

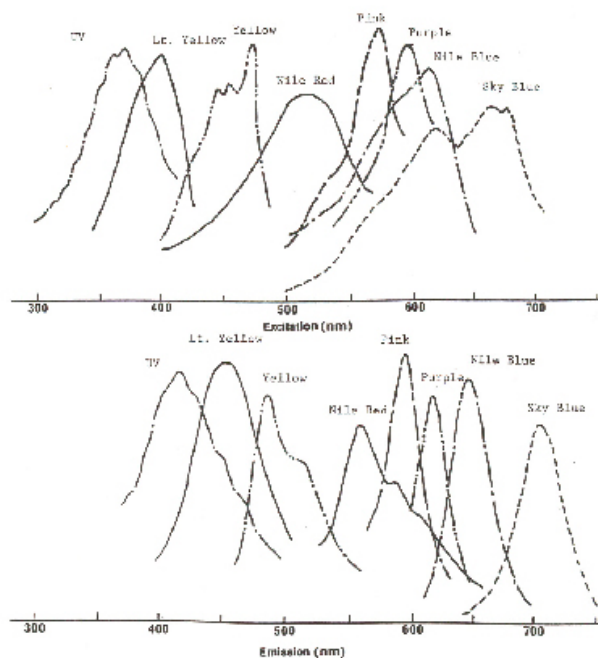
## SPHERO™ Fluorescent Particles

The SPHERO™ fluorescent microparticles are prepared by either staining the polystyrene particles with a solution of appropriate fluorophore or by polymerizing a fluorophore in styrene in the presence of polystyrene core particles. As a result, a wide variety of fluorescent particles can be prepared ranging in size, type of fluorophore, fluorescence intensity and surface functional groups. Most of the fluorophores chosen for use in the preparation of SPHERO™ fluorescent particles are water insoluble and therefore are very stable. These fluorophores, once incorporated into the particles, do not leach and their color and fluorescence remains stable for long periods of time under proper storage conditions.

The SPHERO™ fluorescent particles are available in single or multiple fluorophores of various sizes and fluorescence intensities with very small coefficient of variation in both size and fluorescence. They can be used for latex agglutination, fluorescence microscopy, confocal fluorescence microscopy and flow cytometry. The flow cytometer histograms of 2.9 μm Nile Red Particles (Catalog # FP-3056-2) at four channels are shown in Figure 2. More flow cytometry data for SPHERO™ fluorescent particles is shown on page 10.

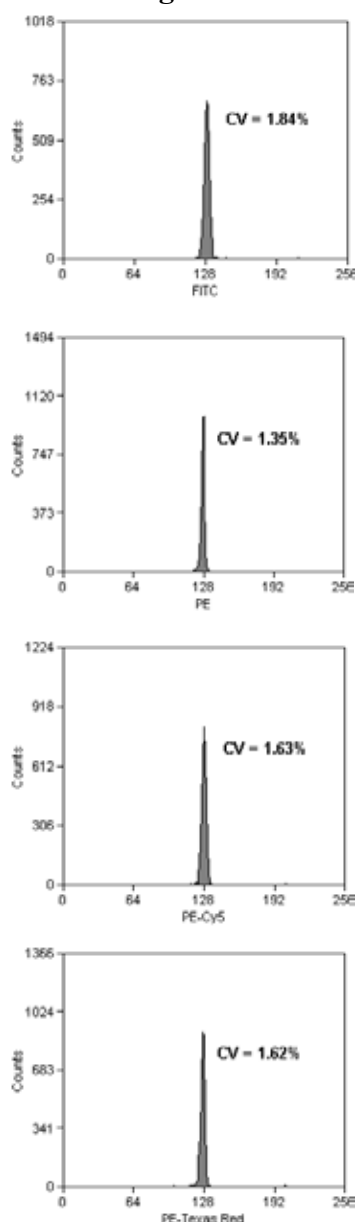
The excitation and emission spectra of some of the fluorophores used in the SPHERO™ fluorescent particles are shown in Figure 1.

