



Annexin V / Annexin A5 (RUU-WAC2A)

CATALOG NUMBER: MUB0106S

CLONE: RUU-WAC2A

ISOTYPE: Mouse IgG1

PRODUCT FORM: culture supernatant of monoclonal antibody

BACKGROUND

Annexin A5 (or annexin V) is a cellular protein of the annexin family. Annexin A5 has been proposed to play a role in the inhibition of blood coagulation by competing for phosphatidylserine binding sites with prothrombin, and also to inhibit the activity of phospholipase A1. Annexin A5 forms a shield around negatively-charged phospholipid molecules, which blocks the entry of phospholipids into coagulation (clotting) reactions. Antibodies directed against annexin A5 are the cause of the antiphospholipid syndrome in which the formation of the shield is disrupted by antibodies. Without the shield, there is an increased quantity of phospholipid molecules on cell membranes, speeding up coagulation reactions and causing the blood-clotting characteristic of the antiphospholipid antibody syndrome.

Annexin A5 is used as a probe in the annexin A5 affinity assay to detect cells that have expressed phosphatidylserine on the cell surface, a feature found in apoptosis as well as other forms of cell death (1,2). Platelets also expose phosphatidylserine on their surface when activated, which serves as binding site for various coagulation factors.

Annexin A5 has been shown to interact with kinase insert domain receptor (3) and integrin beta 5 (4).

RUU-WAC2a is a monoclonal that specifically reacts with annexin A5. This monoclonal antibody is directed to the phospholipid binding domain of annexin A5 and as such it inhibits annexin A5 binding to anionic phospholipid membranes for more than 95%. RUU-WAC 2A does not remove annexin A5 bound to phospholipid surfaces indicating that the epitope is not accessible after interaction with annexin A5.

SOURCE

RUU-WAC2a is a mouse monoclonal IgG1 antibody obtained by fusion of P3-X63-Ag 8,653 mouse myeloma cells with spleen cells from a BALB/c mouse immunized with recombinant annexin A5.

PRODUCT

Each vial contains 1 ml of culture supernatant of monoclonal antibody containing 0.09% sodium azide.

SPECIFICITY

RUU-WAC2A reacts against human recombinant and endogenous annexin A5. It shows no cross reactivity with Annexins I, II, III, IV, VI, VII and VIII in ELISA or in immunoprecipitation. It does not recognise monkey and rat Annexin V in ELISA.

The monoclonal antibody is suitable for ELISA in combination with a polyclonal antibody raised against Annexin V in rabbit. It can be applied in immunofluorescence to paraformaldehyde fixed cell samples and in immunohistochemistry of frozen sections and formaldehyde fixed and paraffin embedded tissue specimens. For immunohistochemistry of paraffin sections a pretreatment step with 2% amino-propyl-triethoxysilan (in 100% ethanol) and 0,2 % BSA is recommended. The sections are heated for 15 minutes at 100°C in a citrate buffer (10 mM trisodium citrate, pH 6,0). The sections are then further processed by routine procedures (see below).

RUU-WAC2a inhibits annexin A5 binding to anionic phospholipid-containing vesicles, activated platelets and apoptotic cells for more than 95%.

Optimal antibody dilution should be determined by titration; recommended range is 1:50 – 1:100 for immunohistochemistry with avidin-biotinylated horseradish peroxidase complex (ABC) as detection reagent, and 1:100 – 1:500 for immunoblotting applications.

SPECIES REACTIVITY

Human.

Not reactive with monkey and rat.

STORAGE

Store at 4°C, or in small aliquots at –20°C.

REFERENCES

1. Koopman, G., Reutelingsperger, C.P., Kuijten, G.A.M. et al. (1994). Annexin V for flow cytometric detection of phosphatidylserine expression on B cells undergoing apoptosis. *Blood* 84, 1415-20.
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WARNING and CAUTION

This product is intended FOR RESEARCH USE ONLY, and FOR TESTS IN VITRO, not for use in diagnostic or therapeutic procedures involving humans or animals.

This product contains sodium azide. To prevent formation of toxic vapors, do not mix with strong acidic solutions. To prevent formation of potentially explosive metallic azides in metal plumbing, always wash into drain with copious quantities of water.

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3. Wen, Y., Edelman J.L., Kang, T. and Sachs, G. (1999). Lipocortin V may function as a signaling protein for vascular endothelial growth factor receptor-2/Flk-1. *Biochem. Biophys. Res. Commun.* **258**, 713–21.
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