

# Multifunctional Hybrid Silica Particles for Multicolor Imaging and Multiplex Tasking

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Multifunctional submicron particles have recently emerged as great promises for biological applications such as bio-labeling, medical diagnostics, and drug delivery.<sup>1,2,3,4,5,6</sup> These particles are non-toxic and have size comparable to biomolecules, becoming very suitable for biomedical applications. The particle structures can be engineered to perform multicolor imaging, as well as multiplex tasking such as site-selective binding, detection, and separation.<sup>7,8,9,10,11</sup> In addition, these particles can load a large amount of fluorescent dye for signal amplification.<sup>12,13,14,15,16</sup> Several synthetic techniques have been developed for a variety of submicron particles, including core-shell synthesis, layer-by-layer techniques, multi-block polymer emulsifications, and surface modifications.<sup>17,18,19,20,21,22</sup> Silica particles are one of the most extensively studied particles as they can be easily modified with organic functionalities. However, these silica particles usually have only a single type of functional group. Therefore, multiple sequential modification steps have to be employed in order to attach multitasking components such as imaging components, drugs and targeting moieties. For wider biological applications, the development of a simple and efficient preparation method for particles with various types of functionalities is of great importance. Herein, a “ONE-POT” synthesis of multifunctional hybrid silica (MHS) particles of uniform size and morphology with homogeneously distributed multiple functional moieties is reported. The MHS particles can be tagged with various multitasking components for detection, selective binding, and separation using different functionalities, in an easier and more flexible way than those particles with a single functional group. Moreover, each functional moiety can be further modified with additional organic or inorganic groups. We demonstrate here following abilities of MHS particles: 1) expansion of surface functionality through the reaction with organic or inorganic compounds; 2) multicolor imaging by surface-conjugation with multiple dyes; 3) controlled assembly of 3-dimensional aggregates from two different types of MHS particles; 4) multiplex tasking through dye-tagging and selective binding to a patterned surface.

MHS particles with various types of functional groups were prepared by one-pot synthesis made of a two-step (acid-base) sol-gel process. In acidic conditions, the hydrolysis of mixture of various silane monomers occurred simultaneously and emulsion droplets with homogeneous silane mixtures were formed. Co-condensation between the hydrolyzed silane mixtures in the emulsion droplets occurred in basic conditions. The mixture of silane monomers

for MHS particles include phenyltrialkoxysilane (PTMS), vinyltrialkoxysilane (VTMS), or mercaptopropyltrialkoxysilane (MPTMS) as the major building blocks and (3-trimethoxysilyl) propylmethacrylate, aminopropyltrimethoxysilane (APTMS), or other functional organo silanes as the minor functional components. For most of the MHS particles, we found that the relative concentration of major silane monomers compared to minor functional silanes should be greater than 70% of the total silane concentration to obtain monodisperse, spherical particles. By changing the reaction conditions such as silane concentrations, temperature, and reaction time, the size of MHS particles could be controlled from 200 nm to 1.4  $\mu$  m. Structural drawings and SEM images of typical di- and tri-functional hybrid silica particles are shown in Figure 1 indicating that all of the MHS particles exhibit monodispersity and spherical morphologies. The existence of the functional groups was confirmed by IR and NMR spectra. IR spectra of MHS particles exhibited characteristic stretching vibrational absorption peaks for corresponding functional groups. For the vinyl, mercapto, and acryl tri-functional hybrid particles,  $\nu$  (C=C),  $\nu$  (S-H), and  $\nu$  (C=O) stretching vibrational absorption peaks from each functional group were observed at 3100, 2300, and 1500 $\text{cm}^{-1}$ , respectively. Solid-state  $^{13}\text{C}$  NMR analysis also showed typical carbon resonance peaks for corresponding functional groups. For phenyl and vinyl based MHS particles, strong absorption peaks near 130 ppm were observed from unsaturated carbons, while aliphatic carbon peaks between 0-50 ppm were observed for mercapto-based MHS particles. The solid-state  $^{29}\text{Si}$  NMR spectrum show two peaks at 78.1 and 69.4 ppm, indicating that the main structure of MHS particles consisted of  $\text{T}^2$  and  $\text{T}^3$  units, similar to those in PTMS and PTMS/VTMS composite particles.<sup>23,24,25</sup> However, the relative amount of  $\text{T}^2$  units was reduced for thiol- and vinyl- based MHS particles compared to those in phenyl-based MHS particles. Thermal behaviors of MHS particles were investigated using thermo-gravimetric analysis (TGA). While decomposition was observed between 300 ~ 700  $^{\circ}\text{C}$  for most of MHS particles, the spherical morphologies of MHS particles were maintained even after heat treatment at 800  $^{\circ}\text{C}$ . The list of synthesized MHS particles and characterization results are included in Supporting Information 1 (SEM, IR and NMR spectra) and 2 (TGA).

