

Annexin V Apoptosis Detection Kit

50/100 Reactions

Kit storage:

Store between 2°C and 8°C. Do not freeze

Verified Reactivity: Human, Mouse, Rat

Component for 50 Rxn

Annexin V-FITC	300	μl
Annexin V Binding Buffer	2X60	ml
Propidium iodide	300	μl

Description: Parstous's FITC Annexin V Apoptosis Detection Kit with PI identifies apoptotic and necrotic cells. Annexin V binds to phosphatidylserine (PS), which is exposed on the cell surface during early apoptosis. Annexin V alone cannot distinguish between apoptotic and necrotic cells, so Propidium iodide (PI) is added. Early apoptotic cells exclude PI, while late apoptotic and necrotic cells take it up, allowing differentiation. PI binds DNA and is detected in the PE/Texas Red® channel, making it useful for cell viability and cell cycle analysis via flow cytometry.

General Protocol (Annexin V Staining):

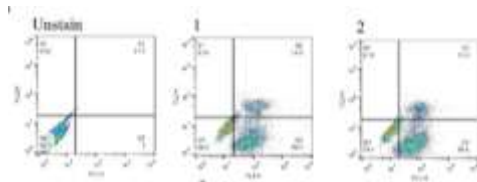
- Wash cells twice with cold PBS and then resuspend cells in 1 ml Binding Buffer at a concentration of $\sim 5 \times 10^5$ cells/ml.
- Centrifuge at 300 x g for 5 minutes at room temperature (15 - 25°C). Remove and discard supernatant.
- Resuspend cells at 0.5 ml of Annexin V Binding Buffer.
- Aliquot 100 μL of cell suspension to individual tubes for staining.
- Add 2-5 μl of Annexin V- FITC.

Note: The amount of purified recombinant Annexin V required to saturate binding sites may vary according to cell type, and stage of apoptosis. In some cases, investigators may also need to reduce the number of cells to 0.5×10^5 /200 μl and still add 2-5 μg of recombinant Annexin V to obtain optimal results.

- Gently mix the cells and incubate for 15 min at RT.
- Add 5 μl of propidium iodide (PI) (optional)
- OPTIONAL: To reduce background staining, wash cells with 0.5 mL of Annexin V Binding Buffer. Centrifuge sample at 300 x g for 5 minutes at room temperature. Remove and discard supernatant.
- Add 200 μL of Annexin V Binding Buffer to each tube
- Analyze by flow cytometry as soon as possible (within 1 hr).

- Analyze annexin V-FITC binding via flow cytometry. Ex = 488nm and Em = 350nm using FITC signal detector (usually FL1)
- If propidium iodide was added, analyze PI staining by the phycoerythrin emission signal detector (usually FL2).

Notes: Caution: Propidium Iodide Solution is toxicogenetic and mutagenic; handle with care.



Disclaimers and Addresses

This product is for research use only and should only be used by trained professionals.

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