

Painless Particles®

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A DIVISION OF POLYSCIENCES, INC.

B E A D S ● A B O V E T H E R E S T™

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QuickCal® v. 2.3

Great News! Our **free** QuickCal Data Analysis tool (for use with our MESF and ABC Kits for quantitative flow cytometry) is easier to use and better than ever. Our version 2.3 automatically calculates the Linearity and Detection Threshold for you! Plus, all scales are now normalized to 256 for easy comparison of calibration curves between cytometers.

"On the Road Again!"

Bangs 2005 Tradeshow Schedule

❖ American Society for Microbiology

Booth #1732
June 6-8, 2005
Atlanta, GA
www.asm.org

❖ American Association for Clinical Chemistry

Booth #733
July 24-28, 2005
Orlando, FL
www.aacc.org

WHAT'S YOUR DYNAMIC RANGE?

NIST Traceable Particle Size Standards

Particle size analysis is critical to particle-based technologies in the life sciences. As the proper function of common products such as pharmaceutical active ingredients, adhesives, and printing inks depends upon the sizes of their constituent particles, the sizing instruments used to support research, manufacturing, and QC efforts in these sectors must be rigorously calibrated and validated.

Particle size standards may be used to validate sizing instruments across their dynamic ranges. They are suitable for the performance of routine calibration checks and corrections for particle sizing instruments, and in the support of practice standards, such as those published by ISO, ASTM International, CEN and other organizations. Additionally, the use of a reference material permits the standardization of results between runs, instruments, and laboratories, and across time.

Bangs Laboratories is proud to present our NIST Traceable Particle Size Standards. Our highly-characterized polymer size standards span the range of 40nm to 175µm. Additional features and benefits include:

- Certificate of Calibration and Traceability
- Convenient and economical packaging (15mL, 1% solids)
- Ease of use (dropper bottles)
- Highly competitive pricing

Bangs' NIST Traceable Particle Size Standards are provided under the following catalog codes. Please visit our website for Lot-specific information.

NT02N: 0.040 - 0.90 µm

NT03N: 1.00 - 2.00 µm

NT04N: 2.50 - 3.50 µm

NT05N: 4.00 - 175.00 µm

For more information or order placement, please contact our Customer Service Department.

Ultra Rainbow Fluorescent Particles

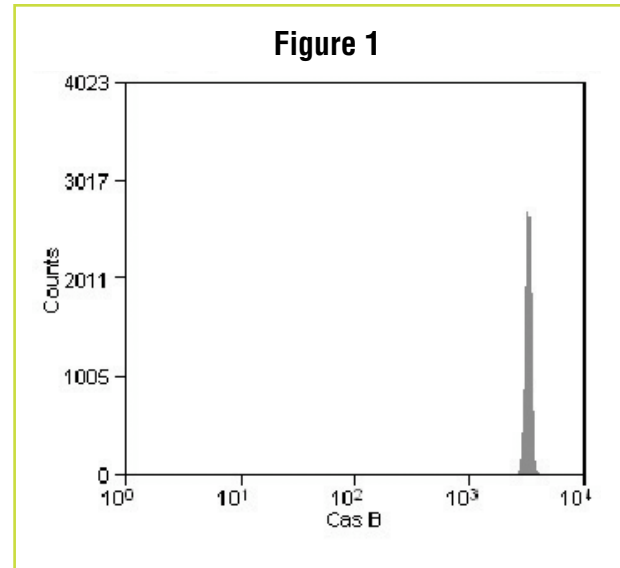
Spherotech™ Particles Now Offered at Bangs Labs

Bangs Laboratories is excited to announce our newest offering — Ultra Rainbow Fluorescent Particles, which are designed for daily QC of your flow cytometer.

Ultra Rainbow Fluorescent Particles are internally labeled with a mixture of fluorophores, which enable them to be excited at wavelengths from 365 nm to 650 nm. Enhanced UV and Far Red capabilities allow for detection in all channels (just one example shown in **Figure 1**) on your flow cytometer using a single particle prep.

Catalog No. 610: single population between 3.61µm - 3.99µm

Catalog No. 611: single population between 8.1µm - 12.0µm



P(articles)₂ = Particles Articles

Cool Articles Citing the Use of Microspheres

❖ **Optimized DNA Immobilization to Silica Microspheres**
Steinberg G, Stromborg K, Thomas L, Barker D, Zhao C. (2004) Strategies for covalent attachment of DNA to beads. *Biopolymers*; 73:597-605.