Chemical reactions of surface functional groups on MHS particles with various organic and inorganic materials, including fluorescent dyes, were performed to endow additional or modified functionalities to the particle surfaces, as shown in Supporting Information 3. Amine functional groups on the surface of MHS particles could be converted to carboxyl groups by reaction with glutaric anhydrides.<sup>26</sup> The formation of carboxyl groups on the surface of MHS particles was confirmed by IR and solid state  $^{13}\text{C}$  NMR spectra (Supporting Information 4). Hydrophobic phenyl based MHS particles could be transformed to hydrophilic groups by treatment with nitric acid, which converts surface phenyl groups to nitro-phenyl groups through the Friedel-Craft addition reaction.<sup>27</sup> Surface modification of MHS particles with inorganic materials such as  $\text{AgNO}_3$  produced metallic hybrid composite particles. Mercapto-

functionalized MHS particles were treated with an aqueous  $\text{AgNO}_3$  solution, converting the S-H functionality to  $\text{S}^-$ - $\text{Ag}^+$ . The resulting MHS particles were reduced with hydrazine to produce Ag nanoparticles on the surface of MHS particles after removing physically adsorbed Ag species through filtration and washing. The SEM image of MHS particles with  $\text{S}^-$ - $\text{Ag}^+$  functionalities did not reveal any noticeable change in surface morphology compared to untreated particles, while nanosized Ag particles were observed on the surface of reduced MHS particles. The XRD spectrum of Ag nanoparticle-doped MHS particles confirmed the formation of metallic silver on the surface of the MHS particles (Supporting Information 5).

To investigate the distribution of surface functional groups and multicolor imaging capability of MHS particles, selective tagging reactions with fluorescent dye molecules were performed. MHS particles containing phenyl, amine and mercapto groups were first treated with an excess amount of fluoresceamine dye to cover all the amine functionalities on the surface of MHS particles, and then further reacted with Rhodamine B isothiocyanate through surface mercapto groups.<sup>28,29</sup> Figure 2 shows confocal fluorescence spectra and the excitation/emission spectra of dye-tagged phenyl-based tri-functional particles with amine and mercapto functionalities. MHS particles tagged with fluoresceamine only exhibited a blue fluorescent emission peak at 475 nm by excitation at 390 nm, while those tagged with two different dyes showed two fluorescent emission peaks (blue and red) at 475 and 570 nm using excitation wavelengths of 390 and 540 nm, respectively. This indicates that the surfaces of MHS particles were covered by different functional groups, demonstrating better and simpler control over the amount of multiple tagging on the nanoparticle surface than those with only a single type of functional group. Therefore, these multi-dye tagged MHS particles have excellent potential for applications to bio- and nanotechnologies such as multiplex signaling and combinatorial chemistry.

The ability of MHS particles to act as potential building blocks was demonstrated by 3-dimensional self-assembled structures constructed through selective binding between two MHS particles with different surface functional groups. The phenyl-epoxy-carboxylic acid functionalized MHS particles of 1.4  $\mu\text{m}$  in diameter and phenyl-epoxy-amine functionalized MHS particles of 200 nm or 600nm in diameter were dispersed together in an aqueous solution to form a self-assembled structure which was confirmed by SEM (Figure 3 and Supporting material 6). The formation of self-assembled structures is probably due to coulombic interaction between negatively charged MHS particles with carboxylate groups and positively charged MHS particles with ammonium functional groups, which were produced by the protonation and deprotonation of corresponding functional groups on each MHS particle. The formation of self-assembled structures was also confirmed by the confocal microscopy, using the phenyl-mercapto-carboxylic acid functionalized MHS particles with the size of 1.4  $\mu\text{m}$  and the phenyl-amine-epoxy functionalized MHS particles with the size of 200 nm. The larger MHS particles were tagged with Rhodamine B isothiocyanate and the smaller ones were partially coordinated with fluoresceamine. The confocal fluorescent images of the self-assembled MHS particles

exhibited red fluorescence from the center of the assembled structure and blue fluorescence mostly from the periphery when excited at 390 and 540 nm, respectively. This result suggests the potential use of the MHS particles for multicolor imaging. These self-assembled MHS particles maintained their morphology even upon severe stirring and sonication and under acidic condition ( $\text{pH} < 2$ ). However, the assembled structure of the MHS particles was easily disintegrated by addition of a base (Supporting Information 6). In the presence of a strong base, the positively charged ammonium group transforms into negatively charged amide group. This induces repulsion between two types of MHS particles, eventually destroying the assembled structure. In an acidic condition, negatively charged carboxylate groups are converted into neutral carboxylic acids, producing a mixture of neutral and positively charged MHS particles, resulting in no coulombic attraction/repulsion between the two types of particles. However, the hydrogen-bonding between carboxyl acid and ammonium functionalities may have prevented the disintegration of the assembled MHS particles. The formation of self-assembled structures of MHS particles and their stability in acidic condition indicate that these particles can be useful for bio-labeling and intracellular or in vivo applications as they can contribute signal amplification consistently. The instability in basic condition may be also useful for the laboratory analysis of specific analytes, enabling detection, signal amplification, separation and then disassembling for further analysis.