**Figure 1**  
EXCITATION & EMISSION SPECTRA



NOTE: All spectra are taken with fluorescent particles in aqueous suspension.

**Figure 2**



## Spherotech, Inc.

27845 Irma Lee Circle, Unit 101, Lake Forest, IL 60045

The flow cytometry data of some of the fluorescent particles are shown in Table 1.

<b>Table 1</b>							
Fluorophores	Catalog No.	Size (µm)	%CV*				
			UV	FL	PE	TR	APC
<b>SPHERO™ Fluorescent Particles, 1.7-2.2 µm.</b>							
UV, Low Intensity	FL-2040-2	1.8	4.0				
UV, High Intensity	FH-2040-2	2.1	2.7				
Light Yellow, Low Intensity	FL-2045-2	2.1	2.2				
Light Yellow, High Intensity	FH-2045-2	2.2	1.9				
Yellow, Low Intensity	FL-2052-2	1.8		2.4	4.5		
Yellow, High Intensity	FH-2052-2	1.8		2.3	3.3		
Nile Red, Low Intensity	FL-2056-2	2.1		3.4	2.6	4.7	
Nile Red, High Intensity	FH-2056-2	2.1			2.8		
Pink	FP-2058-2	1.8			2.8	2.4	3.4
Purple, Low Intensity	FL-2062-2	1.9				2.9	4.2
Purple, Mid-level Intensity	FP-2062-2	2.0				2.4	3.0
Purple, High Intensity	FH-2062-2	1.8				2.2	
Nile Blue	FP-2065-2	2.2				3.6	4.1
Sky Blue	FP-2070-2	2.1					4.3
<b>SPHERO™ Fluorescent Particles, 2.5-4.5 µm.</b>							
Yellow	FP-4052-2	4.1		2.9			
Nile Red	FP-3056-2	2.9		1.9	1.7	2.9	
Nile Blue	FP-3065-2	3.0					3.6
Blue	FP-3068-2	3.3					2.7
<b>SPHERO™ Multiple Fluorophore Particles, 1.7-2.2 µm.</b>							
UV/LY	FP-2042-2	2.0	1.6				
PR/Y, Low Intensity	FL-2060-2	2.2		1.9	2.8	3.9	4.5
PR/Y, Mid-level Intensity	FP-2060-2	2.2		2.4		4.6	
PR/Y, High Intensity	FH-2060-2	2.0		3.0		4.2	
<b>SPHERO™ Multiple Fluorophore Particles, 2.5-5.0 µm.</b>							
UV/LY	FP-3042-2	3.2	2.2				
PK/Y	FP-3055-2	3.0		2.7	3.2		
PR/Y	FP-4060-2	4.0		2.2		5.1	
<b>SPHERO™ Fluorescent Particles, 0.7-0.9 µm.</b>							
Yellow	FP-0852-2	0.85		3.3			
Pink	FP-0858-2	0.91				6.0	
Purple	FP-0862-2	0.84				2.9	

\*Data obtained by Dr. David R. Parks, Dept. of Genetics, Stanford University Medical Center using a modified FACSII with the following Excitation and Emission wavelength:

UV: Ex 350-360, Em. 400-560 nm; FL: Ex. 488, Em. 515-545 nm;

PE: Ex. 488, Em. 560-590 nm; TR: Ex 590, Em. 615-640 nm;

APC: Ex. 590, Em. > 640 nm

Tel.: 800-368 0822 or 847-680 8922; Fax: 847-680 8927; E-Mail: [service@spherotech.com](mailto:service@spherotech.com)

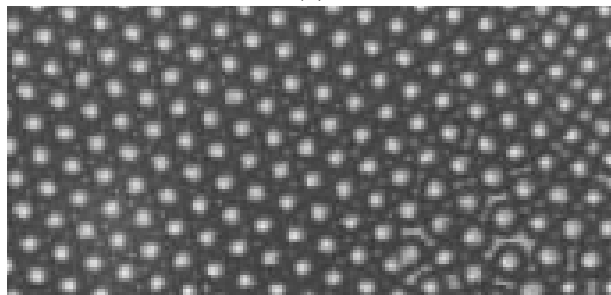
Visit us on the web at <http://www.spherotech.com>

