

## **Product Data Sheet**

## Pacific Blue™ Annexin V

Catalog # / Size: 640917 / 25 tests

640918 / 100 tests

Reactivity: All mammalian species

Preparation: The purified protein was conjugated with Pacific Blue™ under optimal

conditions. The solution is free of unconjugated Pacific Blue™.

Formulation: Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide and

0.2% (w/v) BSA (origin USA).

Storage: The Annexin V solution should be stored undiluted at 4°C, and protected from

prolonged exposure to light. Do not freeze.

## **Applications:**

Applications: FC - Quality tested

Recommended Usage: Each lot of this product is quality control tested by staining of apoptotic cells and analyzing by flow cytometry. The suggested use of this reagent is  $5 \mu l$ 

per 1X10<sup>5</sup> 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. \* Pacific Blue™ has a maximum emission of 455 nm when it is excited at 405 nm. Prior to using Pacific Blue™

conjugate for flow cytometric analysis, please verify your flow cytometer's

capability of exciting and detecting the fluorochrome.

\*\*\* Pacific Blue™ is a registered trademark of Molecular Probes, Inc. Pacific Blue™ dye antibody conjugates are sold under license from Molecular

Probes, Inc. for research use only, except for use in combination with microarrays and high content screening, and are covered by pending and issued patents.

Application Notes: Annexin V Staining

1. Wash cells twice with cold BioLegend cell staining buffer (cat # 420201) and then resuspend cells in Annexin V Binding Buffer (cat # 422201) at a

Add 10 µl of PI solution (cat # 421301) or 7-AAD (cat # 420401).

5. Gently vortex the cells and incubate for 15 min at RT (25°C) in the dark.

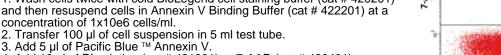
6. Add 400 µl of Annexin V Binding Buffer (cat # 422201) to each tube.

Analyze by flow cytometry.

Application References: 1. Gordy C, et al. 2011 Blood 117:618. PubMed

2. Jia W, et al. 2011. J. Immunol. 186:5313. PubMed.

3. Naegele M, et al. 2012. J. Neuroimmunol. 242:60. PubMed. 4. Jenke AC, et al. 2013. PLoS One. 8:e55636. PubMed.



Annexin V Pacific Blue<sup>1</sup>

Annexin V Pacific Blue™

Human T-cell leukemia cell line, Jurkat, stimulated (4 hours) with

(bottom) or without (top) LÉAF™

purified anti-CD95, clone EOS9.1, then stained with Annexin V Pacific Blue™ and viability dye 7-AAD

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. So, we recommend using our 7-AAD Viability Staining Solution (cat. no. 420401/420402) 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.

Related Products: Product

Annexin V Binding Buffer 7-AAD Viability Staining Solution Propidium Iodide Solution

Clone

**Application** 