As an example to show multiplex tasking ability of MHS particles, the assembly of dye-doped MHS particles on the patterned glass surface was investigated. MHS particles with thiol and carboxylic acid functional groups were first reacted with Rhodamine B to load fluorescence dye. The dye-doped MHS particles were then attached to the glass surface with patterned lines created by treating the glass surface with amino and octadecyl silanes, which prepared with slight modification of the method described earlier.<sup>30</sup> The acid-base reaction between amine groups on the glass surface and carboxylic groups on the surface of MHS particles leads to the formation of the patterned assembly of MHS particles on the glass, as shown in Figure 4.

In conclusion, multifunctional hybrid silica particles were prepared by facile one-pot synthesis. The types and numbers of surface functional moieties can be controlled with great flexibility and the size of the particles can also be controlled between 200 nm and 1.4  $\mu\text{m}$  with a narrow size distribution. The surface can be further modified in order to introduce additional organic functional groups or to carry various dyes or molecules such as fluorescent image contrast agents, targeting ligands or drugs, making them useful for various biomedical applications such as multicolor imaging. The self assembly behavior of MHS particles with other MHS particles or on a patterned glass surface demonstrates the ability of MHS particles to act as potential building blocks for next generation multitasking nano-devices which can detect multiple analytes and amplify the signals.

## METHODS

### Typical Synthesis of MHS particles.

#### 1) Vinyl and methacrylate based di-functional particles

One hundred and fifty milliliters of de-ionized water was placed in a 250 mL three-neck round bottom flask while maintaining a temperature of 40 °C, and 0.2 mL of nitric acid (60 wt%, 2.67 mmole) was added with stirring at 300 rpm. Five milliliters of VTMS (32.06 mmole) was added to an acidic aqueous solution, followed by the addition of (3-trimethoxysilyl) propylmethacrylate, (0.5 mL, 2.06 mmole). After stirring for one minute, 40 mL of ammonium hydroxide (30 wt%, 306 mmole) was poured into the reaction mixture to induce condensation. The solution became turbid, indicating the formation of colloidal particles. After stirring for 4 hours, the suspension was filtered through a membrane (Osmonics, pore size of 400 nm) and the filtered product was washed with 10 mL of de-ionized water and 10 mL of ethanol two times. The solution was dried under vacuum and 2.7 g of vinyl and methacrylate based tri-functional particles were obtained.

#### 2) Phenyl-amine-mercapto based tri-functional particles

A 250 mL Erlenmeyer flask maintained at 40 °C was charged with 150 mL of deionized water and 0.2 mL of nitric acid (2.67 mmol, 60 wt%) was added while stirring at 300 rpm. Five milliliters of PTMS (24.89 mmole) was first injected to an acidic aqueous solution, followed by the addition of 0.5 mL APTMS (2.75 mmole) and 0.5 ml MPTMS (2.57 mmol). The resulting solution was stirred for 1 minute to allow hydrolysis followed by the addition of 40 mL of ammonium hydroxide (305 mmole, 30 wt%) to induce condensation. The mixture became turbid and was stirred for an additional 4 hours. The resulting multifunctional hybrid silica particles were filtered through a membrane and washed with 10 mL of deionized water and 10 mL of ethanol several times. The filtered particles were dried under vacuum and 2.8 g of phenyl-amine-thiol based tri-functional particles were obtained.

#### **Surface modification reactions of MHS particles.**

##### 1) Organic modification with glutaric anhydride.

Vinyl based MHS particles with mercapto and amine functional groups (0.2 g) were placed in a 250 mL three-neck round bottom flask and the reaction vessel was heated to 85 °C. Glutaric anhydride (0.5 g, 4.16 mmole) was added to the MHS particles and the resulting mixture was stirred for 3 hours. One hundred milliliters of de-ionized water and 100 mL of ethanol were poured into the reaction flask for dilution and dispersion of the resulting mixture. The suspension was stirred for 5 minutes and filtered through a membrane. The solution was dried in air and 0.2 g of surface modified MHS particles were obtained.

##### 2) Hydrophilic modification with nitric acid.

Three grams of phenyl-based MHS particles with mercapto and amine functional groups were placed in a round bottom flask in an ice bath and 6 ml (60%, 80 mmol) of nitric acid was added to the flask. A solution containing 0.5 ml (98%, 10 mmol) of sulfuric acid was added drop-wise and the reaction mixture was stirred for 2 hours. For dilution, 500 ml of cold water was poured into the reaction flask and after 10 min, the resulting particles were

collected with a membrane filter. The particles were dried in a vacuum oven at 60° C for 4 hours and 2.8 g of surface modified MHS were obtained.

3) Inorganic modification with Ag(NO)<sub>3</sub> and the reduction to Ag nanoparticles with hydrazine.

