abcam

Product datasheet

Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free ab267397

Recombinant RabMAb

13 Images

Overview

Product name	Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free
Description	Rabbit monoclonal [EPR22927-54] to Thrombospondin 1 - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), WB, ICC/IF, IHC-P, IP
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: 3T3-L1 starved with 0.4% serum for 24 hours, then cultivated with 15% serum for 6 hours, whole cell lysate. HUVEC and mouse platelet lysates and rat platelet whole cell lysate. IHC-P: Human spleen, human bone marrow, human cervical carcinoma and mouse spleen tissues. ICC/IF: HUVEC cells and PC-12 cells. Flow Cyt (intra): HUVEC, 3T3-L1 and PC-12 cells. IP: HUVEC and mouse platelets lysate.
General notes	ab267397 is the carrier-free version of **ab267388**. 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 cell-based 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® 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**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR22927-54
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab267397 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.
WB		Use at an assay dependent concentration. Predicted molecular weight: 129 kDa.
ICC/IF		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IP		Use at an assay dependent concentration.

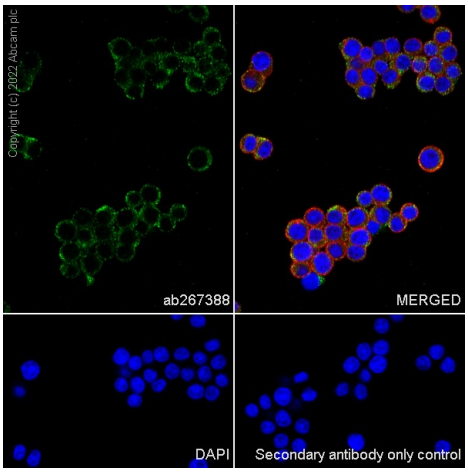
Target

Function	Adhesive glycoprotein that mediates cell-to-cell and cell-to-matrix interactions. Binds heparin. May play a role in dentinogenesis and/or maintenance of dentin and dental pulp (By similarity). Ligand for CD36 mediating antiangiogenic properties. Plays a role in ER stress response, via its interaction with the activating transcription factor 6 alpha (ATF6) which produces adaptive ER stress response factors.
Sequence similarities	Belongs to the thrombospondin family. Contains 2 EGF-like domains. Contains 1 laminin G-like domain. Contains 1 TSP C-terminal (TSPC) domain.

Contains 3 TSP type-1 domains.
Contains 8 TSP type-3 repeats.
Contains 1 VWFC domain.

Cellular localization Endoplasmic reticulum. Sarcoplasmic reticulum.

Images

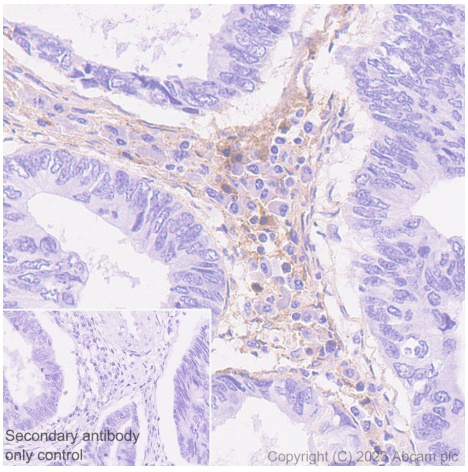


Immunocytochemistry/ Immunofluorescence - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

Immunofluorescent analysis of 100% Methanol-fixed, 0.1% TritonX-100 permeabilized PC-12 (rat adrenal gland pheochromocytoma cell) cells labelling Thrombospondin 1 with **ab267388** at 1/100 (5.3 µg/ml) dilution, followed by **ab150077** AlexaFluor® 488 Goat anti-Rabbit secondary antibody at 1/1000 (2 µg/ml) dilution (Green). Confocal image showing cytoplasmic staining in PC-12 cell line. Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8). **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 (2.5 µg/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is **ab150077** AlexaFluor® 488 Goat anti-Rabbit secondary at 1/1000 (2 µg/ml) dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.



