# abcam

## Product datasheet

## Anti-Glutathione Synthetase antibody [EPR6563] - BSA and Azide free ab236062





## 5 Images

#### Overview

**Product name** Anti-Glutathione Synthetase antibody [EPR6563] - BSA and Azide free

**Description** Rabbit monoclonal [EPR6563] to Glutathione Synthetase - BSA and Azide free

**Host species** Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), IHC-P, WB

Species reactivity Reacts with: Human

Predicted to work with: Mouse, Rat

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control 293T, HeLa, Daudi and HT-1080 cell lysates; Human colon tissue

**General notes** ab236062 is the carrier-free version of ab133592.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> is a trademark of Fluidigm Canada Inc.

This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

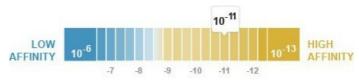
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#### **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at +4°C. Do Not Freeze.

**Dissociation constant (K<sub>D</sub>)**  $K_D = 2.74 \times 10^{-11} M$ 



Learn more about K<sub>D</sub>

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal
Clone number EPR6563

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab236062 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		Use at an assay dependent concentration. <b>ab172730</b> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB		Use at an assay dependent concentration. Predicted molecular weight: 52 kDa.

**Target** 

Pathway Sulfur metabolism; glutathione biosynthesis; glutathione from L-cysteine and L-glutamate: step

2/2.

Involvement in disease Defects in GSS are the cause of glutathione synthetase deficiency (GSS deficiency)

[MIM:266130]; also known as 5-oxoprolinuria or pyroglutamic aciduria. It is a severe form characterized by an increased rate of hemolysis and defective function of the central nervous

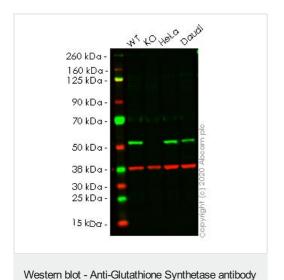
system.

Defects in GSS are the cause of glutathione synthetase deficiency of erythrocytes (GLUSYNDE) [MIM:231900]. Glutathione synthetase deficiency of erythrocytes is a mild form causing hemolytic anemia.

#### Sequence similarities

Belongs to the eukaryotic GSH synthase family.

#### **Images**



[EPR6563] - BSA and Azide free (ab236062)

All lanes: Anti-Glutathione Synthetase antibody [EPR6563] (ab133592) at 1/1000 dilution

Lane 1: Wild-type HEK-293T cell lysate

Lane 2: GSS knockout HEK-293T cell lysate

Lane 3: HeLa cell lysate Lane 4: Daudi cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

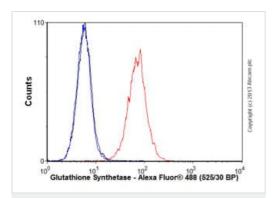
Observed band size: 50 kDa

Predicted band size: 52 kDa

This data was developed using the same antibody clone in a different buffer formulation (ab133592).

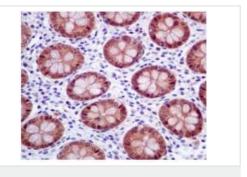
Lanes 1-4: Merged signal (red and green). Green - ab133592 observed at 50 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.

ab133592 was shown to react with GSS in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266342 (knockout cell lysate ab257460) was used. Wildtype HEK-293T and GSS knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab133592 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup>680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Flow Cytometry (Intracellular) - Anti-Glutathione Synthetase antibody [EPR6563] - BSA and Azide free (ab236062)

Overlay histogram showing Jurkat cells stained with <u>ab133592</u> (red line). The cells were fixed with 80% methanol (5 min)/ and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (<u>ab133592</u>, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor<sup>®</sup> 488 goat anti-rabbit lgG (H&L) (<u>ab150077</u>) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit lgG (monoclonal) (0.1µg/1x10<sup>6</sup> cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab133592</u>).

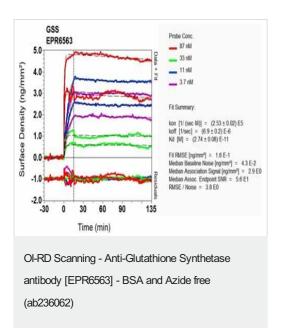


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Glutathione Synthetase antibody [EPR6563] - BSA and Azide free (ab236062)

Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling Glutathione Synthetase with <u>ab133592</u> at 1/50 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab133592</u>).

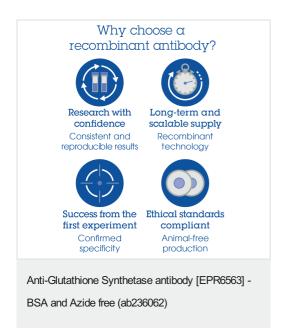
Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.



Equilibrium disassociation constant ( $K_D$ ) Learn more about  $K_D$ 

#### Click here to learn more about KD

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab133592</u>).



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