Cell Apoptosis DAPI Detection Kit **Cat. No. L00312**

Technical Manual No. 0358 **Version 11172008**

I. INTRODUCTION

Cell Apoptosis DAPI Detection Kit (Cat.No.L00312) provides a rapid and convenient assay for apoptosis based upon fluorescent detection. 4, 6-Diamidino-2-phenylindole (DAPI) is a kind of specific dye for binding DNA. This dye is not completely permeability. Once it overpasses cell membranes of normal cells, the blue fluorescence will be observed by fluorescent microscopy. With the process of apoptosis, the ability of permeability for dye is improved and the apoptotic cells will produce high blue fluorescence. At the same time, for normal cells, round nucleus is stained uniformity and its margin is clear. But, for apoptotic cells, the margin of nucleus is abnormity and the condensed chromosome is easily stained. So it can be observed apoptotic cells according to the strength of fluorescence and the conformation of nucleus by using this kit.

II. KIT CONTENTS

Cell Apoptosis DAPI Detection Kit (Cat.No.L00312) employs DAPI assaying normal and apoptotic cells. Each kit contains enough reagents for one hundred apoptosis assays.

Kit Components	100 Assays
DAPI	10 ml
Buffer A	25 ml

III. KEY FEATURES

- Easy to perform: simple and rapid procedure to perform.
- Fast and quick: all of the procedures are less than 20 minutes.
- Versatile: directly analyze normal and apoptotic cells by fluorescence microscopy.
- Ready to use
- Highly competitive price

- A visual presentation of apoptosis

IV. STORAGE

This kit remains stable for at least six months if stored at 4°C and protected from light.

V. CELL APOPTOSIS DAPI DETECTION KIT PROTOCOL

Note:

1. Before the experiment, dilute DAPI to 2 µg/ml work buffer by adding 90 ml of methanol.
2. Protect DAPI reagent from light all the time. And DAPI is a known mutagen and should be handled with care. It is necessary to use with appropriate precautions.

For Adherence Cells

1. Discard the cell media on the cover slip for adherence cells. And add 500 µl of DAPI work reagent to wash once. Then, discard the DAPI work reagent.
2. Add 500 µl of DAPI work weagent and incubate at 37°C for 15 minutes.
3. Discard the work reagent and potch cells once using methanol.
4. Place glycerol or buffer A on cells.
5. Observe at 340/380 nm of excitation wavelength by fluorescent photometer.

For Suspension Cells

1. Harvest the cells by centrifugation at 2000 rpm for five minutes.
2. Wash cells by adding 500 µl of DAPI work reagent.
3. Add 500 µl of DAPI work reagent and suspend cells. Incubate the cells at 37°C for 15 minutes.
4. Centrifuge cells and discard the supernatant.
5. Add buffer A to suspend cells and place the suspension cells on a glass slipe.
6. Observe at 340/380 nm of excitation wavelength by fluorescent photometer.

VI. RELATED PRODUCTS

Cell Cycle Analysis Kit:	Cat.No.L00287.
Annexin V-EGFP Apoptosis Detection Kit:	Cat.No.L00288.
Caspase-3 Colorimetric Assay Kit:	Cat.No.L00289.
Double Stain Apoptosis Detection Kit (Hoechst 33342/PI):	Cat.No.L00309.
Cell Apoptosis PI Detection Kit:	Cat.No.L00311.

VII. EXAMPLES

P388 cells were induced apoptosis by 10 μM camptothecin for one hour at 37°C, and procedures were accomplished according with protocol as described as above. The result is as follow:

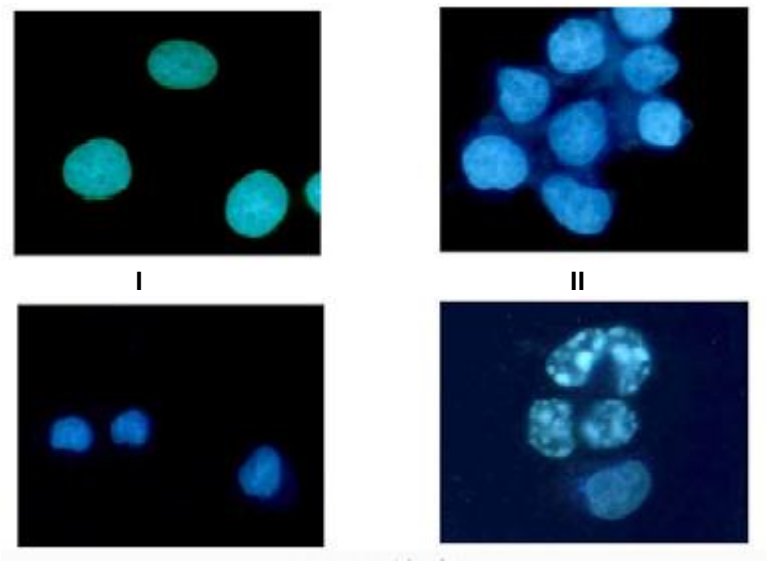

Fig.1 The morphological change of nuclear chromatin in apoptosis observed by fluorescent microscopy
I: Control; II: Stage I; III: Stage IIa; IV: Stage IIb

VIII. ORDERING INFORMATION

Cell Apoptosis DAPI Detection Kit: Cat.No. L00312

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