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TECHNICAL DATA SHEET 635

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Silica Microspheres

Description

Inorganic supports such as silica microspheres have become increasingly important for a variety of applications. They offer the combined benefits of working with a bead platform and the unique properties of a silica substrate. The benefits include the flexibility to coat any number of bead populations with the biomolecule of choice, a large specific surface area, and improved binding kinetics over planar surfaces. They are also amenable to different silanization chemistries and have a unique refractive index and density, low auto-fluorescence, low nonspecific binding of many biomolecules, hydrophilicity, and are easy to manipulate. Our inorganic microspheres, made from pure silica (SiO₂), are available as aqueous suspensions. These particles are important in a variety of diverse applications, such as: nucleic acid purification, flat panel displays, velocimetry studies, and immunoassays.

Characteristics

Mean diameters available: 0.01µm to 5µm Standard Deviation: <10% Density: 1.96 g/cm³ Refractive Index: 1.42-1.46 (at 589nm) Dielectric Constant: 2.8 Porosity: None Surface: Hydroxyl



Procedure

Researchers are advised to optimize the use of particles in any application.

Nucleic Acid Isolation

Negatively charged biomolecules, such as nucleic acid, will bind to silica in the presence of divalent cations (e.g. Ca^{2+} , Mg^{2+}). Protocols have also been developed for the adsorption of nucleic acid to siliceous supports in the presence of salt and chaotropes.^{1, 3} In addition, newly developed protocols for binding of DNA to glass surfaces may be adapted for use with silica microspheres.^{2,4} In general nucleic acid is isolated from materials such as serum or cell lysates, by mixing the material with a chaotropic substance and a nucleic acidbinding solid phase (silica). For example, DNA can be isolated from human serum when 3M KI, Nal, or NaSCN is used in combination with 8M urea as chaotropic substances. It is also possible to reverse the overall charge of silica and adsorb negatively charged DNA. This is accomplished by rinsing clean silica in a 0.1-1M CaCl₂ solution. The Ca²⁺ ions would normally coagulate the microspheres; but, if microspheres are put into a solution with excess Ca²⁺ ions very quickly, with good mixing, then every negative charge will pick up a Ca²⁺ ion. The result will be a positively charged surface. Wash the microspheres with very clean deionized water to remove excess Ca²⁺ and counterions. Negatively charged DNA will then adsorb directly onto positively charged silica microspheres.

Cell and Biomolecule Purification

With its high density, silica has also been utilized as an alternative to other supports (e.g. magnetic particles) for the separation of cells or biomolecules. An antibody or other suitable capture molecule is first bound to functionalized silica. The coated silica microspheres are then mixed with sample to bind the targeted cell population(s) or analyte(s). Centrifugation or simple settling may then be utilized to isolate the targeted cells or biomolecules.

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Spacers in Flat Panel Displays

Microspheres maintain a uniform gap between the two glass panels used to form a flat panel display (such as the display on a laptop computer). Our silica microspheres are the perfect material for displays that require great uniformity in gap space, coupled with little compressibility and high temperature tolerance (~1000°C). Especially suited for use in epoxy seals that require high curing temperatures, these chemically inert microspheres will not stack atop each other, as cut class often does. We are able to accommodate display manufacturers by supplying our microspheres as either free-flowing powders or in aqueous suspensions.

Seed Particles for Velocimetry

Because of our silica microspheres' unique properties, they are suitable for Laser Doppler Velocimetry (LDV), Particle Imaging Velocimetry (PIV), Digital Imaging Velocimetry (DIV), Laser Speckle Velocimetry (LSV), and other methods of flow visualization and measurement. The wide variety of diameters permits optimum choice of microspheres large enough to give good signal-to-noise ratios, yet small enough to accurately follow the flow. Tight control of the microsphere diameter (standard deviations typically <10%) means that the microspheres respond uniformly, with all particles moving at the same speed in a flow stream. High temperature resistance (~1000°C) makes them especially well suited for studying flows at elevated temperatures.

