

Viahance™: Dead Cell, Stripped Nuclei and Free Oligonucleotide Removal Kit Instructions

Viahance™ dead cell removal kit enhances the viability ratio of live to dead cells in cell culture through removal of dead cells and cellular debris using magnetic negative selection. Viahance removes dead cells, stripped nuclei and free oligonucleotides. Other dead cell separation kits only remove dead cells. Viahance's ability to remove dead cells, stripped nuclei, and free oligonucleotides from your cell culture will increase cell culture viability, improve data quality and favorably impact downstream results. Eliminating stripped nuclei and free oligonucleotides cleans up a cell preparation for future molecular biology work.

Unique Advantages of Viahance

1. Simultaneously removes stripped nuclei, free oligonucleotides and dead cells
2. Annexin-V free
3. Contains no proteins, no protein contamination
4. Provided autoclaved
5. Does not require calcium containing buffers
6. Does not require expensive magnetic filters

Sample Protocol

This protocol serves as a generic template for magnetic removal of dead cells, free nuclei, and free oligonucleotides. Modifications may be necessary to optimize the protocol for your specific needs.

Step 1: For up to 5 mL of $[(1.0 \times 10^5) \text{ to } (1.0 \times 10^6)]$ cells per ml. Collect cells in a sterile 5 ml polystyrene culture tube and determine the cell number.

Step 2: Centrifuge the cell suspension at 300g for 5 minutes. Remove the supernatant and re-suspend the pellet in 1ml sterile PBS.

Unlike Annexin V cell separation kits, Viahance does not require calcium for cell separation. Viahance performs well in PBS or HBSS. It is better to minimize the amount of serum during the separation process. Serum can interfere with separation.

Step 3: Add 0.1 ml of Viahance to the re-suspended cell pellet from Step 2. Gently pipette the solution up and down two times to ensure proper mixing. Incubate at room temperature for 5 minutes.

Step 4: Place the culture tube from Step 3 into a magnetic separator rack for 15 minutes. Dead cells, stripped nuclei, and free oligonucleotides will aggregate on the side closest to the magnet. Minimize the distance between the magnet and the tube.

Step 5: Leave the culture tube from Step 4 in the magnetic separator rack. Carefully pipette the cell suspension from the tube (~1 ml) and transfer into a sterile 15 mL centrifuge tube. Be careful to not disturb the magnetic pellet in the culture tube.

Step 6: Bring the volume of the purified cells to 10 mL with fresh buffer and pellet the cells by centrifugation (300g, 5 mins). Aspirate and discard the supernatant. This removes the unbound Viahance.

Step 7: Re-suspend the pellet from the wash into cell culture medium and process the cells according to your laboratory protocol.

Verification

Optional – Verification of dead cell removal can be done with trypan blue or propidium iodide staining.

The Products Available

- Cat No: CP-50VQ02 2 mL of Viahance
Cat No: CP-50VQ10 10 mL of Viahance

Items Needed

1. Centrifuge
2. Sterile polystyrene 12x75mm 5 mL culture tubes
3. Sterile 15 mL centrifuge tube
4. Magnetic separator
5. Sterile PBS

Frequently Asked Questions

1. *My cells grow in clumps. Can I still use this kit?*
Yes, you must first dissociate your cells. Follow your dissociation procedure, e.g., trypsin/EDTA, then properly quench before following Steps 1 through 6.
2. *You recommend minimizing serum, what is the maximum percent that I can use?*
You will need to test serum concentrations to optimize your system. We routinely perform cell purification without serum.
3. *Can I use a higher density of cells than (1.0×10^6) cells/ml?*
Yes, the density of cells can be increased up to (1.0×10^7) cells/ml. However, dead cell removal may be less efficient and slower.