

# BD™ CompBead and BD™ CompBead Plus Compensation Particles

## Features

Ideal for experiments with dimly expressed antigens, antibodies that have low affinity for receptors on the cell surface, and positive populations of rare cells

Ideal for conserving cells, since no cells are used

Optimal for compensating experiments with larger cell types, for example, human embryonic stem cells (hESCs), using BD CompBead Plus particles

Recommended for use in experiments in which tandem dye conjugates (eg, PE-Cy™7, APC-Cy7) are incorporated

BD™ CompBead particles are polystyrene microparticles used to optimize fluorescence compensation settings for multicolor flow cytometric analysis. When mixed with a fluorochrome-conjugated antibody, BD CompBead particles provide distinct positive and negative (background fluorescence) stained populations that can be used to set compensation levels manually or with the automated compensation setup available in BD FACSDiva™ software.

### Simplify experimental setup with easier compensation for multicolor analysis

BD CompBead compensation particles are beads coupled to an antibody specific for the kappa light chain of immunoglobulin (Ig) from mouse, rat, or rat/hamster. Mix them with any kappa-bearing fluorochrome-conjugated antibody used in an experiment to serve as the single-stain control. Each BD CompBead set also contains a negative control, with no binding capacity, which consists of particles labeled with BSA or FBS.

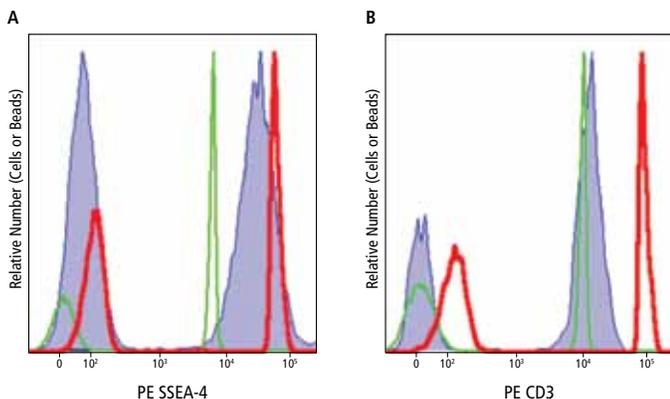
### Ultimate convenience for use with precious samples, tandem dyes, and dim or rare markers

BD CompBead particles adjust compensation using the same fluorochrome-labeled antibody used in the experiment. This method allows the researcher to more accurately establish compensation corrections for spectral overlap for any combination of fluorochrome-labeled antibodies, without having to use valuable cell samples. It also eliminates reliance on hard-dyed beads that could have potentially mismatched fluorescence spectra. And, if the positive population is dim or rare, which might compromise spectral overlap determinations, BD CompBead particles can be used instead of sample. Spectral overlap values determined from single-stained BD CompBead particles have been shown to closely match those from single-stained cells (Crowther, et al, 2002).

BD CompBead compensation particles are highly recommended for all experiments using tandem dye conjugates (eg, PE-Cy7, APC-Cy7, etc), which may have distinct spectral characteristics for each specific conjugation. Use BD CompBead particles to capture different lots of a given tandem fluorophore so that each can be measured.

### New BD CompBead Plus particles for larger cells

BD now offers BD CompBead Plus particles, which are optimal for compensating experiments with larger cell types, for example, human embryonic stem cells (hESCs), or for very bright markers. These 7.5-µm beads have forward scatter properties better suited for compensating larger cell types than the standard BD CompBead particles (3.0 to 3.4 µm) (Figure 1A). For lymphocytes and other smaller cell types, BD CompBead Plus particles are too bright (Figure 1B).



**Figure 1.** Comparison of BD CompBead and BD CompBead Plus particles on different sample types.

**A.** H9 human embryonic stem cells (WiCell, Madison, WI), and BD CompBead (Cat. No. 552843) and BD CompBead Plus particles (Cat. No. 560497) were stained with PE mouse anti-SSEA-4 (Cat. No. 560128) and analyzed on a flow cytometer. Both unstained and stained hESCs are shown in blue, negative and stained BD CompBead particles in green, and negative and stained BD CompBead Plus particles in red. Flow cytometry was performed on a BD™ LSR II flow cytometry system.

**B.** Lysed whole blood (LWB), BD CompBead particles, and BD CompBead Plus particles were stained with PE mouse anti-human CD3 (Cat. No. 555333) and analyzed on a flow cytometer. Stained LWB is shown in blue, negative and stained BD CompBead particles in green, and negative and stained BD CompBead Plus particles in red. Flow cytometry was performed on a BD LSR II flow cytometry system.

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# BD™ CompBead and BD™ CompBead Plus Compensation Particles

**NOTE:** BD Horizon™ V500- and AmCyan-conjugated reagents can show significant differences in emission spectrum on stained cells and when captured on BD CompBead particles. Thus, spillover values for these dyes evaluated with BD CompBead particles may not provide correct compensation for cells. Therefore, single-stained cellular controls are recommended for setting up compensation for AmCyan and BD Horizon V500 reagents. BD Horizon™ V500-C has been modified to minimize these spectral differences and BD CompBead particles can be used to determine spillover values for RUO antibodies conjugated to BD Horizon V500-C.

## References

Crowther EJ, Stall A, Bishop JE, Sasaki D. A compensation tool: capture beads plus software for determination of spectral overlap. Presented at ISAC XXI; May 4–9, 2002, San Diego, CA. Poster No. 57644.

## Ordering Information

### BD™ CompBead Compensation Particles

| Description            | Reg | Size      | Cat. No. |
|------------------------|-----|-----------|----------|
| Anti-rat Ig, κ         | RUO | 6 mL each | 552844   |
| Anti-rat/hamster Ig, κ | RUO | 6 mL each | 552845   |
| Anti-mouse Ig, κ       | RUO | 6 mL each | 552843   |

### BD™ CompBead Plus Particles

| Description      | Reg | Size      | Cat. No. |
|------------------|-----|-----------|----------|
| Anti-mouse Ig, κ | RUO | 6 mL each | 560497   |
| Anti-rat Ig, κ   | RUO | 6 mL each | 560499   |



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APC-Cy7: US patent 5,714,386

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