

Streptavidin Microbubbles Kit

with Akadeum BACS™ Technology



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Components of Streptavidin Microbubbles (# 11110-000)

IN THE BOX:

1. BACS™ Streptavidin Microbubbles

PACKAGED SEPARATELY:

1. Separation Buffer Ca²⁺ and Mg²⁺ free PBS containing 2 mM EDTA and 0.5% biotin-free BSA
2. 5 mL Eppendorf Tubes

Additional Recommended Supplies:

1. Low retention 1,000 µL pipet tips
VWR Part #89174-530 or similar
2. Centrifuge with swinging bucket rotor
3. Vacuum aspirator

PRODUCT OVERVIEW

- > Akadeum's streptavidin microbubbles are high-buoyancy hollow glass microspheres coated in streptavidin.
- > Useful for binding biotinylated target molecules such as antibodies, proteins or DNA. This permits the capture and sorting of targets such as cells, bacteria, viruses, or molecular analytes from common samples in the life sciences.

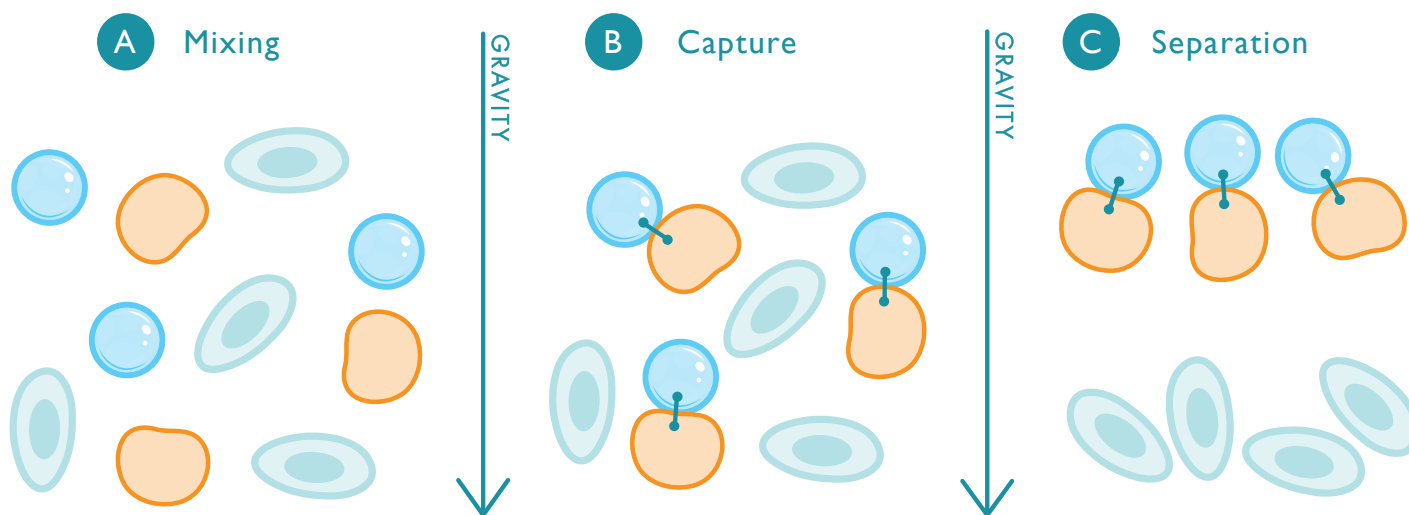
Average Particle Size: 15 µm (polydisperse mixture)

Quantity/Volume: 13.5 mL

Format: Liquid solution containing 0.09% sodium azide

Storage: Store at 4°C

MICROBUBBLE WORKFLOW



A. Akadeum Microbubbles (blue spheres) are mixed with sample containing target cells (brown) and non-target cells (teal). B. Microbubbles capture target cells by streptavidin-biotin interaction. C. Microbubbles float to the top of the sample while non-targeted cells are pelleted at the bottom of the tube.

SAFETY INFORMATION

For research use only. Not intended for any animal or human therapeutic or diagnostic use.

Before use, please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

GENERAL NOTES

For additional technology or product information please visit www.akadeum.com

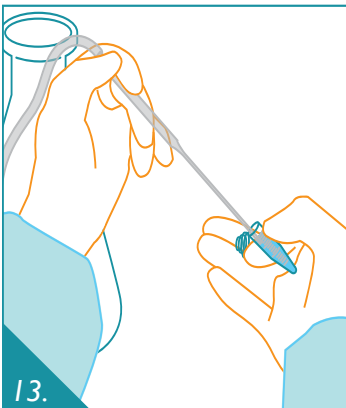
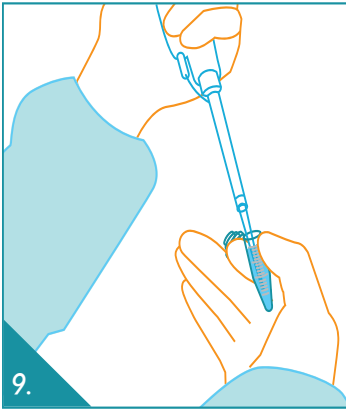
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ANTIBODY ON CELL PROCEDURE FOR MAMMALIAN CELLS

1. Begin with a single cell suspension.
2. Resuspend cell pellet at 1×10^7 cells per 50 μL of separation buffer and transfer up to 1×10^8 cells to a 5 mL Eppendorf tube.
3. Add biotin-labeled antibody to the cells (the antibody dosing must be determined for the specific application empirically). Mix briefly.

Note: The amount of antibody will vary based on target cell types and antibodies used. Akadeum suggests starting with the antibody vendors' recommended dosages for optimization.

4. Incubate sample for 20 min at 4°C .
5. Add 1 mL separation buffer and centrifuge (5 min, $400 \times g$). Aspirate supernatant.
6. Resuspend cells at 1×10^7 cells per 50 μL of separation buffer.
7. Prepare microbubbles by resuspending microbubbles in solution (mixture should be a homogeneous white solution, i.e. look like milk). Vigorously mix or pipet. Immediately proceed to Step 8.
8. Add user-determined amount of microbubbles to the first sample.

Note: The amount of microbubbles used in this step depends on target cell number and antibody performance. Akadeum recommends titrating in the range of 10-30 μL microbubbles per 10^6 target cells. Example: If depleting T cells from a 5×10^7 cell sample containing 40% T cells, titrate using 200-600 μL of microbubbles.

9. Set the pipet volume to 50% of the total sample volume (cell suspension + microbubbles) and mix with gentle trituration for 30 pipet strokes using low retention 1000 μL pipet tip.

Note: Mixing at this pipet setting ensures adequate mixing of microbubbles and cells for binding.

Example: If 5×10^7 total cells are suspended in 250 μL of buffer and 400 μL of microbubbles are added to the sample, set the pipet volume to 325 μL for mixing.

10. Immediately add 3 mL separation buffer to sample.
Note: This facilitates separation of the microbubbles from the cell pellet.
11. Repeat steps 8-10 for remaining samples making sure to resuspend microbubbles before each sample.
12. Centrifuge samples (5 min, $400 \times g$). (Akadeum recommends using a swinging bucket rotor for this step to facilitate microbubble aspiration).
13. Aspirate off white microbubble layer (containing cells targeted for depletion) and supernatant. Take care not to aspirate cell pellet.
14. Resuspend cell pellet in the desired amount of separation buffer or other buffer/media by pipetting and transfer cells to the appropriate tube/plate for use.