LeviSelect Cell Debris Depletion Kit (10 rxn)

PRODUCT DATA SHEET

Catalog# 1004060

Description

The LeviSelect™ Cell Debris Depletion Kit is designed to efficiently remove debris from cell suspensions during viable cell enrichment on a LeviCell® instrument and cartridge. During processing, the majority of debris binds to the LeviCell cartridge consumable, while viable, untouched cells are simultaneously separated from dead cells. These enriched, healthy cells are then collected in the top fraction output of the cartridge.

Kit Components

Component	Tube PN	Quantity	Storage Conditions	Shelf Life
LeviSelect Debris Depletion Nanospheres	6000132	1x 100 µL	Store at 2-8°C. Do not freeze	Stable until expiration date on label
LeviSelect Debris Depletion Buffer	6000141	2x 1.5 mL	Store at 2-8°C. Do not freeze	Maintain at RT

Additional Tools and Consumables Required

LeviCell 1.0 System; LevitasBio PN 1000001
Or LeviCell EOS System PN 1000020
LeviCell S2.3 Cartridge; LevitasBio PN 1002010
Or LeviCell S2.3-IR Cartridge; PN1002012
LeviCell EOS-4 Cartridge; LevitasBio PN 1002101 (if using the LeviCell EOS)
Levitation Agent; LevitasBio PN 1003001 or PN 1003007
1.5 - 2.0 mL low-binding microcentrifuge tubes
2-20 µL, and 200-1000 µL pipettes and tips

Data example

Without Cell Debris depletion kit

With Cell Debris depletion kit

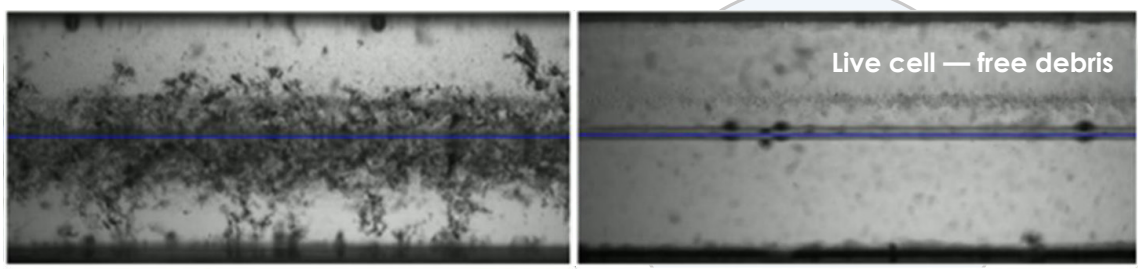
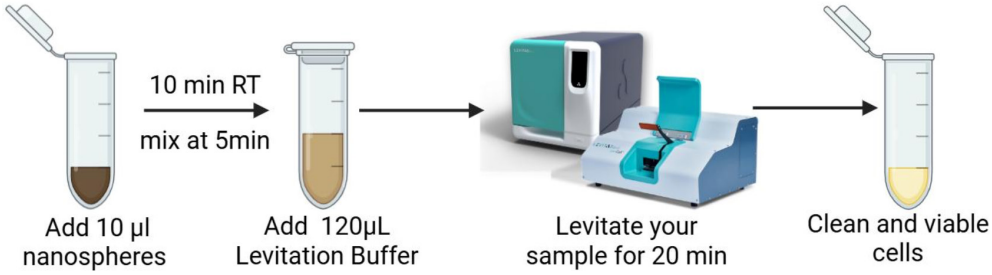


Figure 1. Depletion of debris using the Cell Debris depletion kit during a LeviCell run. Debris from dissociated chicken gizzard was mixed with 100,000 H358 cells and levitated without (left) and with the Cell Debris Depletion Kit (right).

LeviSelect Cell Debris Depletion Kit Protocol

Workflow



Prepare Levitation Buffer

Table 1. Preparation of the Levitation Buffer to achieve a concentration of 150 mM Levitation Agent in the final sample.

Reagent Volume	For 1 sample (µL; includes 10% overage)	For 4 samples (µL; includes 10% overage)
1X LeviSelect Debris Depletion Buffer	93	372
Levitation Agent	40	160
Total	133	532

LeviCell Run with Cell Debris Depletion

1. Prepare a cell suspension from tissue/cells of interest.
2. Count cell suspensions for both viability and cell concentration. If possible, take images in both the brightfield channel as well as using nuclei-specific fluorescent dyes to detect live and dead nucleated cells, for example, a combination of acridine orange and propidium iodide (AOPI). Depending on the type of debris, distinguishing between actual cells and debris can be challenging. Therefore, for the first run, aliquot no more than 2 x 10^6 total events based on the brightfield counts from the prepared cell suspension into a new 1.5 mL or 2 mL low-binding tube. Centrifuge the cells at 300 RCF for 5 minutes.
3. Remove and discard the supernatant. Resuspend cell pellet in 110 µL of 1x LeviSelect Buffer.
4. Vortex the tube containing the LeviSelect Debris Depletion Nanospheres.
5. Add 10 µL of Debris Nanospheres to the resuspended cells. Pipette mix with >80 µL 10 times. The total volume is now 120 µL.
6. Incubate the cell suspension for a total of 10 minutes at room temperature. Pipette mix the tube at 5 minutes into the incubation.
7. After incubation, add 120 µL of prepared Levitation Buffer to the 2 mL tube. The total volume is now 240 µL.
8. Pipette mix with the same pipette tip 10 times. The magnetic beads should be uniformly dispersed throughout the solution.
9. Set up the LeviCell/EOS cartridge on the instrument following the instructions on the Experiment Manager User Interface, selecting the “standard/medium” option.
10. With a P1000 pipette set to 220 µL, pipette up and down 5X to thoroughly mix (avoid bubble formation) and load 220 µL of cell suspension into the inlet well of the cartridge. The pipette tip should be placed near the backside of the well, slightly above the entrance to the flow channel.
    Note: Avoid introducing bubbles into the inlet well by not depressing the pipette plunger past its initial stop.
11. Start the LeviCell run.
12. Follow all instructions on the LeviCell up to and including the sample retrieval step. A split of 0 is advised.