MHS particles (0.2 g) obtained from (1) were dispersed in 100 mL of ethanol. AgNO<sub>3</sub> (0.2g, 1.18 mmole) in 10 mL of de-ionized water was added and the resulting solution was stirred for 4 hours. To remove unreacted AgNO<sub>3</sub>, the reaction mixture was filtered and the obtained particles were washed with 10 mL of de-ionized water at least three times. The resulting particles were re-dispersed in 100 mL of ethanol and 0.2 mL of hydrazine monohydrate (4.19 mmole) was added to reduce the Ag<sup>+</sup> ions attached to thiol functional groups. The Ag nanoparticle doped MHS particles were filtered through a membrane filter and washed with de-ionized water and ethanol several times. The particles were air-dried and 0.2 g of the composite MHS particles were obtained.

3) Dual dye-tagging reaction of MHS particles.

Phenyl-based MHS particles with amine and mercapto functional groups (0.05 g) were dispersed in 50 mL of ethanol and 50 mL of de-ionized water was added. Fluoresecamine (Strem, 0.05g, 0.02 mmole) in 20 mL of an ethanol/water/dichloromethane (V/V/V=1/1/1) mixture was added and the reaction mixture was stirred for 4 hours. The resulting suspension was filtered, washed and dried in air. Fluoresecamine tagged MHS particles were re-dispersed in 50 mL of ethanol and 50 mL of de-ionized water was added. Rhodamine B isothiocyanate (0.05 g, 0.01 mmole) was added to the solution and stirred for 4 hours. The solution was filtered and the particles obtained were washed with de-ionized water and ethanol several times. The particles were air-dried and 0.03 g of dual dye tagged MHS particles were obtained.

#### Reference

1. G. R. Reddy, M. Bhojani, P. M. McConville, J. Moody, B. A. Moffat, D. E. Hall, G. Kim, Y. Koo, M. J. Woollscroft, J. V. Sugai, T. D. Johnson, M. Philbert, R. Kopelman, A. Rehemtulla, & B. D. Rss. Vascular targeted nanoparticles for imaging and treatment of brain tumors. *Clin. Cancer Res.* **12**(22), 6677-6686 (2006).
2. Moritz F. Kircher, Umar Mahmood, Raymond S. King, Ralph Weissleder, & Lee Josephson. A Multimodal Nanoparticle for Preoperative Magnetic Resonance Imaging and Intraoperative Optical Brain Tumor Delineation. *Cancer Res.* **63**, 8122– 8125 (2003).
3. Istvan J. Majoros, Thommey P. Thomas, Chandan B. Mehta, & James R. Baker Jr. Poly(amidoamine) Dendrimer-Based Multifunctional Engineered Nanodevice for Cancer Therapy. *J. Med. Chem.* **48**, 5892-5899 (2005).
4. Omid C. Farokhzad, Jianjun Cheng, Benjamin A. Teply, Ines Sherifi, Sangyong Jon, Philip W. Kantoff, Jerome P. Richie, & Robert Langer. Targeted nanoparticle-aptamer bioconjugates for cancer chemotherapy in vivo. *PNAS* **103**(16), 6315-6320 (2006).
5. Dhruva J. Bharali, Iлона Klejbor, Ewa K. Stachowiak, Purnendu Dutta, Indrajit Roy, Navjot Kaur, Earl J. Bergey, Paras N. Prasad, & Michal K. Stachowiak. Organically modified silica nanoparticles: A nonviral vector for in vivo gene delivery and expression in the brain. *PNAS* **102**(32), 11539-11544 (2005).
6. Lin Wang, Wenjun Zhao, Meghan B. O' Donoghue, & Weihong Tan. Fluorescent Nanoparticles for Multiplexed Bacteria Monitoring. *Bioconjugate Chem.* **18**, 297-301 (2007).
7. William J. Rieter, Jason S. Kim, Kathryn M.L. Taylor, Hongyu An, Weili Lin, Teresa Tarrant, & Wenbin Lin. Hybrid Silica Nanoparticles for Multimodal Imaging. *Angew. Chem. Int. Ed.* **46**, 3680-3682 (2007).
8. Eugenii Katz & Itamar Willner. Integrated Nanoparticle-Biomolecule Hybrid Systems: Synthesis, Properties, and Applications. *Angew. Chem. Int. Ed.* **43**, 6042-6108 (2004).

