



## CellCycle™ PI/RNase Staining Solution

Catalog Number: A056

Table 1. Product Package and Storage

Material	Amount	Concentration	Storage	Stability
CellCycle™ PI/RNase Stain	50 mL	1X	4 °C	One year when stored as directed.

**Number of assays:** 100 assays.

**Approximate fluorescence excitation/emission maxima, in nm:** 535/617, bound to DNA.

### Introduction

Measurement of DNA content allows the study of cell populations in various phases of the cell cycle as well as the analysis of DNA ploidy. In a given population, cells are distributed among three major phases of cell cycle: G<sub>0</sub>/G<sub>1</sub> phase (one set of paired chromosomes per cell), S phase (DNA synthesis with variable amount of DNA), and G<sub>2</sub>/M phase (two sets of paired chromosomes per cell, prior to cell division). DNA content can be measured using fluorescent, DNA-selective stains that exhibit emission signals proportional to DNA mass. Flow cytometric analysis of these stained populations is then used to produce a frequency histogram that reveals the various cell cycle phases. Univariate DNA content analysis is an established assay method and is widely used for studies in oncology, cell biology, and molecular biology.

The CellCycle™ PI/RNase staining solution is a specially formulated ready-to-use reagent. Just adding it to fixed cells, incubate, and acquire on a flow cytometer without washing.

### Materials required but not provided

- Reagents for fixing cells, such as alcohol or formaldehyde
- Wash buffer, such as PBS

### Experimental Protocols

**Note:** The following staining protocol was optimized using Jurkat cells, a human T-cell leukemia cell line, but can be adapted for any cell type.

1. Harvest the cells.
2. Fixed cells according to your preferred protocol.
3. Wash the cells.
4. Prepare flow cytometry samples each containing  $\sim 1 \times 10^6$  cells in suspension.
5. Centrifuge the samples and decant the supernatant, leaving a pellet of cells in each sample tube.
6. Add 0.5 mL of CellCycle™ PI/RNase staining solution to each flow cytometry sample, mix well.
7. Incubate at 37°C for 30 minutes, protected from light.
8. Analyze the samples without washing, using 488 nm or 532 nm excitation, and collect emission using a 585/42 bandpass filter or equivalent.