

## TECHNICAL DATA SHEET 431

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# Fluoresbrite® Microparticles

## Introduction

Polysciences has been a manufacturer of polystyrene-based latex particles for over 20 years. Our expertise in basic polymer particle synthesis and our commitment to innovative research has made us the world leader in the synthesis of fluorescent particles. With over 70 different standard fluorescent products, we offer a variety of particles suitable for any application. Our users have come to expect the highest quality, whether they use 1ml quantities for calibration standards or multi-liter quantities for diagnostic assays.

Over the last decade, we have witnessed an explosion in the use of Fluoresbrite particles. Our Technical Staff has compiled a list of frequently asked questions.

## Frequently Asked Questions

**Q:** How many types of dyes can be found in Fluoresbrite particles?

**A:** There are a number of standard dyes which we can easily incorporate into these polymer particles. Beyond these dyes, we can custom manufacture a Fluoresbrite particle with a dye of your choice. Our most popular dyes match the following filter settings:

BB	≡	Coumarin
YG	≡	Fluorescein
YO	≡	Rhodamine
PC Red	≡	Phycocerythrin

Dyed Particle	Excitation Max. (nm)	Emission Max. (nm)
BB	360	470
YG	441	486
YO	529	546
PC RED	491 & 512	554

**Q:** How concentrated are the particles?

**A:** The number of particles per ml may be estimated with the following equation:

$$\text{Number of particles per ml} = \frac{6W \times 10^{12}}{\rho \times \pi \times \phi^3}$$

W	=	grams of polymer per ml in latex (0.025g for a 2.5% latex)
$\phi$	=	diameter in microns of latex particles (consult label)
$\rho$	=	density of polymer in grams per ml (1.05 for polystyrene)

**Q:** How stable are the particles?

**A:** We guarantee a one year shelf life. No biocides or stabilizers are added and they are shipped in DI water only. All Fluoresbrite products should be stored at 4°C, protected from light, and protected from freezing. For applications where longer storage is required, an addition of biocide is recommended.

**Q:** How are Fluoresbrite microspheres manufactured?

**A:** Polystyrene-based spheres are internally dyed using solvent swelling / dye entrapment. The highly hydrophobic dyes (BB, YG, PC Red) remain trapped in the beads in aqueous environments. YO has limited water solubility, and some leaching may occur with aggressive washing.

**Q:** Are the Fluoresbrite particles sufficiently intense for visualization?

**A:** In a planktonic community feeding study, our particles were reported to be highly visible inside the organisms, did not decompose

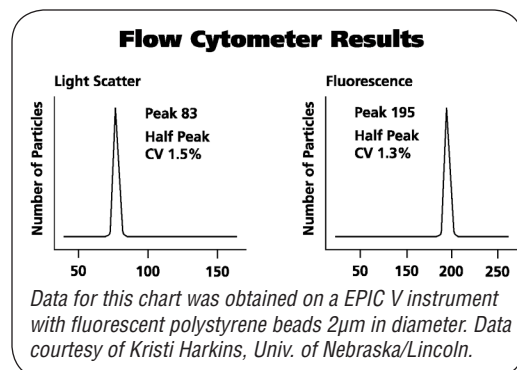
or fade, and were nonhazardous.<sup>1</sup> In fact, some customers claim that our fluorescent intensities are too strong and we offer a graded series of less intense beads to service their needs.

**Q:** Can I couple proteins to Fluoresbrite particles?

**A:** Fluoresbrite particles are internally dyed, which frees the surface of the beads for protein adsorption or covalent linking via functional groups. Polysciences is a world leader in placing particles in diagnostic assays. Our abilities to manufacture custom lots with relative control of dye content per bead has allowed for major advances in the quantification of bead-protein interactions. Even DNA has been bound to polystyrene particles.<sup>2,3</sup> See Technical Data Sheets 238C (Covalent Coupling to COOH Microspheres) and 238E (Protein Adsorption to Polystyrene Microspheres).

**Q:** Can Fluoresbrite particles be used to calibrate flow cytometers?<sup>4, 5, 6, 7</sup>

**A:** Our Fluoresbrite particles have had a long history of use as flow cytometry standards. As a manufacturer of the base particle, as well as an innovator in the incorporation of fluorescent dyes, we have advised even the instrument manufacturer on the proper types of beads to use for aligning flow cytometers. We have a special grade of calibration grade particles, pre-screened on a flow cytometer and validated as calibration quality. We also sell Bangs Laboratories' Flow Cytometry Standards, if additional products are needed.



**Q:** Can I use Fluoresbrite particles for phagocytosis<sup>8, 9, 10, 11, 12</sup> or retrograde transport?<sup>13, 14</sup>

**A:** Yes, the consistently sized polystyrene particles are ideal for cell interaction. Identification is made easy by the intense fluorescence and polystyrene has long been recognized as a biologically active surface for cell attachment. Using two different fluorescent particles allows researchers to track two populations in one study. See Technical Data Sheet 430 (Phagocytosis and Microparticles).

**Q:** Can I embed tissues which contain particles?

**A:** Latex microspheres have been visualized by light microscopy in unembedded coverslip monolayers,<sup>15</sup> in fixed or unfixed frozen sections,<sup>16, 17</sup> in paraffin section,<sup>18</sup> and glycol methacrylate kits.<sup>19</sup> For paraffin sections, n-butyl alcohol must be used for clearing and deparaffination since the typical organic solvents will destroy the beads. Successful TEM embeddings in Epon<sup>20</sup> and Spurr's<sup>21</sup> have also avoided harsh organic solvents.

**Q:** What are other applications for Fluoresbrite particles?

**A:** Our technical staff is constantly amazed at the vast array of applications for these monodisperse, fluorescent particles. Most applications depend on the intense fluorescence of Fluoresbrite microparticles to aid in the detection of the submicron particles. Their intense fluorescence has allowed medical researchers to eliminate the use of radiolabelled markers in coronary artery flow visualization studies.<sup>22</sup> The uniform size was important for a study which attempted to mimic the transport of bacteria through groundwater.<sup>23</sup> Fluorescent markers have also been used to track cells which have ingested specific particles.<sup>24, 25</sup> The diversity of fluorescent beads allowed these researchers to simultaneously track two different fluorescently tagged cell lines. The unique abilities of Polysciences to manufacture particles which emit at wavelengths from the ultraviolet to near infrared has attracted the developers of new fluorescent microscopy techniques.<sup>26</sup>

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