## Spherotech, Inc.

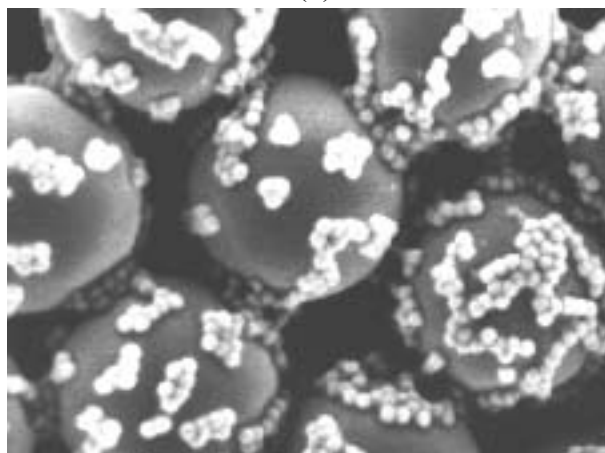
27845 Irma Lee Circle, Unit 101, Lake Forest, IL 60045

Scanning Electron Microscope (SEM) photos showing (a) 10  $\mu\text{m}$  Nile Red particles as seen under fluorescence microscope (40x ), (b) 0.4  $\mu\text{m}$  Avidin coated fluorescent particles binding to the surface of 6.0  $\mu\text{m}$  Biotin coated polystyrene particles (5000x).

(a)



(b)



### SPHERO™ Fluorescent Polystyrene

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
Yellow	0.04-0.06	1.0	FP-00552-2	2 mL
Nile Red	0.04-0.06	1.0	FP-00556-2	2 mL
Pink	0.04-0.06	1.0	FP-00558-2	2 mL
Purple	0.04-0.06	1.0	FP-00562-2	2 mL
Sky Blue	0.04-0.06	0.25	FP-00570-2	2 mL
Light Yellow	0.1-0.3	1.0	FP-0245-2	2 mL
Yellow	0.1-0.3	1.0	FP-0252-2	2 mL
Nile Red	0.1-0.3	1.0	FP-0256-2	2 mL
Purple	0.1-0.3	1.0	FP-0262-2	2 mL
Sky Blue	0.1-0.3	0.25	FP-0270-2	2 mL
Light Yellow	0.4-0.6	1.0	FP-0545-2	2 mL
Yellow	0.4-0.6	1.0	FP-0552-2	2 mL
Nile Red	0.4-0.6	1.0	FP-0556-2	2 mL
Pink	0.4-0.6	1.0	FP-0558-2	2 mL
Purple	0.4-0.6	1.0	FP-0562-2	2 mL
Sky Blue	0.4-0.6	1.0	FP-0570-2	2 mL
Light Yellow	0.7-0.9	1.0	FP-0845-2	2 mL
Yellow	0.7-0.9	1.0	FP-0852-2	2 mL
Nile Red	0.7-0.9	1.0	FP-0856-2	2 mL
Pink	0.7-0.9	1.0	FP-0858-2	2 mL
Purple	0.7-0.9	1.0	FP-0862-2	2 mL
Blue	0.7-0.9	1.0	FP-0868-2	2 mL
Sky Blue	0.7-0.9	1.0	FP-0870-2	2 mL
Light Yellow, Medium Intensity	1.7-2.2	1.0	FP-2045-2	2 mL
Nile Red	1.7-2.2	1.0	FP-2056-2	2 mL
Pink, Medium Intensity,	1.7-2.2	1.0	FP-2058-2	2 mL
Purple , Medium Intensity	1.7-2.2	1.0	FP-2062-2	2 mL
Nile Blue	1.7-2.2	1.0	FP-2065-2	2 mL
Blue	1.7-2.2	1.0	FP-2068-2	2 mL
Sky Blue	1.7-2.2	1.0	FP-2070-2	2 mL
Yellow	2.5-4.5	1.0	FP-3052-2	2 mL