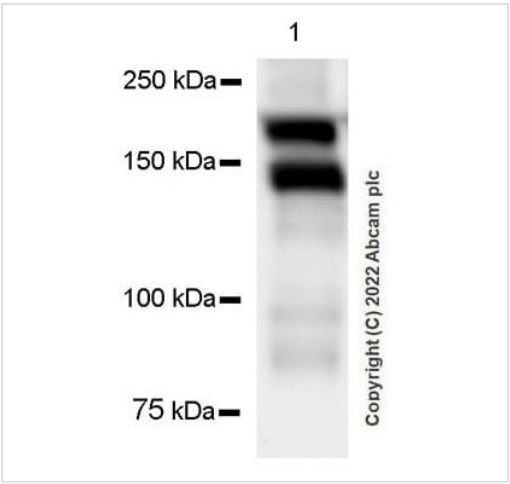
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

Immunohistochemical analysis of paraffin-embedded Human cervical carcinoma tissue labelling Thrombospondin 1 with **ab267388** at 1/5000 (0.101 µg/ml) followed by a Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) at a Ready to use dilution. Positive staining on extracellular matrix of human cervical carcinoma. The section was incubated with **ab267388** for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) at Ready to use dilution.

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.



Western blot - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

Anti-Thrombospondin 1 antibody [EPR22927-54] (**ab267388**) at 1/1000 dilution + Rat platelet whole cell lysate at 20 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 129 kDa

Observed band size: 180,140 kDa

Blocking buffer and concentration : 5% NFDM/TBST

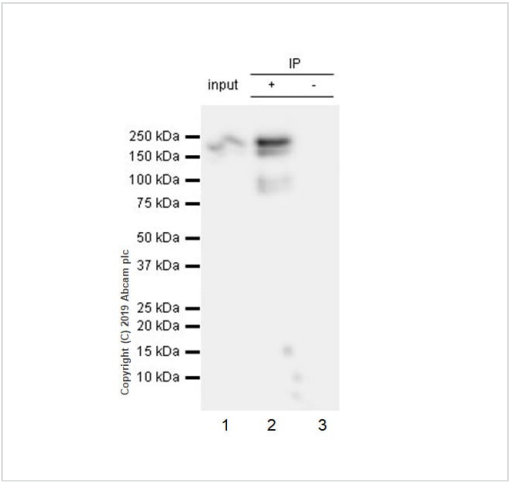
Diluting buffer and concentration : 5% NFDM/TBST

The full-length TSP 1 (180 kDa) and a ~140 kDa band, likely to be a TSP 1 isoform or fragment, are observed.

The molecular weight observed is consistent with what has been described in the literature (PMID:1426766, 27588705).

Exposure time : 5.5 seconds

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.



Immunoprecipitation - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.

Thrombospondin 1 was immunoprecipitated from 0.35 mg mouse platelets whole cell lysate 10µg with **ab267388** at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using **ab267388** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.

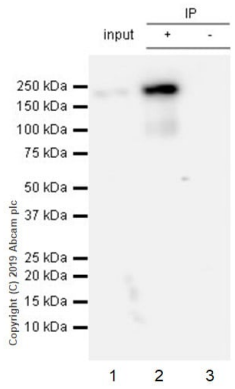
Lane 1: Mouse platelets whole cell lysate 10µg.

Lane 2: **ab267388** IP in mouse platelets whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab267388** in mouse platelets whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 8 seconds.



Immunoprecipitation - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.

Thrombospondin 1 was immunoprecipitated from 0.35 mg HUVEC (human umbilical vein endothelial cell) whole cell lysate 10µg with **ab267388** at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using **ab267388** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used at 1/5000 dilution.

