

Anti-Flagellin FliC

Monoclonal antibody against flagellin from *Salmonella typhimurium*

Catalog code: mabg-flic

<https://www.invivogen.com/anti-flagellin>

For research use only, not for diagnostic or therapeutic use

Version 22K09-MM

PRODUCT INFORMATION

Contents: 100 µg of purified Anti-Flagellin FliC, provided azide-free and lyophilized

Target: Flagellin from *Salmonella typhimurium*

Specificity: Purified or recombinant flagellin from *S. typhimurium*

Clone: X5A12

Isotype: Mouse IgG1

Light chain type: Kappa

Formulation: 0.2 µm filtered solution in a sodium phosphate buffer with glycine, saccharose, and stabilizing agents

Applications: Detection by ELISA and Western blot

Antibody resuspension (0.1 mg/ml)

Add 1 ml of sterile water per 100 µg vial.

Storage and stability

- Product is shipped at room temperature. Upon receipt, store lyophilized antibody at -20°C.
- Reconstituted antibody is stable for 1 month at 4°C and for 1 year at -20°C. Avoid repeated freeze-thaw cycles.

Quality control

- This product has been validated by ELISA.
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

BACKGROUND

Flagellin is the major component of the bacterial flagellar filament, which confers motility to a wide range of bacterial species. This pro-inflammatory molecule is present in Gram-negative and Gram-positive bacteria. It is recognized by distinct types of pattern recognition receptors (PRRs). Extracellular flagellin is recognized by surface-localized Toll-like receptor 5 (TLR5), which promotes the activation of NF-κB and the subsequent production of cytokines^{1,2}. Flagellin is also recognized by the cytosolic NOD-like receptors (NLRs) NAIP5/NAIP6 sensors of the NLR4 inflammasome^{3,4}. Flagellin monomers are translocated into the host cell cytosol by a mechanism that requires bacterial secretion systems. This triggers the formation of an NAIP-NLR4 inflammasome leading to caspase-1-mediated secretion of IL-1β and IL-18^{3,4}.

1. Song WS. et al., 2017. A conserved TLR5 binding and activation hot spot on flagellin. *Sci Rep.* 7:40878. 2. Hayashi F. et al., 2001. The innate immune response to bacterial flagellin is mediated by Toll-like receptor 5. *Nature.* 410(6832):1099-103. 3. Duncan JA. & Canna SW., 2018. The NLR4 Inflammasome. *Immunol Rev.* 281(1):115-123. 4. Zhao Y. et al., 2011. The NLR4 inflammasome receptors for bacterial flagellin and type III secretion apparatus. *Nature.* 477(7366):596-600.

DESCRIPTION

Anti-Flagellin FliC is a mouse monoclonal antibody against flagellin from *S. typhimurium*. It was generated by DNA immunization of Balb/c mice with the *fliC* gene. This antibody has been produced in hybridomas and purified by affinity chromatography.

Anti-Flagellin FliC was screened for its ability to bind flagellin from *S. typhimurium* using ELISA. It can be used to detect purified or recombinant flagellin from *S. typhimurium* using ELISA and Western blot.

APPLICATIONS

Anti-Flagellin FliC can be used for ELISA and Western blot.

Indirect ELISA with HRP-conjugated secondary antibody

1. Coat wells of a transparent 96-well ELISA plate with 50 µl of RecFLA-ST (recombinant flagellin from *S. typhimurium*) at 1 µg/ml per well in a carbonate/bicarbonate coating buffer and incubate overnight at room temperature (15-25°C). Wash the microtiter plate with phosphate-buffered saline (PBS) containing 0.05% Tween® 20 (PBS-T).
2. Add 200 µl phosphate-buffered saline (PBS) containing 4% bovine serum albumin (BSA) to each well to prevent non-specific binding and incubate for 2-4 hours at 37°C. Wash the microtiter plate with PBS-T.
3. Distribute 150 µl of Anti-Flagellin FliC (100 ng-10 µg/ml final concentration) diluted in PBS-T containing 1% BSA to each well and incubate for 1 hour at 37°C. Wash the microtiter plate twice with PBS-T.
4. Add 100 µl of HRP-conjugated secondary antibody diluted in PBS-T containing 1% BSA to each well and incubate for 30 min to 1 hour at 37°C. Wash the microtiter plate twice with PBS-T.
5. Add HRP substrate and read absorbance using a microplate reader.

Western blot

Recommended working dilution: 0.1-1 µg/ml

RELATED PRODUCTS

Product	Description	Cat. Code
Mouse IgG1 Control	Isotype control antibody	mabg1-ctrlIm
FLA-ST	Flagellin <i>S. typhimurium</i>	tlrl-stfla
FLA-ST Ultrapure	Ultrapure flagellin	tlrl-epstfla
Rec FLA-ST	Recombinant flagellin	tlrl-flic-10

TECHNICAL SUPPORT

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