Researchers at Illumina, Inc. evaluated several strategies for the covalent immobilization of DNA to **Bangs' silica microspheres**, and their effects on hybridization kinetics and hybridization efficiency.

❖ **Beads, Beacons and Flow**
Horejsh D, Martini F, Poccia F, Ippolito G, Di Caro A, Capobianchi MR. (2005) A molecular beacon, bead-based assay for the detection of nucleic acids by flow cytometry. *Nucleic Acids Res*; 33(2):e13.

Biotinylated molecular beacons were conjugated to **streptavidin-coated microspheres** for the detection of SARS coronavirus and other respiratory pathogens via flow cytometry.

Ask "The Particle Doctor®":

Q: Do you have any products that support quantitative applications in molecular biology?

A: Our **Quantum MESF** kits have been utilized to quantitate fluorescence intensity for Flow-FISH (fluorescence *in situ* hybridization)¹ and bead-based hybridization assays² via flow cytometry. Provided that you're using a fluorescent reporter for which we offer an MESF kit (**FITC, PE, PE-Cy5, APC**), and you're evaluating the fluorescence of a "particle," be it a fluorescently-labeled cell or microsphere, our kits permit quantitation of fluorescence intensity in MESF (Molecules of Equivalent Soluble Fluorochrome) units.

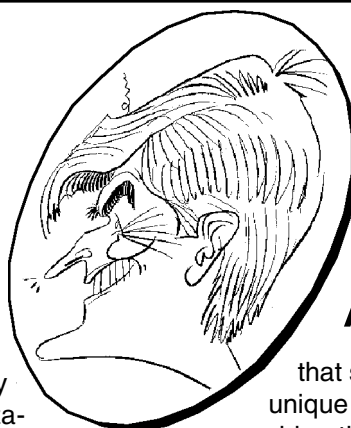
To accomplish this, the fluorescent bead set is run on the flow cytometer using the same instrument settings as for the labeled samples. The median channel values (fluorescence intensity) for the beads are entered against their assigned MESF values using the **QuickCal**® analysis template that we provide; this generates a calibration curve relating fluorescence intensity to MESF value. Channel values for the samples may then be entered into QuickCal to obtain their MESF assignments.



Telomere length measurements via flow-FISH

References:

1. Baerlocher GM, Lansdorp PM. (2003) Telomere length measurements in leukocyte subsets by automated multicolor flow-FISH. *Cytometry Part A*; 55A:1-6.
2. Spiro A, Lowe M. (2002) Quantitation of DNA sequences in environmental PCR products by multiplexed, bead-based method. *Appl Environ Microbiol*; 68(2):1010-1013.



Q: I am interested in developing an immunoassay using 100 nm microspheres. Could you offer me some tips for working with this size of particle?

A: Yes — don't.

Just kidding — although it is important to note that small polymer microspheres ($\leq 300\text{nm}$) present unique challenges, and there will be special handling considerations.

Particles in this size range are more prone to aggregation than larger spheres due to their very high surface area : volume ratios, and may require more surfactant and sonication than their larger-diameter counterparts. In fact, you may find it useful to sonicate the suspension before, during (e.g. ~every 15 minutes), and after coating. An automated particle sizer can aid in determining the level of monodispersity (i.e. fluctuation in mean diameter), as can traditional microscopy. Although you will not be able to visualize individual particles with a standard microscope, aggregates should be visible using 400X magnification.

Regarding washes, centrifugation will not be a suitable means of separation, as pelleting is likely to cause irreversible aggregation. Rather, washes are typically performed using filtration or dialysis. Filter companies can provide suggestions regarding a suitable filter or dialysis membrane or cartridge, i.e. a pore size or MWCO (Molecular Weight Cut-Off) that will retain particles, while allowing the removal of unbound antibody and blocking molecule.

Mail Bonding

(Subscribers "do the 'write' thing"!)

By far the best customer service with any sort of chemical company I have dealt with in my lab. R.W., UT

I just found what I was looking for, right in front of my nose. Thanks for a great website. J.R., New Zealand

"All truths are easy to understand once they are discovered; the point is to discover them." - Galileo Galilei

Technical References – See our website (www.bangslabs.com) for “downloadable” TechNotes, papers and Technical Data Sheets, or ask for copies by mail or fax. We continually update and add newest TechNotes and Data Sheets to our website.

