

## Brilliant Violet 421™ Annexin V

<b>Catalog# / Size</b>	640923 / 25 tests 640924 / 100 tests
<b>Regulatory Status</b>	RUO
<b>Other Names</b>	Annexin A5
<b>Description</b>	<p>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.</p>

### Product Details

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<b>Verified Reactivity</b>	All mammalian species
<b>Formulation</b>	Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide and BSA (origin USA).
<b>Preparation</b>	The protein was purified by affinity chromatography and conjugated with Brilliant Violet 421™ under optimal conditions.
<b>Concentration</b>	Lot-specific (to obtain lot-specific concentration, please enter the lot number in our <a href="#">Concentration and Expiration Lookup</a> or <a href="#">Certificate of Analysis</a> online tools.)
<b>Storage &amp; Handling</b>	The solution should be stored undiluted between 2°C and 8°C, and protected from prolonged exposure to light. <b>Do not freeze.</b>
<b>Application</b>	<a href="#">FC - Quality tested</a>
<b>Recommended Usage</b>	<p>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.</p> <p>Brilliant Violet 421™ excites at 405 nm and emits at 421 nm. The standard bandpass filter 450/50 nm is recommended for detection. Brilliant Violet 421™ is a trademark of Sirigen Group Ltd.</p> <p><a href="#">Learn more about Brilliant Violet™.</a></p> <p>This product is subject to proprietary rights of Sirigen Inc. and is made and sold under license from Sirigen Inc. The purchase of this product conveys to the buyer a non-transferable right to use the purchased product for research purposes only. This product may not be resold or incorporated in any manner into another product for resale. Any use for therapeutics or diagnostics is strictly prohibited. This product is covered by U.S. Patent(s), pending patent applications and foreign equivalents.</p>
<b>Excitation Laser</b>	Violet Laser (405 nm)
<b>Application Notes</b>	<p><b>Annexin V Staining</b></p> <ol style="list-style-type: none"><li>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 1x10<sup>6</sup> cells/mL</li><li>2. Transfer 100 µL of cell suspension in 5 mL test tube.</li><li>3. Add 5 µL of Brilliant Violet™ 421 Annexin V.</li><li>4. Add 10 µL of PI solution (Cat. No. 421301) or 7-AAD (Cat. No. 420403/420404).</li><li>5. Gently vortex the cells, and incubate for 15 min at room temperature (25°C), in the dark.</li><li>6. Add 400 µL of Annexin V Binding Buffer (Cat. No. 422201) to each tube. Analyze by flow cytometry.</li></ol>

**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.

#### Additional Product Notes

View more applications data for this product in our [Scientific Poster Library](#).

#### 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](#)

#### Product Citations

1. Uroda T, et al. 2019. *Mol Cell.* 75:982. [PubMed](#)
2. Bauché D et al. 2018. *Immunity.* 49(2):342-352. [PubMed](#)
3. Nkwouano V, et al. 2017. *PLoS One.* 12(2):e0171817. [PubMed](#)
4. Murakami K, et al. 2021. *Cell Reports.* 34(1):108579. [PubMed](#)
5. Ward D, et al. 2016. *Haematologica.* 101: 286 - 296. [PubMed](#)
6. Di Franco S, et al. 2021. *STAR Protoc.* 2:100880. [PubMed](#)

#### RRID

not an antibody (BioLegend Cat. No. 640923)  
AB\_2686928 (BioLegend Cat. No. 640924)

## Antigen Details

#### Biology Area

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

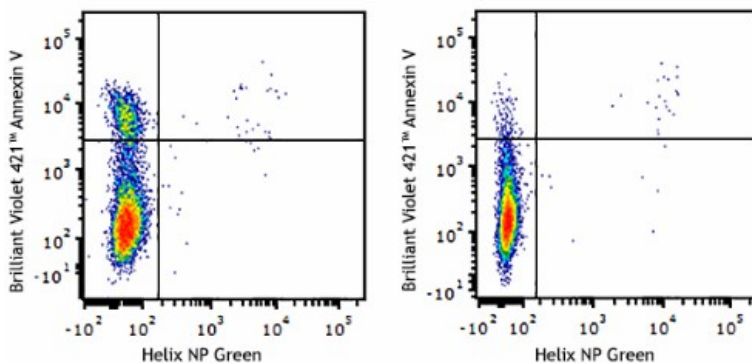
#### Gene ID

[308](#)

## Related Protocols

[Cell Surface Flow Cytometry Staining Protocol](#)

## Product Data



Human T leukemia cell line Jurkat, treated (left) or non-treated (right) with BioLegend's anti-human CD95 (EOS9.1) mAb (Cat. No. 305704) for 4 hours at 37°C, then stained with Annexin V-Brilliant Violet 421™ for 15 minutes at 37°C in Annexin V Binding buffer. Helix NP Green (Cat. No. 425303 at 1.25 nM) was added 5 minutes at room temperature prior to running tubes.

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