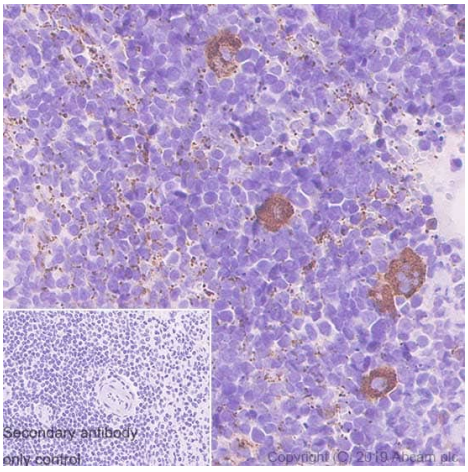
Lane 1: HUVEC whole cell lysate 10µg.

Lane 2: **ab267388** IP in HUVEC whole cell lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab267388** in HUVEC whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 8 seconds.



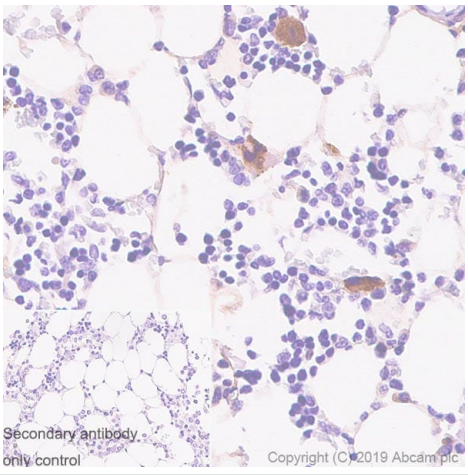
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling Thrombospondin 1 with **ab267388** at 1/5000 dilution (0.1 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer). Positive staining on the megakaryocytes and platelets in the mouse spleen is observed. The section was incubated with **ab267388** for 10 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.



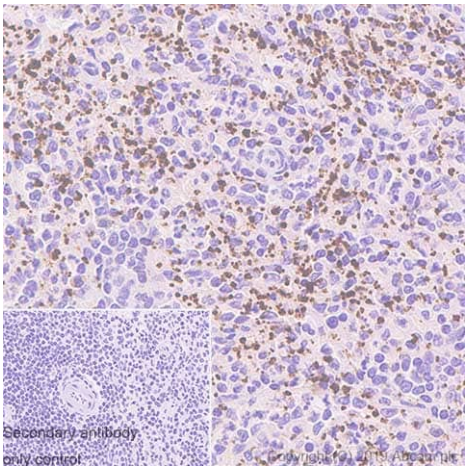
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.

Immunohistochemical analysis of paraffin-embedded human bone marrow tissue labeling Thrombospondin 1 with **ab267388** at 1/5000 dilution (0.1 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer). Positive staining on the megakaryocytes in the human bone marrow (PMID: 28239144). The section was incubated with **ab267388** for 10 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.



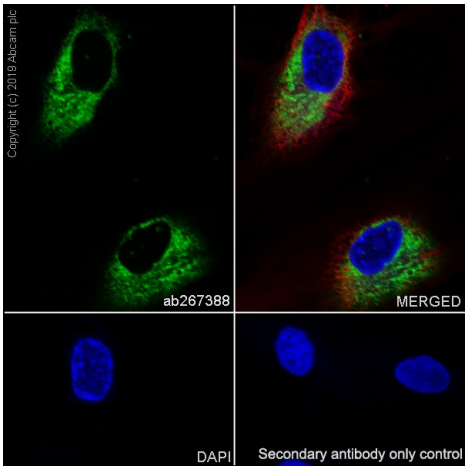
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.

Immunohistochemical analysis of paraffin-embedded human spleen tissue labeling Thrombospondin 1 with **ab267388** at 1/5000 dilution (0.1 µg/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer). Positive staining on the platelets in the human spleen (PMID: 28239144). The section was incubated with **ab267388** for 10 mins at room temperature. The immunostaining staining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP Polymer).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.



