PolyFacts

Vol. 8 | No. 2 *Microspheres/Particles*

News | Views | Insights *from*

Polysciences, Inc.
Chemistry beyond the ordinary

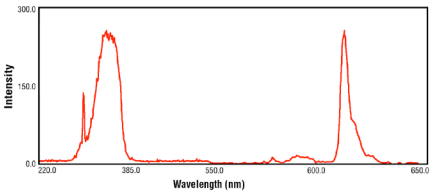
Tr(Eu) Love

Europium Chelate Spheres are Here

Despite the popularity of traditional fluorophores, distinguishing their signal from background autofluorescence can present a challenge in applications where extremely low detection limits are required. Whereas most fluorophores have pico- or nanosecond fluorescence lifetimes, compounds such as rare earth lanthanide chelates exhibit longer (microsecond) lifetimes, allowing fluorescence decay to be monitored over time. This technique provides a means to separate "true" fluorescence signal from short-lived background fluorescence, and an opportunity to improve assay sensitivity.

These same compounds are also characterized by long Stokes shifts, or intervals between fluorescence excitation and emission maxima. This property also lends itself to low background signal, and avoids regions of fluorescence overlap with other common reporters in multicolor assays.

That sounds pretty great, doesn't it?! Well, as it so happens, we were convinced, too! So... we started working on a thing or two in the lab, and are now pleased to offer europium chelate microspheres in diameters of 0.1µm, 0.2µm, and 0.3µm to address the needs of individual assays, including immunochromatographic and microwell-based formats. Our new europium products offer extremely bright fluorescence (excitation: 365nm; emission: 610nm) and exceptional stability, in addition to well-functionalized carboxylated surfaces for the covalent attachment of ligand.



Cat. No.	Description
25488	**Fluoresbrite® Europium (Eu) Carboxylate Microspheres, 0.1µm**
25489	**Fluoresbrite® Europium (Eu) Carboxylate Microspheres, 0.2µm**
25490	**Fluoresbrite® Europium (Eu) Carboxylate Microspheres, 0.3µm**

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The Latex Course™ 2012 Book is Now Available!

Unable to attend The Latex Course™ last September? Well, never fear...The Latex Course™ 2012 Book is here!

At only $279, this 270-page course book contains manuscripts from the presentations given at the conference by our speakers. Tests and assays are addressed in a range of microsphere-based formats, including turbidimetry assays and immobilization strategies.

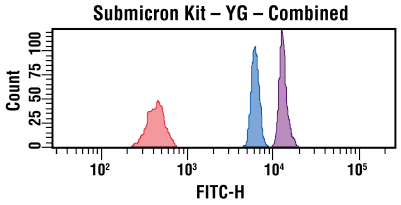
Contact us today!

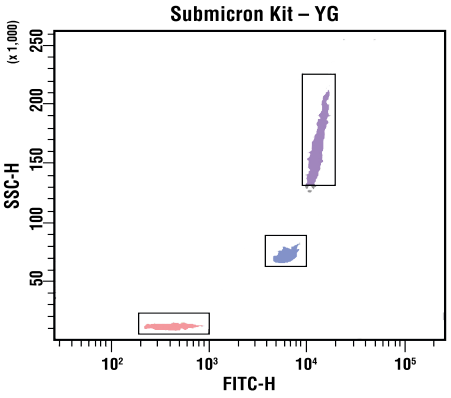
New Small Bead Calibration Kits...

... for Flow Cytometry

Current applications in flow cytometry extend far beyond traditional lymphocyte immunophenotyping, with some involving the analysis of very small particles such as platelet- and endothelial-derived microparticles, subcellular organelles, liposomes, or microbial species. While traditional flow cytometers may be used for these analyses, gating strategies are often modified to overcome the limitations of typical FSC detectors and ensure resolution of small particulates.

Our new Small Bead Calibration Kits for flow cytometry permit users to verify the resolution capabilities of the flow cytometer, and to establish appropriate instrument settings and population gates (e.g. SSC / fluorescence) for specific analyses. The use of appropriate bead standards is important for initial instrument qualification and validation for the intended application, and for ongoing QC purposes.





Cat. No.	Description
BLI832	**Submicron Bead Calibration Kit • 0.2µm, 0.5µm, 0.8µm**
BLI833	**Micron Bead Calibration Kit • 1.0µm, 3.0µm, 6.0µm**

Everyone Needs a Friend

Bead Coating Companion Reagents

The art and science of bead coating is a rather specialized pursuit. As we never want you to feel alone in it, it's become our mission to ensure that you understand the community you'll enjoy when working with us! Our fierce dedication to technical support means you'll always have someone to run ideas, questions and challenges by, and our extensive catalog of microspheres and collection of companion reagents ensures that you'll have the right tools for the task.

We have cross-linking / activation reagents for immobilizing ligand, surfactants for addressing aggregation and stabilizing suspensions, and buffers to support washing, processing and general bead health. But that's not all – we'll even tell you how to best use them! We have loads of protocols in our Technical Data Sheets, and are always happy to receive your questions via phone or email.

Cat. No.	Description
BLI5288	**DEPC-Carbodiimide (EDAC)**
BLI1909	**Glutaraldehyde, EM Grade, 25%**

Cat. No.	Description
BLI4605	**Triton® X-100 Nonionic Surfactant**
BLI6110	**Tween® 20 Nonionic Surfactant**
BLI3945	**Sodium Dodecyl Sulfate Anionic Surfactant**

And if that isn't friendship, well, we don't know what is.

