



APC Annexin V

Catalog# / Size

640919 / 25 tests 640920 / 100 tests 640941 / 300 tests

Regulatory Status

RUO

Other Names

Annexin A5

Description

Annexin V (or Annexin A5) is a member of the annexin family of intracellular proteins that binds to phosphatidylserine (PS) in a calcium-dependent manner. PS is normally only found on the intracellular leaflet of the plasma membrane in healthy cells, but during early apoptosis, membrane asymmetry is lost and PS translocates to the external leaflet. Fluorochrome-labeled Annexin V can then be used to specifically target and identify apoptotic cells. Annexin V Binding Buffer (Cat. No. 422201) is recommended for use with Annexin V staining. Annexin V binding alone cannot differentiate between apoptotic cells and necrotic. Therefore, we recommend using our Helix NP™ Blue (Cat No. 425305), Helix NP™ Green (Cat No. 425303) or Helix NP™ NIR (Cat. No. 425301). Early apoptotic cells will exclude 7-AAD and PI, while late stage apoptotic cells and necrotic cells will stain positively, due to the passage of these dyes into the nucleus where they bind to DNA.

Product Details

Verified Reactivity

All mammalian species

Formulation

Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide and BSA (origin USA)

Preparation

The purified protein was conjugated with APC under optimal conditions.

Concentration

Lot-specific (to obtain lot-specific concentration and expiration, please enter the lot number in our <u>Certificate of Analysis</u> online tool.)

Storage & Handling

The Annexin V solution should be stored undiluted between 2°C and 8°C, and protected from prolonged exposure to light. **Do not freeze**.

Application

FC - Quality tested

Recommended Usage

Each lot of this product is quality control tested by immunofluorescent staining with flow cytometric analysis. For flow cytometric staining, the suggested use of this reagent is 5 μ l per 100,000 - million cells in a 100 μ l volume of Annexin V Binding Buffer (Cat No. 422201). It is recommended that the reagent be titrated for optimal performance for each application.

Excitation Laser

Red Laser (633 nm)

Application Notes

Annexin V Staining

- 1. Wash cells twice with cold BioLegend Cell Staining Buffer (Cat. No. 420201) and then resuspend cells in Annexin V Binding Buffer (Cat. No. 422201) at a concentration of 1x10⁶ cells/ml.
- 2. Transfer 100 µl of cell suspension in 5 ml test tube.
- 3. Add 5 µl of APC Annexin V.
- 4. Add 10 µl of Pl solution (Cat. No. 421301) or 7-AAD (Cat. No. 420403/420404).
- 5. Gently vortex the cells, and incubate for 15 min at room temperature (25°C), in the dark.
- 6. Add 400 µl of Annexin V Binding Buffer (Cat. No. 422201) to each tube. Analyze by flow cytometry.

For a better experience detecting apoptosis, we now recommend <u>Apotracker™</u>. Cell staining with Apotracker™ is Calcium independent. Thus, no special buffers are required, and the protocol can be shortened for single-step co-staining with other reagents

Application References

(PubMed link indicates BioLegend citation)

- 1. Koopman G, et al. 1994. Blood 84:1415.
- 2. Vermes I, et al. 1995. J. Immunol. Methods 184:39.
- 3. Dachary-Prigent J, et al. 1993. Blood 81:2554.
- 4. Sekine C, et al. 2009. Int Immunol. PubMed
- 5. Grujic M, et al. 2010. J. Immunol. 185:1730. PubMed
- 6. Hussain MS, et al. 2013. Hum Mol Genet. 22:5199. PubMed
- 7. Feng Q, et al. 2014. PLoS One. 9:95927. PubMed
- 8. Isobe T, et al. 2014. eLife. 3:1977. PubMed

Product Citations

- 1. Tang Z, et al. 2017. Cell Death Differ. 24:2127. PubMed
- 2. Patel RP, et al. 2020. Transl Oncol. 14:100917. PubMed
- 3. Preda MB, *et al.* 2021. Cell Death Dis. 12:566. <u>PubMed</u>
- 4. Chugh RM, et al. 2021. Stem Cell Res Ther. 12:388. PubMed
- 5. Inclan-Rico JM, et al. 2020. Nat Immunol. 21:1181. PubMed
- 6. McCartin C, et al. 2022. Cancers (Basel). 14:. PubMed
- 7. Isobe T, et al. 2014. Elife. 3:1977. PubMed
- 8. Wu N, et al. 2020. Cell Rep. 30:1129. PubMed
- 9. Wiley CD, et al. 2019. Cell Rep. 28:3329. PubMed
- 10. Kuljanin M, *et al.* 2018. Cell Rep. 25:2524. <u>PubMed</u>

11. Wu SJ, et al. 2017. Cell Rep. 2.628472222. PubMed

12. Schick M, et al. 2022. Nat Commun. 13:281. PubMed

not an antibody (BioLegend Cat. No. 640919) AB_2561515 (BioLegend Cat. No. 640920) AB_2616657 (BioLegend Cat. No. 640941)

Antigen Details

Biology Area

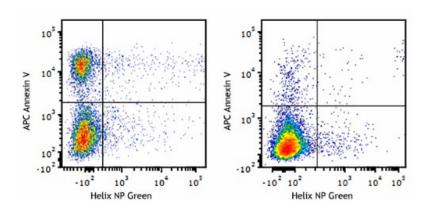
Apoptosis/Tumor Suppressors/Cell Death, Cell Biology, Neuroscience

Gene ID

RRID

308

Product Data



Human T leukemia cell line Jurkat, treated (left) or non-treated (right) with BioLegend's antihuman CD95 (EOS9.1) mAb (Cat. No. 305704) for 4 hours, then stained with Annexin V- APC and Helix NP Green (Cat. No. 425303 at 5 nM) in Annexin V Binding buffer for 15 minutes at 25°C.

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