Annexin V-Cy3 Apoptosis Detection Kit

Cat. No. CTG-AP0006 1 Kit (100 tests)

Procedure

1. Induce apoptosis by desired method. Concurrently incubate a control culture without induction.
2. Collect 1-5 x 10^5 cells by centrifugation.
3. Resuspend cells in 500 µl of 1X Binding Buffer.
4. Add 5 µl of Annexin V-Cy3 and 1 µl of SYTOX Green dye. Note: Thaw the SYTOX Green dye in room temperature before use.
5. Incubate at room temperature for 5-10 min in the dark.
6. Analyze the stained cells by flow cytometry using FL1 channel for SYTOX Green dye (Ex = 488 nm; Em = 530 nm) and FL2 channel for Annexin V-Cy3 (Ex = 543 nm; Em = 570 nm).

The cell population should separate into three groups: live cells with only a low level of fluorescence, apoptotic cells with red fluorescence and necrotic cells with green fluorescence. The flow cytometric results can also be confirmed by viewing the cells under a fluorescence microscope using FITC filter for SYTOX and rhodamine filter for Annexin V-Cy3.

Notes: For adherent cells, gently trypsinize and wash cells once with serum-containing media before incubation with Annexin V-Cy3 and SYTOX dye.

References

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