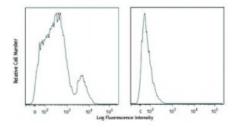
## SONY

## **Product Data Sheet**

## **Purified Annexin V**

Catalog # / Size:	3804505 / 100 μg 3804510 / 500 μg
Isotype:	
Reactivity:	Human,Mouse,Non-human primate,Other,Rat
Preparation:	The protein was purified by affinity chromatography.
Formulation:	Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide.
Concentration:	0.5



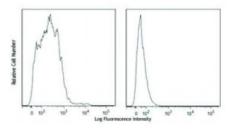
Human T cell leukemia cell line, Jurkat, was treated (4 hours) with (top 2 panels) or without (bottom two panels) LEAF<sup>™</sup> purified anti-CD95 (clone EOS9.1), then stained with Annexin V FITC (panels 1 & 3) or with purified recombinant Annexin V to bloc

## **Applications:**

Applications:	Flow Cytometry
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**Recommended** Usage: Each lot of this product is quality control tested by immunofluorescent staining with flow cytometric analysis. For flow cytometric application, the suggested use of this reagent is 5 - 15 microg per 100,000 - million cells in a 100 microL volume of Annexin V Binding Buffer (Cat No. 422201). It is recommended that the reagent be titrated for optimal performance for each application.

Application **Annexin V Staining** 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  $1 \times 10^6$  cells/ml. 2. Transfer 100 microL of cell suspension in 5 ml test tube. 3. Add 5 microL of purified Annexin V. 4. Add 10 microL 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 microL of Annexin V Binding Buffer (Cat. No. 422201) to each tube. Analyze by flow cytometry.



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Application	1. Koopman G, et al. 1994. <i>Blood</i> 84:1415.
<b>References:</b>	2. Vermes I, et al. 1995. J. Immunol. Methods 184:39.
	3. Dachary-Prigent J, et al. 1993. <i>Blood</i> 81:2554.
	4. Sekine C, <i>et al.</i> 2009. <i>Int Immunol.</i> <u>PubMed</u>
	5. Grujic M, <i>et al.</i> 2010. <i>J. Immunol.</i> 185:1730. <u>PubMed</u>

**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 and necrotic cells. Therefore, we recommend using our 7-AAD Viability Staining Solution (Cat. No. 420403/420404) or Propidium Iodide Solution (Cat. No. 421301). 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.