SPHERO™ Functionalized Polystyrene Particles

Specific particle surface chemistry enables a broad range of coating and binding applications. SPHERO™ functionalized polystyrene particles provide reactive groups on uniform microparticles for consistent and repeatable coating and binding. There are several ways to prepare particles with functionalized surfaces.

- Polymerization initiator selection
- Functionalized monomer grafting

A variety of functional groups can be provided on the microparticle's exterior surface by selecting the appropriate polymerization initiator. For instance, if potassium persulfate is used as an initiator for polymerization the particle will have sulfate groups. Similarly, other functional groups can be introduced on the surface of the particles by using other functionalized initiators.

Another method for providing surface functional groups is by grafting functionalized monomer after the polymerization process. This type of functionalized polystyrene particles are prepared by coating a thin layer of functionalized monomer onto the surface of plain particles. As a result, all of the functional groups are on the surface of the particles. The functional groups are attached to the surface of the particles by alkyl chains of two to eight carbons in length depending upon the type of functionalized monomer used.

Below are the surface charge densities for 0.8 µm carboxyl and amino particles:

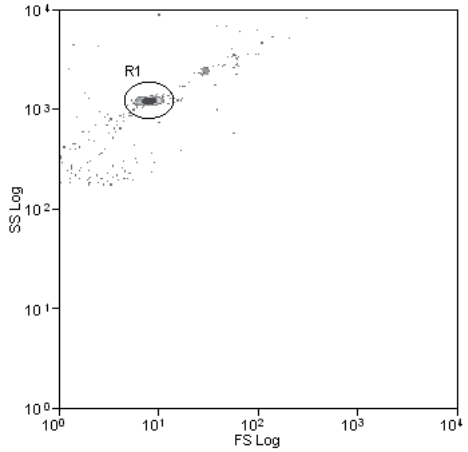
- SPHERO™ carboxyl polystyrene particles with a diameter of 0.8 µm typically contain ~ 50 µeq/g of carboxyl groups on their surface.
- SPHERO™ amino polystyrene particles with a diameter of 0.8 µm typically contain ~ 15 to 20 µeq/g of amino groups on their surface.

Nonetheless, the choice of particle size and type is dependent upon the intended application. For instance, particles with size of 0.4 to 2.0 µm are suitable for latex agglutination assay, solid phase enzyme immunoassay or solid phase fluorescence immunoassay. Particles with size of 2.0 µm or larger are preferred for flow cytometry applications.

SPHERO™ Carboxyl Polystyrene

Particle Type and Surface	Size, µm	% w/v	Catalog No.	Unit
Carboxyl-polystyrene	0.05-0.1	2.5	CP-008-20	20 mL
			CP-008-200	200 mL
Carboxyl-polystyrene	0.2-0.3	5.0	CP-025-10	10 mL
			CP-025-100	100 mL
Carboxyl-polystyrene	0.4-0.6	5.0	CP-05-10	10 mL
			CP-05-100	100 mL
Carboxyl-polystyrene	0.7-0.9	5.0	CP-08-10	10 mL
			CP-08-100	100 mL
Carboxyl-polystyrene	1.0-1.4	5.0	CP-10-10	10 mL
			CP-10-100	100 mL
Carboxyl-polystyrene	1.5-1.9	5.0	CP-15-10	10 mL
			CP-15-100	100 mL
Carboxyl-polystyrene	2.0-2.4	5.0	CP-20-10	10 mL
			CP-20-100	100 mL
Carboxyl-polystyrene	2.5-2.9	5.0	CP-25-10	10 mL
			CP-25-100	100 mL
Carboxyl-polystyrene	3.0-3.4	5.0	CP-30-10	10 mL
			CP-30-100	100 mL
Carboxyl-polystyrene	3.5-3.9	5.0	CP-35-10	10 mL
			CP-35-100	100 mL
Carboxyl-polystyrene	4.0-4.4	5.0	CP-40-10	10 mL
			CP-40-100	100 mL
Carboxyl-polystyrene	4.5-4.9	5.0	CP-45-10	10 mL
			CP-45-100	100 mL
Carboxyl-polystyrene	5.0-5.9	5.0	CP-50-10	10 mL
			CP-50-100	100 mL
Carboxyl-polystyrene	6.0-8.0	5.0	CP-60-10	10 mL
			CP-60-100	100 mL

Figure 9 Flow cytometry forward scatter vs. side scatter dot plot for Cat. No. CP-08-10 Lot AD01.