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
Nile Red	2.5-4.5	1.0	FP-3056-2	2 mL
Pink	2.5-3.4	1.0	FP-3058-2	2 mL
Nile Blue	2.5-4.5	1.0	FP-3065-2	2 mL
Blue	2.5-4.5	1.0	FP-3068-2	2 mL
Ocean Blue	2.5-4.5	1.0	FP-3069-2	2 mL
Sky Blue	2.5-4.5	1.0	FP-3070-2	2 mL
Yellow	2.5-4.5	1.0	FP-4052-2	2 mL
Nile Red	2.5-4.5	1.0	FP-4056-2	2 mL
Pink	2.5-4.5	1.0	FP-4058-2	2 mL
Purple	2.5-4.5	1.0	FP-4062-2	2 mL
Sky Blue	2.5-4.5	0.2	FP-4070-2	2 mL
Yellow	5.0-7.9	1.0	FP-6052-2	2 mL
Nile Red	5.0-7.9	1.0	FP-6056-2	2 mL
Pink	6.0-8.0	1.0	FP-6058-2	2 mL
Ocean Blue	5.0-7.9	0.2	FP-6069-2	2 mL
Yellow	7.0-7.9	1.0	FP-7052-2	2 mL
Sky Blue	7.0-7.9	0.25	FP-7070-2	2 mL
Nile Red	8.1-9.9	1.0	FP-9056-2	2 mL
Light Yellow	10.0-14.0	1.0	FP-10045-2	2 mL
Yellow	10.0-14.0	1.0	FP-10052-2	2 mL
Nile Red	10.0-14.0	1.0	FP-10056-2	2 mL
Sky Blue	10.0-14.0	0.2	FP-10070-2	2 mL
Yellow	15.0-19.0	1.0	FP-15052-2	2 mL
Nile Red	15.0-19.0	1.0	FP-15056-2	2 mL
PAK Blue	15.0-19.0	1.0	FP-15067-2	2 mL
Sky Blue	15.0-19.0	0.2	FP-15070-2	2 mL
Yellow	18.0-24.9	1.0	FP-20052-2	2 mL
Nile Red	18.0-24.9	1.0	FP-20056-2	2 mL
Yellow	25.0-35.0	1.0	FP-30052-2	2 mL
Nile Red	25.0-35.0	1.0	FP-30056-2	2 mL
Purple	25.0-35.0	1.0	FP-30062-2	2 mL

Tel.: 800-368 0822 or 847-680 8922; Fax: 847-680 8927; E-Mail: [service@spherotech.com](mailto:service@spherotech.com)

Visit us on the web at <http://www.spherotech.com>

## Spherotech, Inc.

27845 Irma Lee Circle, Unit 101, Lake Forest, IL 60045

### SPHERO™ High Intensity Fluorescent Polystyrene

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
UV	1.7-2.2	1.0	FH-2040-2	2 mL
Light Yellow	1.7-2.2	1.0	FH-2045-2	2 mL
Yellow	1.7-2.2	1.0	FH-2052-2	2 mL
Nile Red	1.7-2.2	1.0	FH-2056-2	2 mL
Purple	1.7-2.2	1.0	FH-2062-2	2 mL
Sky Blue	1.7-2.2	0.2	FH-2070-2	2 mL
Nile Red	10.0-14.0	1.0	FH-10056-2	2 mL

### SPHERO™ Fluorescent PMMA

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
Yellow	38.0-44.0	1.0	FPMA-40052-5	5 mL
Nile Red	38.0-44.0	1.0	FPMA-40056-5	5 mL
Purple	38.0-44.0	1.0	FPMA-40062-5	5 mL
Nile Red	45.0-52.0	0.2	FPMA-50056-5	5 mL
Purple	53.0-62.0	1.0	FPMA-60062-5	5 mL