Product-Specific TechNotes:

- 101. ProActive® Microspheres** – Handling tips + protocols for streptavidin, Protein A, and goat anti-mouse coated microspheres.
- 102. Magnetic Microparticles** – Characteristics, handling tips and applications for superparamagnetic particles.
- 103. Fluorescent/Dyed Microspheres** – Applications, fluorescence spectra, & product descriptions. Plus QuantumPlex™ microspheres for multiplexing, flow cytometry, and confocal microscope standards.
- 104. Silica Microspheres** – For immunoassays, nucleic acid capture, velocimetry (LDV, PIV), flat panel display spacers, others.
- 105. Microsphere Size Standards** – Beads for cell size estimation, filter challenge, and instrument checks and calibrations. NIST-traceable standards from 0.27µm to 25µm.
- 106. Confocal Standards** – Using our three, bright, single-label 60 nm fluorescent beads in confocal microscopy.

Handling-Specific TechNotes:

- 201. Working with Microspheres** – Choosing, cleaning, characterizing, coating beads, etc.
- 202. Microsphere Aggregation** – Preventing, detecting, and reversing aggregation. Chemicals and equipment sources.
- 203. Washing Microspheres** – Variety of methods for cleaning microspheres; advantages/disadvantages of methods; suppliers of equipment.
- 204. Adsorption to Microspheres** – Adsorbing proteins onto particles; use of “surface diluents” (blockers); recipes and references.
- 205. Covalent Coupling** – Chemical attachment of proteins, nucleic acids, etc. to various types of surface-functionalized microspheres; recipes for buffers, blockers; misc. coupling ideas, vendor info., and refs.
- 206. Equations** – For calculating particles/ml, area/g, “parking area”, settling velocity @ 1G and in centrifuge, etc.
- 208. Microsphere Sizing** – Various manual and automated methods are described and discussed, with references and supplier list.

Application-Specific TechNotes:

- 301. Immunological Applications** – Review of commercial applications of microspheres.
- 302. Molecular Biology** – Overview of purification and solid phase separation methods.
- 303. Lateral Flow Tests** – Putting dyed particles on membranes so they will move properly.
- 304. Light-Scattering Assays** – Turbidimetric and nephelometric applications of microspheres.

Reprints:

- 402. Microspheres, part 1: Selection, cleaning, and characterization, and part 2: Ligand attachment and test formulation** – LB Bangs & Mary Meza, *IVD Technology (in Medical Device & Diagnostic Industry)*, **17**, #3, 18-26, March, and #4, 20-26, April, 1995. (Note that you can download these papers at the IVD website: www.devicelink.com/ivdt/archive/95/03/009.html and [.../95/04/006.html](http://www.devicelink.com/ivdt/archive/95/04/006.html))
- 403. New Developments in Particle-Based Immunoassays: Introduction** – Leigh B. Bangs, *Pure & Appl. Chem.*, **68**, #10, 1873-1879 (1996) Review of 40 years of diagnostic uses of microspheres– from LATs to biosensors.
- 405. Applications of Magnetic Particles in Immunoassays** – Mary Meza, Ch. 22 (pp. 303-309) in *Scientific and Clinical Applications of Magnetic Carriers*, U Häfeli, *et al.*, Eds. Plenum Press, New York, 1997.
- 406. Measuring microsphere binding capacity** – JM Duffy, JV Wall, MB Meza, LJ Janski, *IVD Technology*, **4**, #7, 28-34 (1998). (No reprints are available; you can download from our website.)
- 407. Bead-based HTS applications in drug discovery** – MB Meza, *Drug Discovery Today: HTS supplement*, **1**, #1, 38-41 (2000).

Flow Cytometry Standards? See the “flow” portion of our website for lots of technical information about flow cytometry standardization in general and our expanding line of flow cytometry standards products in particular.

If you aren't able to locate answers to your microsphere application or handling/use questions (within our TechNotes, PolyLink and BioMag® Technical Data Sheets, FAQs, References, Product Brochures, or Product Inserts), we invite you to call us directly, or to contact “The Particle Doctor®” through our website.