Annexin V-FITC Apoptosis Analysis Kit

Catalog Number	Vial Size
AO2001-02P-G	25 tests
AO2001-02P-H	100 tests



Important Note: Each Buffer should be diluted by using the same pH PBS as mentioned below. This product is guaranteed up to one year from purchase.

Description

AAnnexin 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.

Products List

AO2001-02	Annexin V-FITC	Keep as concentrated solution. Store at 4°C and protected from prolonged exposure to light. Do not freeze.
AO2002	Propidium lodide Solution	Keep as concentrated solution. Store at 4°C and protected from prolonged exposure to light. Do not freeze.
AB2000	Annexin V Binding Buffer	Keep as concentrated solution. Store at 4°C as an undiluted liquid. For extended storage aliquot contents and freeze at -20°C.
AO2004	Apoptosis Positive Control Solution	Keep as concentrated solution. Store at 4°C and protected from prolonged exposure to light. Do not freeze.

Suggested Staining Protocol A.Parameters regulation

1. Harvest cell(1×10^6 - 3×10^6 cells), then sepatate the cells in two parts. Wash cells with cold PBS, then centrifuge the cells and disgard the supernatant.

2. Suspend one part of cells in 200µL 1× binding buffer, store at 4 $^\circ\!C$ for use.

3. Suspend the other part of cells in 500µL Apoptosis Positive Control Solution, and incubate for 10 minutes in room temperature. Wash cells with more than 3.0 mL cold PBS, blot the supernatant, then suspend the cells in 200µL 1× binding buffer.

4. Mix the two parts cells together, then separate the cells in three tubes, and add 100 μ L of cells in each tube.

5. The first tube is Blank Control, the second one adds

5 μL of Annexin V-FITC, and the third one adds 5 μL PI solution.

6. Gently vortex each tube and incubate for 5 minutes in room temperature, protected from light.

7. Before analyzing by flow cytometry, using the blank control and single dye sample to regulate voltage and compensation, as shown in Figure Parameters regulation.

B.Sample detection

1. Dilute 3 mL 10× binding buffer with 27 mL distilled water for 10 tests.

2. Harvest cell[®]about 1×10^5 cells per test[®]then wash with cold PBS.

3. Suspend cells in 1 mL 1× Binding Buffer, 300×g centrifugation for 10 minutes, then remove the Binding Buffer from the cell pellet.

4. Resuspend cells in 1 mL 1× Binding Buffer , adjust cell concentration to 1×10^6 cells/mL.

5. Add 100 μL of cells (1×10 $^{\rm 5}$ cells) to each labeled tube.

6. Add 5 μL of Annexin V-FITC to appropriate tubes.

7. Gently vortex each tube and incubate for 10 minutes in room temperature, protected from light.

8. Add 5 μ L PI solution incubation for 5min in room temperature, protected from light.

9. Add PBS to 500µL and vortex gently.

10. Analyze by flow cytometry in 1 hour.

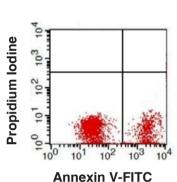
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	天津三箭生物技术股份有限公司 Tianjin Sungene Biotech Co., Ltd. 称准 高效 穆定 Precision Efficient Stable
Market	400-621-0003 marketing@sungenebiotech.com
Support	022-66211636-8024 techsupport@sungenebiotech.com
Web	www.sungenebiotech.com

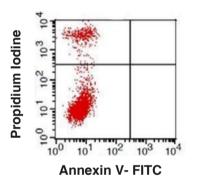
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Illustration of Immunofluorescent Staining



Parameters regulation

Annexin V- FITC Single Staining Control



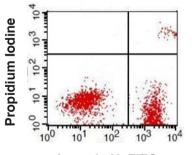
Propidium Iodine Single Staining Control

References

Fadok,V.A.et al. 1992.J.Immunol. 148:2207
Tait,J.F., et al. 1989.J.Biol.Chem.264:7944..
Andree,H.A., et al. 1990.J.Biol.Chem.265:4923.
Tiagarajan,P., et al. 1990.J.Biol.Chem.265:17420.
Dachary,P.J., et al. 1993.Blood 81:2554.
Koopman,G., et al. 1994.Blood 84:1415.
Martin,S.J., et al. 1995.J.Exp.Med.182:1545.
Wood,B.L., et al. 1996.Blood 88:1873.

For Research Use Only.

Sample detection



Annexin V- FITC Camptothecin treated Jurkat Cell stained with Annexin V- FITC and PI