### SPHERO™ Low Intensity Fluorescent Polystyrene

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
UV	1.7-2.2	1.0	FL-2040-2	2 mL
Light Yellow	1.7-2.2	1.0	FL-2045-2	2 mL
Yellow	1.7-2.2	1.0	FL-2052-2	2 mL
Nile Red	1.7-2.2	1.0	FL-2056-2	2 mL
Purple	1.7-2.2	1.0	FL-2062-2	2 mL
Blue	1.7-2.2	1.0	FL-2068-2	2 mL
Blue	2.5-4.5	1.0	FL-3068-2	2 mL
Sky Blue	2.4-4.5	1.0	FL-4070-2	2 mL
Nile Red	5.0-7.9	1.0	FL-6056-2	2 mL
Blue	6.0-8.0	1.0	FL-6068-2	2 mL
Nile Red	10.0-14.0	1.0	FL-10056-2	2 mL

## SPHERO™ Functionalized Fluorescent Particles

The carboxyl and amino fluorescent particles are used for covalent coupling of proteins or ligands. For example, these fluorescent particles can be coated with Avidin, Biotin, Protein A, Goat anti-Mouse IgG, or other protein of interest. The coated fluorescent particles can then be used as fluorescent tracers for cell surface markers in fluorescence microscopy and flow cytometry.

### SPHERO™ Amino Fluorescent Particles

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
Yellow	0.09-0.3	1.0	AFP-0252-2	2 mL
Yellow	0.4-0.6	1.0	AFP-0552-2	2 mL
Pink	0.4-0.6	1.0	AFP-0558-2	2 mL
Yellow	0.7-0.9	1.0	AFP-0852-2	2 mL
Nile Red	0.7-0.9	1.0	AFP-0856-2	2 mL
Pink	0.7-0.9	1.0	AFP-0858-2	2 mL
Nile Blue	0.7-0.9	1.0	AFP-0865-2	2 mL
Yellow	38.0-44.0	1.0	AFP-40052-5	5 mL

### SPHERO™ Carboxyl High Intensity Fluorescent Particles

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
Yellow	5.0-5.9	0.5	CFH-5052-2	2 mL
Nile Red	5.0-5.9	0.5	CFH-5056-2	2 mL
Pink	5.0-5.9	0.5	CFH-5058-2	2 mL
Nile Blue	5.0-5.9	0.5	CFH-5065-2	2 mL

### SPHERO™ Carboxyl Fluorescent Particles

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
Yellow	0.04-0.08	1.0	CFP-00552-2	2 mL
Nile Red	0.04-0.08	1.0	CFP-00556-2	2 mL
Pink	0.04-0.08	1.0	CFP-00558-2	2 mL
Yellow	0.09-0.3	1.0	CFP-0252-2	2 mL
Nile Red	0.09-0.3	1.0	CFP-0256-2	2 mL
Pink	0.09-0.3	0.9	CFP-0258-2	2.2 mL
Yellow	0.4-0.6	1.0	CFP-0552-2	2 mL
Nile Red	0.4-0.6	1.0	CFP-0556-2	2 mL
Pink	0.4-0.6	1.0	CFP-0558-2	2 mL
Purple	0.4-0.6	1.0	CFP-0562-2	2 mL
Sky Blue	0.4-0.6	0.25	CFP-0570-2	2 mL
Yellow	0.7-0.9	1.0	CFP-0852-2	2 mL
Nile Red	0.7-0.9	1.0	CFP-0856-2	2 mL
Pink	0.7-0.9	1.0	CFP-0858-2	2 mL
Purple	0.7-0.9	1.0	CFP-0862-2	2 mL
Sky Blue	0.7-0.9	1.0	CFP-0870-2	2 mL
Yellow	1.7-2.2	1.0	CFP-2052-2	2 mL
Pink	1.7-2.2	1.0	CFP-2058-2	2 mL
Purple	1.7-2.2	1.0	CFP-2062-2	2 mL
Light Yellow	5.0-5.9	0.5	CFP-5045-2	2 mL
Yellow	5.0-5.9	0.5	CFP-5052-2	2 mL
Nile Red	5.0-5.9	0.5	CFP-5056-2	2 mL
Pink	5.0-5.9	0.5	CFP-5058-2	2 mL
Nile Blue	5.0-5.9	0.5	CFP-5065-2	2 mL
Sky Blue	5.0-5.9	0.5	CFP-5070-2	2 mL

