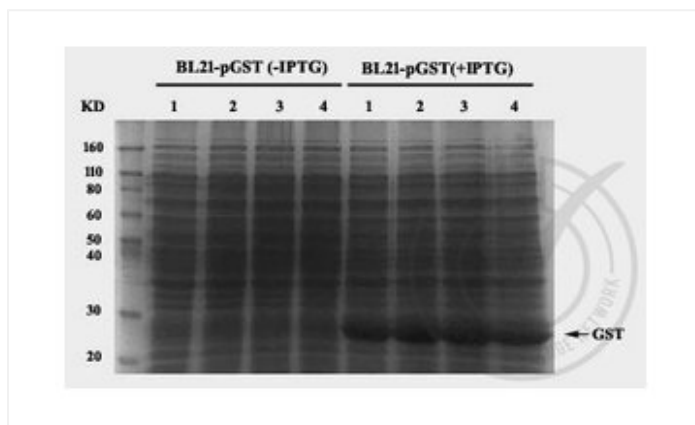
Date: May 19 2016

Target:	Mouse anti-GST monoclonal antibody
Lot Number:	1214
Method validated:	Western Blotting (WB)
Positive Control:	BL21 bacteria cells were transformed with an in-house GST expression vector pGST
Negative Control:	Empty vector cell lysate
Notes:	Based on the 12% GST content in total cell lysate of BL21-pGST (+IPTG), there is an estimated 1 ng of GST protein in 0.01 ug of total protein with IPTG induction. Therefore, the detection limit of mouse anti-GST monoclonal antibody is lower than 1 ng of GST protein.
Primary Antibody:	- Antibody: Mouse anti-GST monoclonal antibody - Supplier: Clonogene LLC - Supplier catalog number: CG0512 - Lot number: 1214 - Dilution: 1:1000
Secondary Antibody:	- Antibody: HRP labeled secondary anti-mouse antibody - Supplier: GE Healthcare Life Sciences - Supplier catalog number: NXA931 - Lot number: 390630 - Dilution: 1:2000
Controls:	<ul style="list-style-type: none">• Positive control: BL21 bacteria cells were transformed with an in-house GST expression vector pGST• Negative control: Empty vector cell lysate
Protocol:	<ul style="list-style-type: none">• **I. Reagent preparation**• 1. BL21 bacteria cells were transformed with an in-house GST expression vector pGST.• 2. Individual colony was inoculated in 2xYT medium and cultured at 37°C with shaking for 5 hrs followed by IPTG (1mM) induction for 2 hrs. Bacteria was grown to OD600=0.6-0.8 (5 hr) when IPTG was added.• 3. Cells were centrifuged at top speed and pellets were collected.• 4. Cell pellets were lysed in SDS-PAGE sample buffer.• 5. Protein concentration was assayed using DC protein assay kit (Bio-Rad).• 6. 50 ug of cell lysate was loaded and protein was separated by SDS-PAGE.• 7. Gel was stained with SimplyBlue Safe Stain (Invitrogen) at room temperature.• 8. GST content in IPTG induced samples was analyzed by Quantity One software (Bio-Rad).• **II. Western blot validation**• 1. Cell lysates from BL21 host cells, BL21-pGST transformed cells with or without IPTG

induction were loaded and proteins were separated by SDS-PAGE followed by Western transfer to a nitrocellulose membrane.

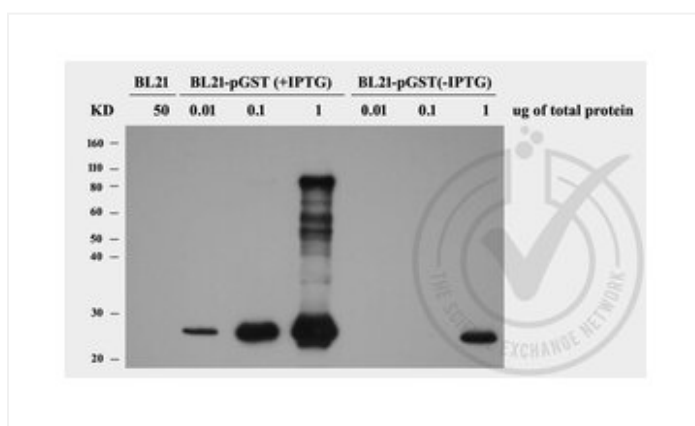
- 2. Membrane was blocked with 2% powder milk (Bio-Rad) for 1 hr at room temperature.
- 3. Membrane was incubated with mouse anti-GST monoclonal antibody (1:1000 dilution) at 4°C overnight with shaking.
- 4. Membrane was washed with TBST three times for 10 min.
- 5. After washing, membrane was incubated with HRP labeled secondary anti-mouse antibody (1:2000 dilution, GE Healthcare) with shaking at room temperature for 1 hr.
- 6. Membrane was washed with TBST three times for 10 min.
- 7. After washing, membrane was incubated with SuperSignal West Pico substrate (Thermo Scientific) for 5 min at room temperature.
- 8. Membrane was analyzed by ChemiDoc XRS gel documentation system (Bio-Rad).

Images for Validation report #029860



Validation image no. 1 for anti-GST-Tag antibody (ABIN3045984)

Figure 1: GST content of BL21-pGST(+IPTG) is 12% of total protein. GST content in IPTG induced samples was analyzed by Quantity One software (Bio-Rad).



Validation image no. 2 for anti-GST-Tag antibody (ABIN3045984)

Figure 2: Western blot for mouse anti-GST monoclonal antibody. The first lane shows the empty vector cell lysate (negative control). This is followed by a 0.01, 0.1, and 1 ug of total protein from BL21-pGST, with (Lanes 2, 3, and 4) or without (Lanes 5, 6, and 7) IPTG induction, respectively. Based on the 12% GST content in total cell lysate of BL21-pGST (+IPTG), there is an estimated 1 ng of GST protein in 0.01 ug of total protein with IPTG induction. Therefore, the detection limit of mouse anti-GST monoclonal antibody is lower than 1 ng of GST protein. The higher than expected

molecular weight may be the result GST dimers or aggregates (Riley et al. Protein Engineering vol.9 no.2 pp.223-230, 1996).