A new standard of Automated Cell Counter

ADAM MC2

Application Note

NanoEnTek

Counting of Mammalian Cells using ADAM MC2

General Description

The ADAM-MC2, an automated cell counter, is a benchtop auto-mated cell counter that performs cell count and viability measurements using AccuStain Solution.

Principle

The ADAM-MC2 is based on staining mammalian cell DNA with a fluorescent dye, Propidium Iodide (PI). PI does not enter cells with intact membranes or active metabolism. In contrast, cells with damaged membranes or with inactive metabolism are unable to prevent PI entering the cell. As a result, the nuclei of non-viable cells will only be stained. The ADAM-MC2 provides two kinds of staining solutions.

The AccuStain Solution T for the total cell counting is composed of the fluorescent dye (PI) and lysis solution. The AccuStain Solution N for the non-viable cell counting is composed of the fluorescent dye and PBS. In order to measure the total concentration of cells, the plasma membranes of all the cells must be disrupted to stain all the Nuclei with PI. The process of disrupting and staining is achieved by treatment with the AccuStain Solution T.

In the second solution, live cells remain intact and are not stained. Only the non-viable cells are stained and detected. After treatment, the prepared cells will be loaded into the chip. The viability will be automatically calculated in the ADAM-MC2 software after each measurement of the total cells and the non-viable cells.

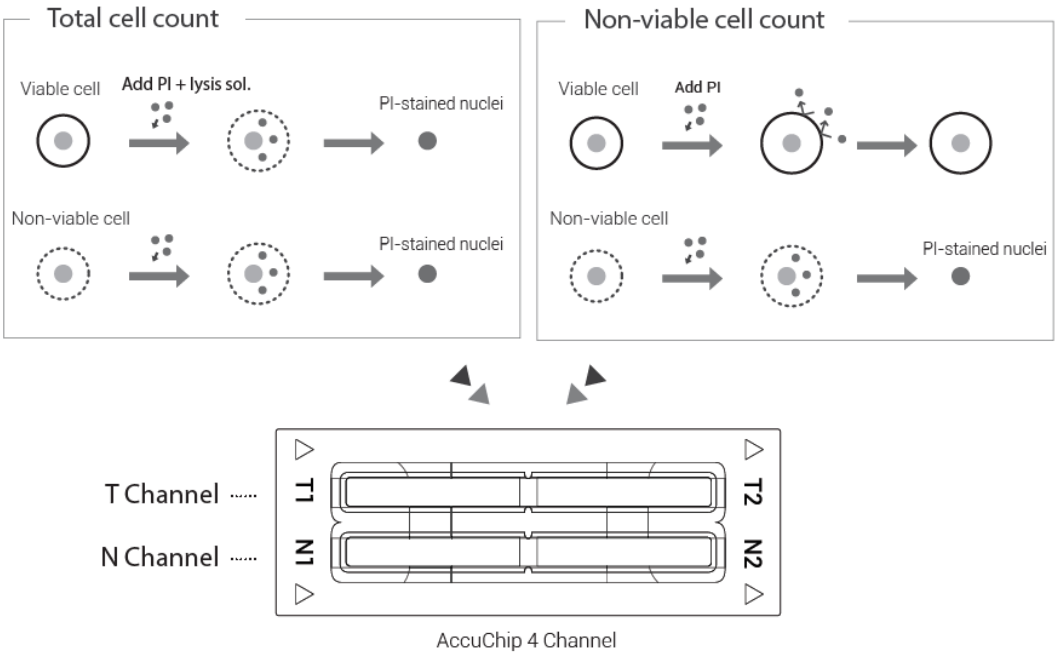

Figure 1 The principle of AccuStain solution

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Materials and Methods

- Cells to be counted
- AccuStain solution T/N
- AccuChip 4X

Sample preparation

1. Cultivate the required number of cells.
2. Add an appropriate volume of growth media or PBS to dilute to a final concentration of 1x106 cells/mL.
3. Thoroughly mix the cell pellet by vortexing.
4. Check visually if any cell clumps or agglomerates are remained.

Counting cell – Total cell

1. Add 50 µL of your sample to 50 µL supplied AccuStain Solution T.
2. Voltex the tube vigorously.
3. Load 13 µL sample mixture to the AccuChip on T1 or T2 channel. Then, wait 1 minute for the sample settling.