9. Claire S. Peyratout & Lars Dähne. Tailor-made Polyelectrolyte Microcapsules: From Multilayers to Smart Containers. *Angew. Chem. Int. Ed.* **43**, 3762-3783 (2004).
10. Ana B. Descalzo, Ramón Martínez-Mañez, Félix Sancenón, Katrin Hoffmann, & Knut Rurack. The Supramolecular Chemistry of Organic-Inorganic Hybrid Materials. *Angew. Chem. Int. Ed.* **45**, 5924-5948 (2006).
11. Joseph M. Slocik, Felicia Tam, Naomi J. Halas, & Rajesh R. Naik. Peptide-Assembled Optically Responsive Nanoparticle Complexes. *Nano Lett.* **7**(4) 1054-1058 (2007).
12. Lin Wang, Chaoyong Yang, & Weihong T. Dual-Luminophore-Doped Silica Nanoparticles for Multiplexed Signaling. *Nano Lett.* **5**(1) 37-43 (2005).
13. Daniel C. Pregibon, Mehmet Toner, & Patrick S. Doyle. Multifunctional Encoded Particles for High-Throughput Biomolecule Analysis. *Science* **315** 1393-1396 (2007).
14. Maria Arduini, Silvia Marcuz, Mariachiara Montoli, Enrico Rampazzo, Fabrizio Mancin, Silvia Gross, Lidia Armelao, Paolo Tecilla, & Umberto Tonellato. Turning Fluorescent Dyes into Cu(II) Nanosensors. *Langmuir* **21**, 9314-9321 (2005).
15. Swadeshmukul Santra, Peng Zhang, Kemin Wang, Rovelyn Tapeç, & Weihong Tan. Conjugation of Biomolecules with Luminophore-Doped Silica Nanoparticles for Photostable Biomarkers. *Anal. Chem.* **73**, 4988-4993 (2001).
16. Robert Wilson, Andrew R. Cossins, & David G. Spiller. Encoded Microcarriers for High-Throughput Multiplex Detection. *Angew. Chem. Int. Ed.* **45**, 6104-6117 (2006).
17. Mark Green. The Organometallic Synthesis of Bifunctional Core/Shell Nanoparticles. *Small* **1**(7), 684-686 (2005).
18. Frank Caruso. Nanoengineering of Particle Surfaces. *Adv. Mater.* **13**(1), 11-22 (2001).
19. Kumares S. Soppimath, Li-Hong Liu, Wei Yang Seow, Shao-Qiong Liu, Ross Powell, Peggy Chan, & Yi Yan Yang. Multifunctional Core/Shell Nanoparticles Self-Assembled from pH-Induced Thermosensitive Polymers for Targeted Intracellular Anticancer Drug Delivery. *Adv. Funct. Mater.* **17**, 355-362 (2007).
20. Frank Hoffmann, Maximilian Cornelius, Jürgen Morell, & Michael Fröba. Silica-Based Mesoporous Organic-Inorganic Hybrid Materials. *Angew. Chem. Int. Ed.* **45**, 3216-3251 (2006).
21. Savka I. Stoeva, Fengwei Huo, Jae-Seung Lee, & Chad A. Mirkin. Three-Layer Composite Magnetic Nanoparticle Probes for DNA. *J. Am. Chem. Soc.* **127**, 15362-15363 (2005).
22. Dejin Li, Xia Sheng, & Bin Zhao. Environmentally Responsive "Hairy" Nanoparticles: Mixed Homopolymer Brushes on Silica Nanoparticles Synthesized by Living Radical Polymerization Techniques. *J. Am. Chem. Soc.* **127**, 6248-6256 (2005).
23. Hoe Jin Hah, Jung Soo Kim, Byung Jun Jeon, Sang Man Koo, & Yong Eun Lee. Simple preparation of monodisperse hollow silica particles without using Templates. *Chem. Commun.* 1712-1713 (2003).
24. Sang Man Koo, Hoe Jin Hah, Dong Il Han, & Chan Yoon Jung. A Self-template Method for Mesoporous Organosilica Particles. *Chem. Lett.* **36**(7), 906-907 (2007).
25. Qiangbin Wang, Yan Liu, & Hao Yan. Mechanism of a self-templating synthesis of monodispersed hollow silica nanospheres with tunable size and shell thickness. *Chem. Commun.* 2339-2341 (2007).
26. Suman Lata & Jacob Piehler. Stable and Functional Immobilization of Histidine-Tagged Proteins via Multivalent Chelator Headgroups on a Molecular Poly(ethylene glycol) Brush. *Anal. Chem.* **77**, 1096-1105 (2005).
27. Chan Yoon Jung, Hoe Jin Hah, Seung Hoon Han, Hong-Woo Park, & Sang Man Koo. Preparation of Tin Oxide-Based Metal Oxide Particles. *J. Sol-Gel Sci. Tech.* **33**, 81-85, (2005).
28. Eun Young Park, Jun Woo Kim, Dong June Ahn, & Jong-Man Kim. A Polydiacetylene Supramolecular System That Emits Red, Green, and Blue Fluorescence. *Macromolecular Rapid Communications* **28**, 171 (2007).
29. Chris R. Miller, Robert Vogel, Peter P. T. Surawski, Kevin S. Jack, Simon R. Corrie, & Matt Trau. Functionalized Organosilica Microspheres via a Novel Emulsion-Based Route. *Langmuir* **21**, 9733-9740 (2005).
30. Mi H. Park, Young J. Jang, Hyung M. Sung-Suh, & Myung M. Sung. Selective Atomic Layer Deposition of Titanium Oxide on Patterned Self-Assembled Monolayers Formed by Microcontact Printing. *Langmuir* **20**, 2257-2260 (2004).

**Supplementary Information** accompanies the paper on [www.nature.com/nature](http://www.nature.com/nature).

Acknowledgement. Authors would like to thank Prof. Myung M. Sung at Hanyang University for generous support on the preparation of patterned glass with aminosilanes and octadecylsilanes.

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## Figure Legends

Figure 1. Structures and SEM images of typical di- and tri-functional hybrid particles.