Cat. No.	Description
24976	**Polysciences Bead Coupling Buffer, pH 4.5**
24977	**Polysciences Bead Coupling Buffer, pH 6.0**
24974	**Polysciences Bead Coupling Buffer, pH 7.4**
24978	**Polysciences Bead Coupling Buffer, pH 9.0**
24979	**Polysciences Bead Storage Buffer, pH 7.4**
24975	**Polysciences Bead Storage Buffer, pH 8.5**
24973	**Polysciences Bead Solution**

For more information in U.S. & Canada, please call (800) 523-2575 or email: info@polysciences.com

Particle Perplexities

Questions & Answers Pertaining to Polysciences' Microspheres / Particles

Q: I need to coat carboxyl microspheres with antibody for an assay that I'm developing. How much antibody should I include in the coating reaction?

A: When developing antibody coatings, the objective is often to achieve full bead coverage in the interest of high specific / low nonspecific binding characteristics. You may conduct the coating with a saturating amount of antibody, and then follow with a blocking molecule (e.g. BSA), or you may conduct a combined coating of Ab and blocker. The latter approach may be particularly useful when attempting to conserve expensive or scarce antibody.

We offer a number of online resources that should be helpful, including Technical Data Sheets (TDS) 238C, *Covalent Coupling of Proteins to COOH Microspheres*, and TDS 644, *PolyLink Coupling Kit*, both of which offer antibody concentrations for specific coating volumes. The antibody concentrations may be used with or without further optimization, depending on your specific requirements. For example, these procedures were developed using ~1µm carboxylate microspheres, and higher or lower antibody concentration may be appropriate for other diameters, i.e. to account for a different surface area. Smaller spheres present much higher surface area per unit weight, as illustrated in the following chart:

Table 1: Sample Values

Diameter (Microns)	Beads per gram	Surface Area ($\mu m^2/g$)
0.1	$1.8x10^{15}$	$5.7x10^{13}$
0.5	$1.5x10^{13}$	$1.1x10^{13}$
1.0	$1.8x10^{12}$	$5.7x10^{12}$
2.5	$1.2x10^{11}$	$2.3x10^{12}$
10.0	$1.8x10^{9}$	$5.7x10^{11}$

Note:
Calculations for 0.1-10.0µm are based on a suspension of polystyrene microspheres (density = 1.05 g/cm^3) at 10% solids (w/v)

Q: I am familiar with your **Quantum™ Simply Cellular® (QSC)** kits that are intended for labeling with primary antibody conjugates for quantitating cell surface markers via flow cytometry. Do you sell kits that are compatible with humanized mouse Abs?

A: **Quantum™ Simply Cellular®** kits are coated with capture antibodies that bind the Fc region of primary antibodies from the specified host, e.g. the anti-Human IgG kit will bind human IgG antibodies via their Fc regions. In the case of recombinant / humanized mouse Abs, the appropriate version of QSC will depend on the Fc region of the antibody, i.e. whether the engineered sequence was from human or mouse. For example, some humanized mouse Abs feature a murine Fc, and we would not expect these to be suited for the anti-Human IgG kit. The anti-Mouse IgG kit may be a fit, though it would need to be tested. As some recombinant antibodies have discrete structural differences from their native counterparts (e.g. lack normal glycosylation), this could impact binding to QSC beads in general. It is probably for these reasons that customers have reported varying levels of success when using recombinant antibodies.

If our antibody capture products aren't a good fit, then we suggest working with our **Quantum™ MESF** kits. These quantitative kits feature beads surface-labeled with many of the same fluorochromes that are used in flow cytometry. See our TDS 917, *Quantitative Cytometry*, for additional details.

Cat. No.	Description
BLI815	**Quantum™ Simply Cellular® anti-Mouse IgG**
BLI816	**Quantum™ Simply Cellular® anti-Human IgG**
BLI817	**Quantum™ Simply Cellular® anti-Rat IgG**

Cat. No.	Description
BLI488	**Quantum™ Alexa Fluor® 488 MESF**
BLI555	**Quantum™ FITC-5 MESF**
BLI555p	**Quantum™ FITC-5 MESF (Premix)**
BLI827	**Quantum™ R-PE MESF**
BLI828	**Quantum™ PE-Cy™5 MESF**
BLI822	**Quantum™ Cy™5 MESF**
BLI647	**Quantum™ Alexa Fluor® 647 MESF**
BLI823	**Quantum™ APC MESF**

Q: I'm planning to use carboxylated magnetic particles for my work. I see that you sell two types, **ProMag™** and **BioMag®**, and I'm not sure which to use. What do you suggest?

A: The ideal particle will depend on your application, and the two microparticles should be considered in the context of needed performance characteristics. **ProMag™** are polymer-based particles that are both spherical and highly uniform in diameter. They offer fast and uniform separations, and their lower densities ensure that they don't settle out of suspension rapidly. **BioMag®** particles are silanized iron clusters with a highly irregular morphology that results in extremely high surface area. BioMag®'s greater density allows efficient separations from difficult (e.g. highly viscous) samples. Because of their respective characteristics, we consider ProMag™ to be well suited for diagnostic assays, and BioMag® to be ideal for bioseparations. That said, these aren't hard and fast guidelines, and it may be important to try different types of particles to assess performance under specific conditions. TDS 778, *Microsphere Selection*, and TDS 855, *Magnetic Particles*, provide particle images and additional information that can aid in your decision making process, and we also invite you to continue the discussion with our technical support group.

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Follow us!

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