

## Pacific Blue™ Annexin V Apoptosis Detection Kit with 7-AAD

<b>Catalog# / Size</b>	640926 / 100 tests
<b>Regulatory Status</b>	RUO
<b>Other Names</b>	Annexin A5 Apoptosis Detection Kit
<b>Description</b>	BioLegend's Pacific Blue™ Annexin V Apoptosis Detection Kit with 7-AAD has been specifically designed for the identification of apoptotic and necrotic cells.

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 is recommended for use with Annexin V staining. Annexin V binding alone cannot differentiate between apoptotic and necrotic cells. To help distinguish between the necrotic and apoptotic cells we recommend use of our 7-amino-actinomycin D (7-AAD) solution. Early apoptotic cells will exclude 7-AAD, while late stage apoptotic cells will stain positively, due to the passage of these dyes into the nucleus where they bind to DNA.

7-AAD (7-amino-actinomycin D) has a high DNA binding constant and is efficiently excluded by intact cells. It is useful for DNA analysis and dead cell discrimination during flow cytometric analysis. When excited by 488 laser light, 7-AAD fluorescence is detected in the far red range of the spectrum (650 nm long-pass filter).

### Product Details

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<b>Verified Reactivity</b>	All mammalian species
<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>	Store between 2°C and 8°C. <b>Do not freeze.</b>  <b>Caution:</b> 7-AAD is a potential carcinogen. It is recommended that the user wear protective clothing, gloves, and eye/face protection in order to avoid contact with skin and eyes.
<b>Application</b>	<a href="#">FC - Quality tested</a>
<b>Recommended Usage</b>	<b>Staining Procedure:</b> <ol style="list-style-type: none"><li>1. Wash cells twice with cold BioLegend's Cell Staining Buffer, and then resuspend cells in Annexin V Binding Buffer at a concentration of 0.25-1.0 x 10<sup>7</sup> cells/mL.</li><li>2. Transfer 100 µL of cell suspension in a 5 mL test tube.</li><li>3. Add 5 µL of Pacific Blue™ Annexin V.</li><li>4. Add 5 µL of 7-AAD Viability Staining Solution.</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 to each tube. Analyze by flow cytometry with proper machine settings.</li></ol>
<b>Application Notes</b>	<b>Materials Provided:</b> 0.5 ml of Pacific Blue™ Annexin V 0.5 ml of 7-AAD Viability Staining Solution 50 ml of Annexin V Binding Buffer  <b>Materials Not Included:</b> Cell Staining Buffer (Cat. No. 420201)  <b>For a better experience detecting apoptosis, we now recommend <a href="#">Apotracker™</a>. Cell staining with <a href="#">Apotracker™</a> is Calcium independent. Thus, no special buffers are required, and the protocol can be shortened for single-step co-staining with other reagents.</b>
<b>Application References</b>	1. Maciel E, <i>et al.</i> 2014. <i>Arch Biochem Biophys.</i> 548:38. <a href="#">PubMed</a>
<b>(PubMed link indicates</b>	

## BioLegend citation)

### Product Citations

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7. Zang RX, *et al.* 2022. ACS Omega. 7:34378. [PubMed](#)
8. van Stevendaal MHME, *et al.* 2021. ACS Appl Mater Interfaces. 13:7879. [PubMed](#)
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12. Calamita P, *et al.* 2017. PLoS Genet. 10.1371/journal.pgen.1006552. [PubMed](#)

## Antigen Details

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### Biology Area

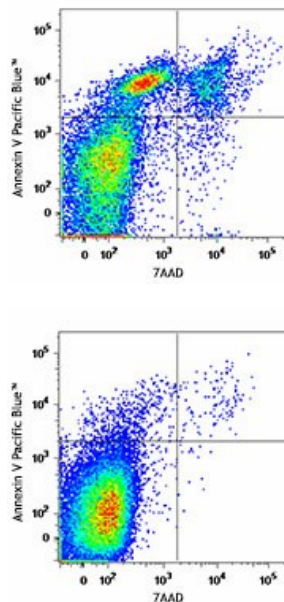
Apoptosis/Tumor Suppressors/Cell Death, Cell Biology, Cell Proliferation and Viability, Neuroscience

### Gene ID

[308](#)

## Product Data

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Human T-cell leukemia cell line, Jurkat, treated (top) or non-treated (bottom) with LEAF™ purified anti-human CD95 (clone EOS9.1) for 4 hours, then stained with Pacific Blue™ Annexin V Apoptosis Detection Kit with 7-AAD.

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