



PE Annexin V

Catalog# / Size 640907 / 25 tests 640908 / 100 tests

640947 / 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. # 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 PE under optimal conditions.

Concentration

Lot-specific (to obtain lot-specific concentration and expiration, please enter the lot number in our Certificate of Analysis 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 µl per 100,000 - million cells in a 100 µl 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

Blue Laser (488 nm)

Green Laser (532 nm)/Yellow-Green Laser (561 nm)

Application Notes

- 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 PE Annexin V.
- 4. Add 10 µL of PI 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 Apotracker™. 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.
- 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

Product Citations

- 1. Roblek M, et al. 2022. Front Oncol. 12:777634. PubMed
- Desikan SA, et al. 2022. STAR Protoc. 3:101388. PubMed
- 3. Symeonidou V, et al. 2021. Cell Rep. 37:109900. PubMed
- 4. Chiu Y, et al. 2017. Nat Commun. 8:14324. PubMed
- Kulkarni PP, et al. 2021. Haematologica. . <u>PubMed</u>
 Carriere PP, et al. 2018. Int J Oncol. 53:1442. <u>PubMed</u>
- 7. Liu Y, et al. 2022. Cancer Discov. :. PubMed
- Hinterbrandner M, et al. 2021. JCI Insight. 6:e151797. PubMed
- 9. Liu J, et al. 2021. Sci Adv. 7: . PubMed
- 10. Herman KD, et al. 2022. Front Immunol. 13:956991. PubMed
- 11. Bartleson JM, et al. 2020. Nat Immunol. 1384:21. PubMed
- 12. Fuhrmann F et al. 2016. eLife. 5 pii: e20616. PubMed

not an antibody (BioLegend Cat. No. 640907) AB_2561298 (BioLegend Cat. No. 640908) not an antibody (BioLegend Cat. No. 640947)

Antigen Details

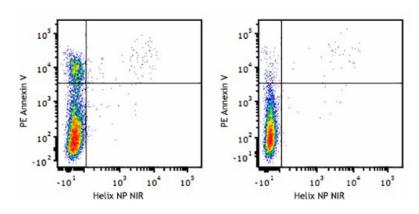
Biology Area

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

Gene ID

<u>308</u>

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 at 37°C, then stained with Annexin V- PE for 15 minutes at 37°C in Annexin V Binding buffer. Helix NP Green (Cat. No. 425303 at 1.25 nM) added 5 minutes at room temperature prior to running tubes.

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8999 BioLegend Way, San Diego, CA 92121 www.biolegend.com Toll-Free Phone: 1-877-Bio-Legend (246-5343) Phone: (858) 768-5800 Fax: (877) 455-9587