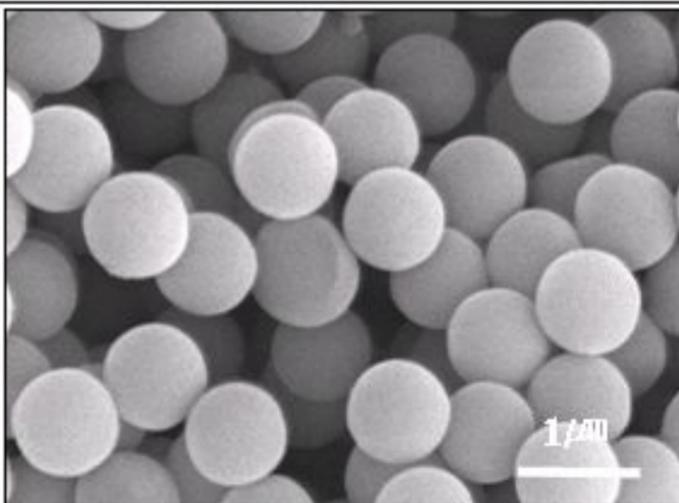
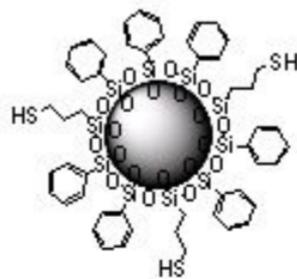
- a) Phenyl based di-functional hybrid silica particles
- b) Vinyl based di-functional hybrid silica particles
- c) Phenyl based tri-functional hybrid silica particles
- d) Mercapto based tri-functional hybrid silica particles

Figure 2. SEM images, photoluminescence spectrum, and confocal fluorescence images of dual dye tagged MHS particles

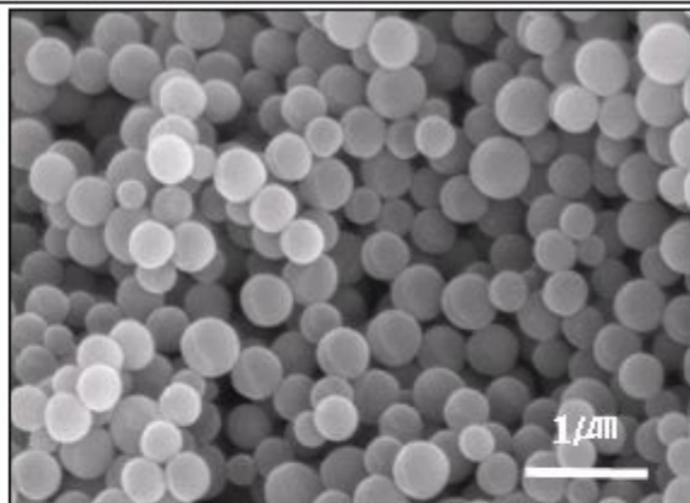
Figure 3. SEM and confocal fluorescence images of self assembled MHS particles with two different sizes

Figure 4. SEM and fluorescence images of dye doped MHS particles attached on the patterned surface.