SPHERO™ Amino Polystyrene

Particle Type and Surface	Size, µm	% w/v	Catalog No.	Unit
Amino-polystyrene	0.2-0.3	2.5	AP-025-10	10 mL
			AP-025-100	100 mL
Amino-polystyrene	0.4-0.6	5.0	AP-05-10	10 mL
			AP-05-100	100 mL
Amino-polystyrene	0.7-0.9	5.0	AP-08-10	10 mL
			AP-08-100	100 mL
Amino-polystyrene	1.0-1.4	5.0	AP-10-10	10 mL
			AP-10-100	100 mL
Amino-polystyrene	2.0-2.4	5.0	AP-20-10	10 mL
			AP-20-100	100 mL
Amino-polystyrene	2.5-2.9	5.0	AP-25-10	10 mL
			AP-25-100	100 mL
Amino-polystyrene	3.0-3.4	5.0	AP-30-10	10 mL
			AP-30-100	100 mL
Amino-polystyrene	3.5-3.9	5.0	AP-35-10	10 mL
			AP-35-100	100 mL
Amino-polystyrene	6.0-8.0	5.0	AP-60-10	10 mL
			AP-60-100	100 mL
Amino-polystyrene	8.0-12.9	1.0	AP-100-10	10 mL

SPHERO™ Jeffamine Polystyrene

Contains a PEG-based spacer arm that is terminated with amine groups

Particle Type and Surface	Size, µm	% w/v	Catalog No.	Unit
Jeffamine®	1.0-1.4	1.0	JAP-10-5	5 mL

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SPHERO™ Sulfonate Polystyrene

Particle Type and Surface	Size, µm	% w/v	Catalog No.	Unit
Sulfonate-polystyrene	0.7-0.9	5.0	SP-08-10	10 mL

SPHERO™ Hydroxy Polystyrene

Particle Type and Surface	Size, µm	% w/v	Catalog No.	Unit
Hydroxy-polystyrene	0.7-0.9	5.0	HP-08-10	10 mL
			HP-08-100	100 mL

SPHERO™ Dimethylamino Polystyrene

Particle Type and Surface	Size, µm	% w/v	Catalog No.	Unit
Dimethylamino-polystyrnene	0.7-0.9	5.0	DP-08-10	10 mL
			DP-08-100	100 mL

SPHERO™ Functionalized Particle Application Examples and Recommendations:

- Carboxyl or Amino functionalized particles are very useful for covalent coupling of proteins, ligands, antibodies or antigens to the surface of the microparticles using water soluble carbodiimide as the coupling agent. Figure 9 is a diagram of antibody and protein coating of carboxyl and amino particles using EDC coupling.
- Varying particle surface charges can be obtained using functionalized particles such as hydroxyl, sulfate and dimethylamino. These particles are used to manipulate the orientation of the coated material by passive adsorption.
- Polyclonal antibodies can be coated to polystyrene particles by passive adsorption. According to our experience, the optimal amount of antibody to particles ratio is ~100 µg of antibody per mL of 0.5% w/v (5 mg solid per mL) of 0.8 µm polystyrene. Since the total surface area of the particles is inversely proportional to the diameter of the particles, the amount of antibody to particles ratio needs to be adjusted accordingly.
- The washing of polystyrene particles to remove unbound proteins or ligands during coating is accomplished by centrifugation or tangential flow filtration for particles with size of 0.4 µm or larger. For smaller size particles gel filtration, dialysis, or tangential flow filtration should be used.

Please refer to SPHERO™ Recommended Coating Procedures catalog pages for more information.

Figure 10 Examples of carbodiimide-mediated coupling processes.