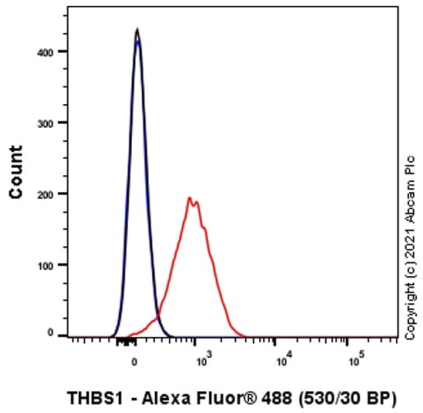
Immunocytochemistry/ Immunofluorescence - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.

Immunofluorescent analysis of 100% methanol-fixed, permeabilized HUVEC (human umbilical vein endothelial cell) cells labeling Thrombospondin 1 with **ab267388** at 1/100 dilution (5 µg/ml), followed by **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary antibody at 1/1000 dilution (Green). Confocal image showing cytoplasmic staining in HUVEC cell line is observed. **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 dilution.

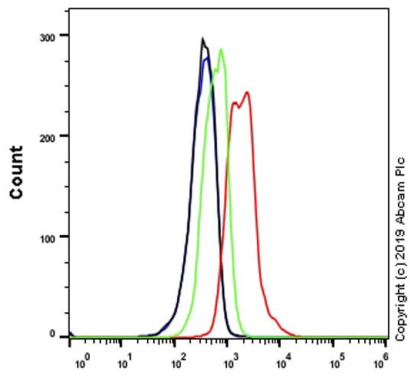
100% methanol fixation is recommended.



Flow Cytometry (Intracellular) - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

Intracellular Flow Cytometry analysis of PC-12 (Rat adrenal gland pheochromocytoma cell line) cells labeling Thrombospondin 1 with **ab267388** at 1/500 dilution (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat Anti-rabbit IgG (Alexa Fluor® 488, **ab150081**) secondary antibody was used at 1/5000 dilution. Isotype control - Rabbit monoclonal IgG (Black) (**ab172730**). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).

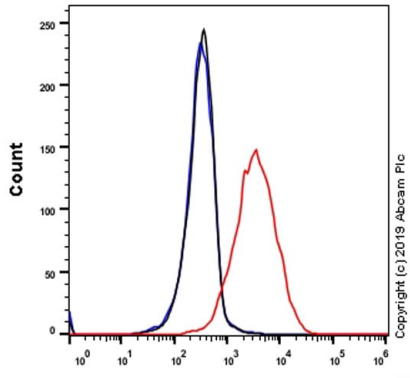
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.



Flow Cytometry (Intracellular) - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.

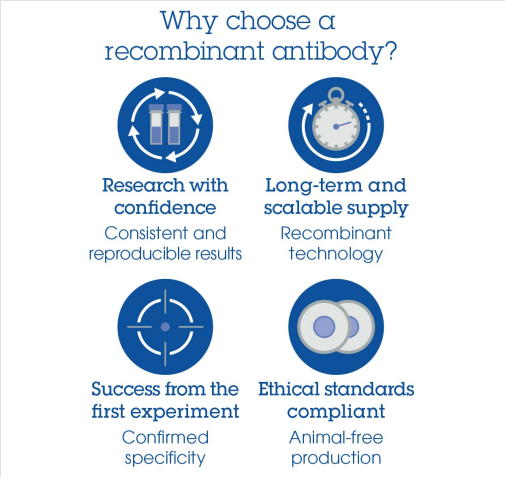
Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized 3T3-L1 (mouse embryonic fibroblast) starved with 0.4% serum for 24h, then cultured with 15% serum for 6h (Red) / Untreated control (Green) cells labeling Thrombospondin 1 with **ab267388** at 1/50 (Red) compared with a Rabbit monoclonal IgG (**ab172730**) / Black isotype control and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.



Flow Cytometry (Intracellular) - Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol and sodium azide **ab267388**.

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized HUVEC (human umbilical vein endothelial cell) cells labeling Thrombospondin 1 with **ab267388** at 1/50 (Red) compared with a Rabbit monoclonal IgG (**ab172730**) / Black isotype control and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.



Anti-Thrombospondin 1 antibody [EPR22927-54] - BSA and Azide free (ab267397)

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