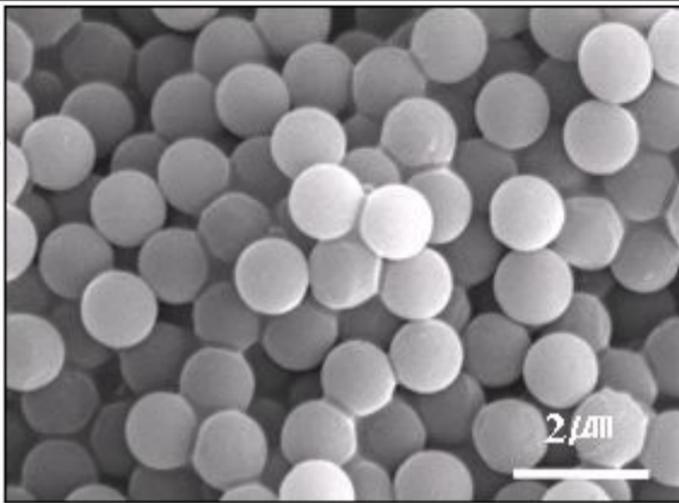
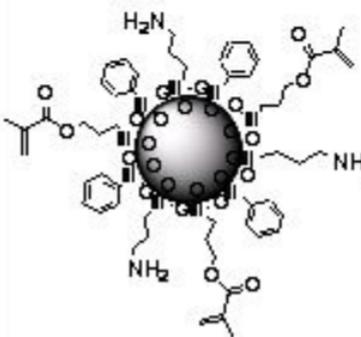
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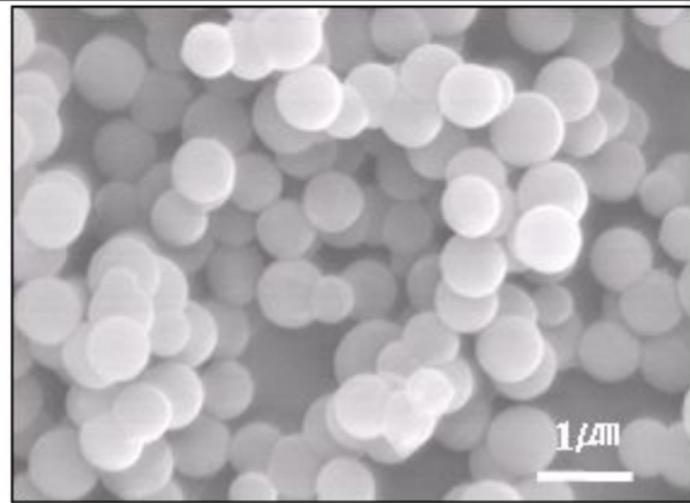
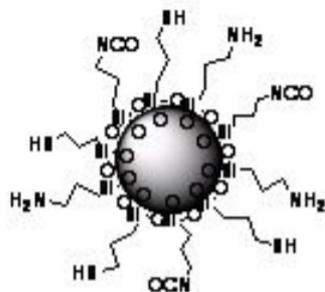
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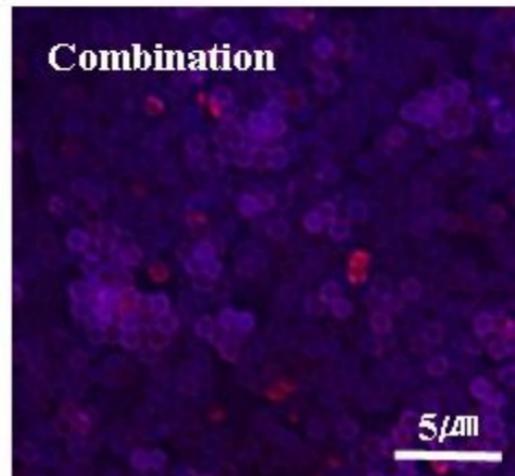
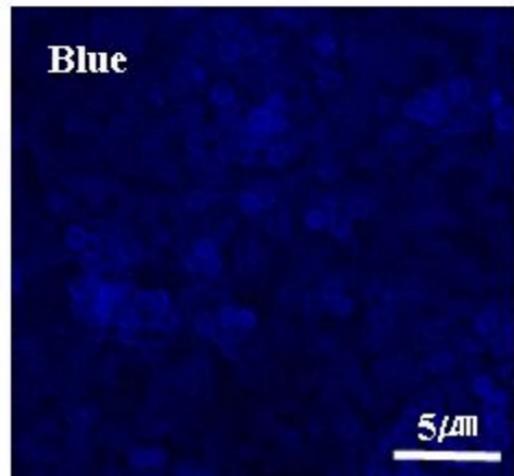
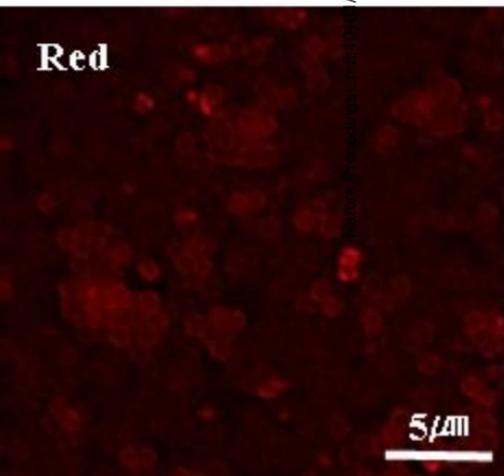
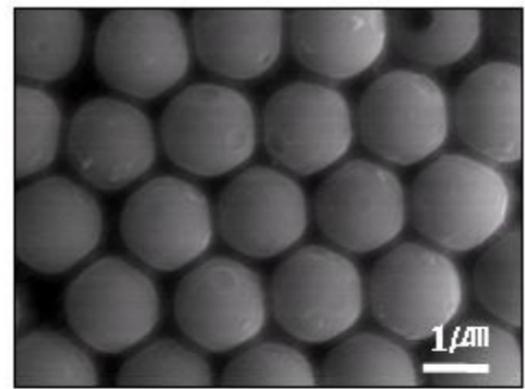
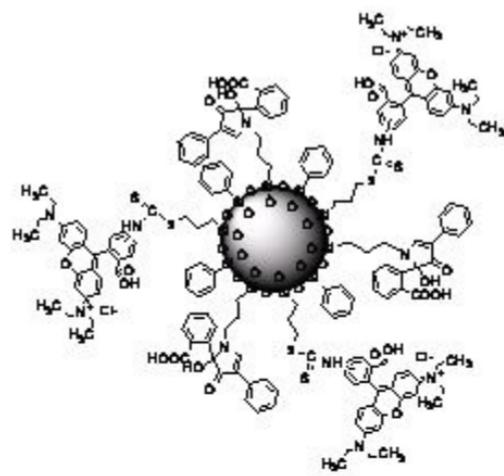
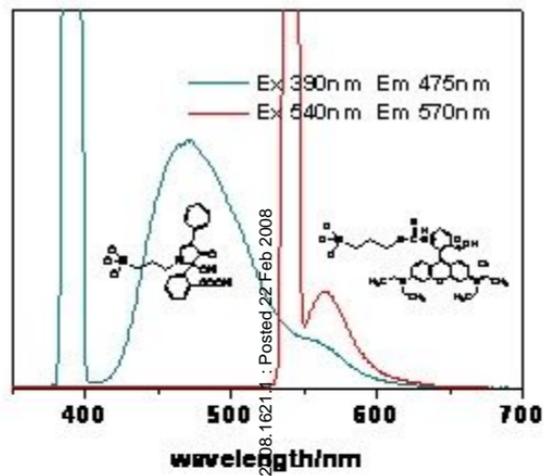