Immunoassays and Miscellaneous Applications

Hydrophilic silica does not adsorb proteins well, therein, our microspheres are used in immunoassays in which very low non-specific protein binding is vital. The density of silica makes it ideal for easy, rapid separation in tests and immunoassays. Optical based tests and assays take advantage of its unique refractive index (1.42-1.46 versus 1.59 for polystyrene, at 589nm). As an example, silica microspheres (1µm in diameter) were used as the solid support in a series of laser trapping experiments to study the interaction of actin and myosin. These microspheres are very easily manipulated.^{5,6,7}

Notes

Drying Silica Microspheres

Silica microspheres (>0.5µm in diameter) can be dried to a free flowing powder; however, they should first be washed with an organic solvent such as ethanol or THF. To change the liquid phase, gradually move the microspheres through water / ethanol (or THF) solutions with increasing solvent concentration. Separate the microspheres from solution by allowing them to settle, then remove most of the liquid. Alternatively, they may be centrifuged or filtered. The microspheres are then dried to form a moist cake, either in the open air or in a drying oven (with or without a vacuum). If using a drying oven at 70°C, it will take approximately 24 hours. After drying, the cake should be crushed with a mortar and pestle and dried again. After final crushing with a mortar and pestle, the microspheres will be in the form of a free flowing powder.

Suspending Powdered Microspheres

Dry silica microspheres can be easily dispersed in water or aqueous solutions. Begin by adding the appropriate weight of silica powder to buffer. Dilute solutions are easier to work with, so using the lowest concentration possible is helpful. Vortex well to mix. Suspend the vial or tube containing the silica suspension in a sonic bath. (Sonic probes are not useful at dispersing powders.) Better sonication is achieved if the vessel containing the suspension is held above the floor of the sonic bath with a clamp, rather than resting on the bottom. The bath must also be filled to the proper level, which depends on the model. Sonicate for approximately 10 minutes, then confirm that the microspheres are dispersed by viewing under a light microscope. Individual microspheres ~1µm in diameter are visible under 1000x magnification. Clumps made of microspheres <1µm in diameter will be clearly visible under the same magnification. If clumps are visible, sonicate again for 10 minutes. Continue with 10 minute cycles until the microspheres are completely dispersed.

Calculating Particles per Milliliter

The number of particles per milliliter will vary with the specified weight to volume (w/v), density of particle composition, and diameter of the particle. The number of particles per milliliter can be calculated using the following equation:

6x • 10 ¹²	x = particle weight (g) per ml
y • π • Z ³	y = density (g/ml)
	z = diameter (µm)

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Storage and Stability

Store at 4°C. Freezing may result in irreversible aggregation and loss of binding activity.

This product is for research use only and is not intended for use in humans or for *in vitro* diagnostic use.

Ordering Information

Cat. #	Description	Size
24298	Silica Microspheres, 0.01µm (broad distribution	on) 10ml
24040	Silica Microspheres, 0.05µm	10ml
24041	Silica Microspheres, 0.10µm	10ml
24320	Silica Microspheres, 0.15µm	15ml
24321	Silica Microspheres, 0.30µm	15ml
24322	Silica Microspheres, 0.40µm	15ml
24042	Silica Microspheres, 0.45µm	10ml
24323	Silica Microspheres, 0.50µm	15ml
24324	Silica Microspheres, 0.70µm	15ml
24325	Silica Microspheres, 0.90µm	15ml
24326	Silica Microspheres, 1.0µm	15ml
24327	Silica Microspheres, 1.5µm	15ml
24328	Silica Microspheres, 2.0µm	15ml
24329	Silica Microspheres, 2.5µm	15ml
24330	Silica Microspheres, 3.0µm	15ml
24331	Silica Microspheres, 4.0µm	15ml
24332	Silica Microspheres, 5.0µm	15ml
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