Counting cell – Non-viable cell

1. Add 50 µL of your sample to 50 µL supplied AccuStain Solution N.
2. Voltex the tube vigorously.
3. Load 13 µL sample mixture to the AccuChip on T1 or T2 channel. Then, wait 1 minute for the sample settling.

* When you load of the sample mixture to the AccuChip, please be careful not to make bubbles.

Result

The ADAM-MC2 software automatically calculates total cell count and viability.
The ADAM-MC2 instrument acquires a image and data for each channel.
The images and data can be easily saved to USB and send the data through the e-mail for additional analysis or data archiving.

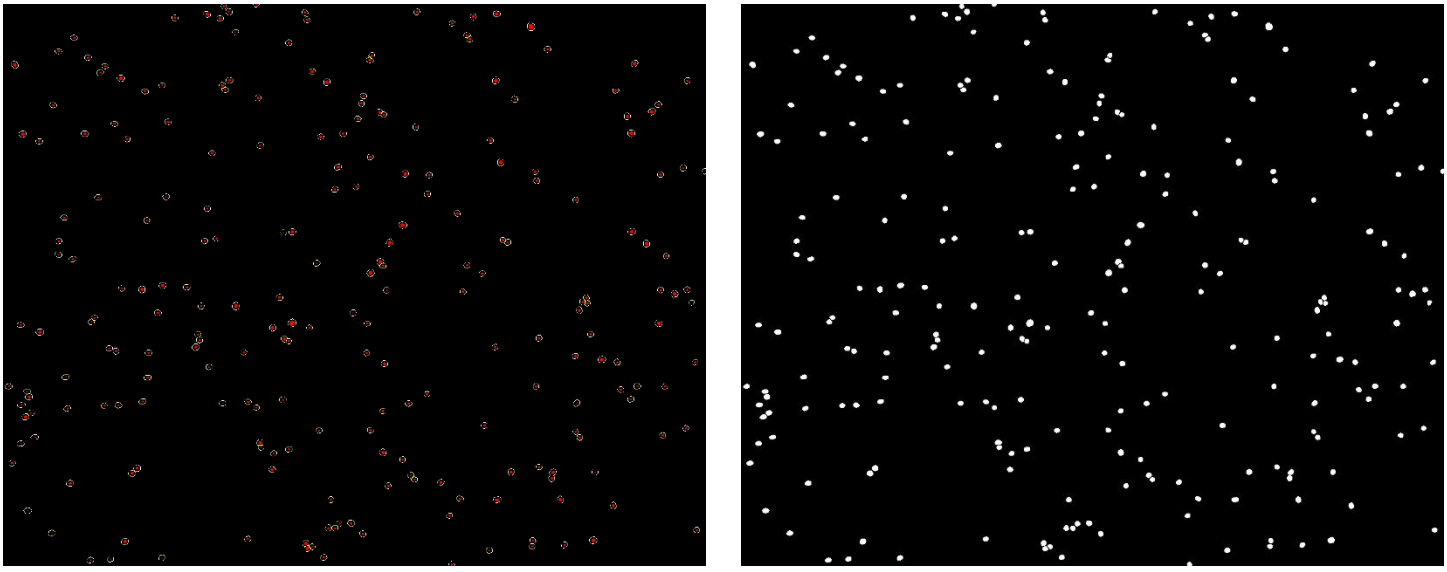

Figure 2 Mark and mask image of HeLa cell

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Trouble shooting

Inaccurate result

- Cell number may be out range.
  - Adjust the number of cells between 5X104~ 4X106cells/mL.
  - Optimal range is 5X105~2X106cells/mL.
- AccuStain Solution has expired.
  - Discard AccuStain that have expired. Purchase the AccuStain.
- Too high clumped cells.
  - Try again after vortexing the cells.

When error message is shown

- When frames with errors are over 50% of total counting frame. (Error message: E)
  - Check the suspension of cells if all cells are fully dissociated into single cells.
  - If contaminants except cells are found, prepare sample again.
- High concentration of cells (Error message: H) & Over detection range (Error message: O)
  - Check if concentration of cell is too high.
  - Dilute the sample and count again.
- Low concentration of cells (Error message: L) & Under detection range (Error message: U)
  - Check if concentration of cell is too low.
  - Use concentrated sample and count again.

Figure 3 Data edit tap - change the setting