Tel.: 800-368 0822 or 847-680 8922; Fax: 847-680 8927; E-Mail: [service@spherotech.com](mailto:service@spherotech.com)

Visit us on the web at <http://www.spherotech.com>

## Spherotech, Inc.

27845 Irma Lee Circle, Unit 101, Lake Forest, IL 60045

### SPHERO™ Carboxyl Low Intensity Fluorescent Particles

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
Yellow	0.7-0.9	1.0	CFL-0852-2	2 mL
Yellow	5.0-5.9	0.5	CFL-5052-2	2 mL
Pink	5.0-5.9	0.5	CFL-5058-2	2 mL
Nile Blue	5.0-5.9	0.5	CFL-5065-2	2 mL

### SPHERO™ Multiple Fluorophore Fluorescent Particles

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
Multiple Fluorophore	0.1-0.3	0.2	FP-0257-2	2 mL
Multiple Fluorophore	0.4-0.6	0.2	FP-0557-2	2 mL
Multiple Fluorophore	0.7-0.9	0.2	FP-0857-2	2 mL
Multiple Fluorophore	1.7-2.2	0.2	FP-2057-2	2 mL
Multiple Fluorophore	2.5-5.0	0.2	FP-3057-2	2 mL
Multiple Fluorophore	6.0-7.9	1.0	FP-6057-2	2 mL
UV / Light Yellow	0.7-0.9	1.0	FP-0842-2	2 mL
UV / Light Yellow	1.7-2.2	1.0	FP-2042-2	2 mL
UV / Purple / Yellow / Pink / Nile Blue	1.7-2.2	1.0	FP-2054-2	2 mL
Purple / Yellow, Medium Intensity	1.7-2.2	1.0	FP-2060-2	2 mL
UV / Light Yellow	2.5-5.0	1.0	FP-3042-2	2 mL
Pink / Yellow	2.5-5.0	1.0	FP-3055-2	2 mL
Nile Red / Blue	2.5-4.5	1.0	FP-3066-2	2 mL
Purple / Yellow	2.5-5.0	1.0	FP-4060-2	2 mL
Nile Red / Blue	4.6-5.9	1.0	FP-5066-2	2 mL
Purple / Yellow, High Intensity	1.7-2.2	1.0	FH-2060-2	2 mL
Purple / Yellow, Low Intensity	1.7-2.2	1.0	FL-2060-2	2 mL

### SPHERO™ Dimethylamino Fluorescent Particles

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
Purple	2.5-3.5	1.0	DFP-3062-2	2 mL

### SPHERO™ Carboxyl Multiple Fluorophore Fluorescent Particles

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
UV / Light Yellow / Yellow	0.04-0.08	1.0	CFP-00546-2	2 mL

### SPHERO™ FITC Polystyrene Particles

The FITC labeled polystyrene particles are used as calibration particles for flow cytometry. In addition, they also are used to cross calibrate different flow cytometers so that data can be normalized.

Particle Type and Surface	Size, $\mu\text{m}$	% w/v	Catalog No.	Unit
FITC	0.7-0.9	0.1	F1CP-08-2	2 mL
FITC	2.0-2.9	0.1	F1CP-20-2	2 mL
FITC	3.0-3.9	0.1	F1CP-30-2	2 mL
FITC	5.0-5.9	0.1	F1CP-50-2	2 mL
FITC	7.0-7.9	0.1	F1CP-70-2	2 mL
FITC	8.0-8.9	0.1	F1CP-80-2	2 mL