DNA Content Quantitation Assay(Cell Cycle)

Cat : CA1510
Size: 20T/50T
Storage: -20℃ away from light, valid for one year.
Product description:

The cell cycle refers to the entire process that continuously dividing cells undergo from the end of one mitosis to the end of the next mitosis. During this process, the cell's genetic material replicates and doubles, and is evenly divided between the two daughter cells at the end of division. The cell cycle can be divided into interphase and mitotic phase. Interphase is often divided into dormant phase (G), DNA synthesis phase (G_0), DNA synthesis phase ($_1S$), DNA synthesis phase (G), and DNA synthesis phase (G_2). The whole cycle can be expressed as $G_1 \rightarrow S \rightarrow G_2 \rightarrow M$. DNA cycle detection can be used to reflect the status of each phase of the cell cycle, that is, the cell proliferation status. By taking advantage of the ability of DNA in cells to bind to fluorescent dyes (such as propyl iodide PI), the fluorescence intensity measured by flow cytometry is also different because of the different DNA content of cells at different stages and the fluorescent dyes bound to them.

During cell apoptosis, due to the concentration of cytoplasm and chromatin, nuclear lysis, apoptotic bodies are produced, which changes the light scattering properties of cells. In the early stage of apoptosis, the ability of the cells to scatter light from the forward Angle is significantly reduced, and the ability to scatter light from the 90° Angle is increased or not changed. In the late stages of apoptosis, the signals of both forward Angle and 90° Angle light scattering were reduced. Therefore, apoptotic cells can be observed by measuring the change of light scattering by flow cytometry. The apoptotic cells were stained with PI. Due to the decrease of the total DNA content of apoptotic cells, the DNA hypochromic cell population appeared before the normal G_0/G_{11} cell population, that is, the subdiploid peak (sub-G_1) appeared before the G peak, that is, the apoptotic group.

This kit can be applied to the detection of DNA content (cell cycle) in cultured cells (suspension, adherence).

Kit composition:

Reagent name	CA1510-20T	CA1510-50T	Storage conditions
RNase A	2.0mL	5.0mL	- 20 ℃
PI stain solution	8.0mL	20.0mL	-20℃ to avoid light

Protocols: (only for reference):

1. Apoptosis was induced by appropriate methods, while a negative control group was set up, and

cells were collected.

2. The cells were washed once with PBS, 1500rpm, centrifuged for 5min, and the cell concentration was adjusted to 1×10^6/mL, and 1mL single-cell suspension was taken.

3. After centrifugation of the prepared single-cell suspension, the supernatant was removed, and 70% pre-cooled ethanol 500μL was added to the cells and fixed for 2 hours to overnight, stored at 4℃, and the fixed solution was washed off with PBS before staining; If necessary, the cell suspension can be filtered once with a 200-mesh cell screen.

4. Add 100μL RNase A solution to cell precipitation, suspend the cells, and bathe in 37℃ water for 30min.

5. Add 400μL PI staining solution and mix well. Incubate at 4℃ for 30min in dark.

6. The red fluorescence at 488nm excitation wavelength was recorded by computer detection.

Note:

1. Propyl iodide (PI) dyeing solution should be kept away from light during storage and use.

2. PI toxic, operation should wear gloves, and avoid pollution.

3. Fluorescent dyes have quenching problems, it is recommended to complete the test on the same day after dyeing.

Related products:

SF9	*Sheep Anti-Mouse IgG-FITC*
S9000	*Extra Grade Fetal Bovine Serum*
12100	*DMEM(H) culture-medium*
S2100	*anti-fluorescence attenuation sealer*
C0080	*Propyl iodide PI Solution (1mg/mL)*
CA1020	*ANNEXIN V-FITC/PI Apoptosis detection Kit*