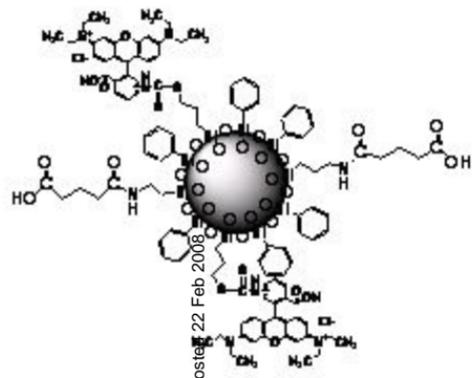
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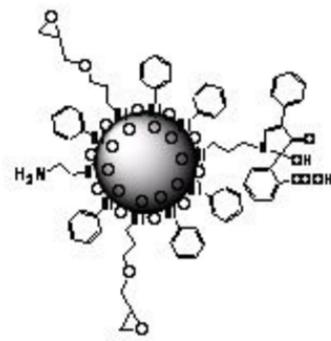
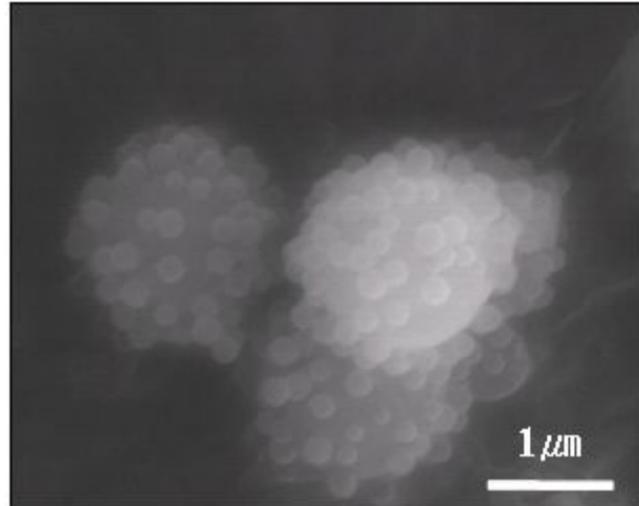
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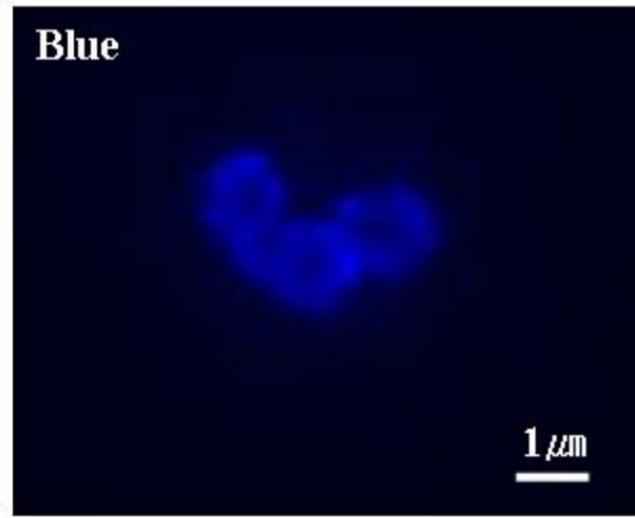
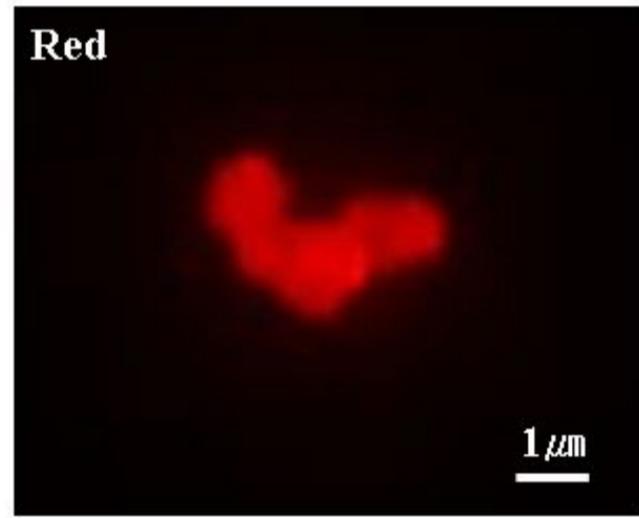
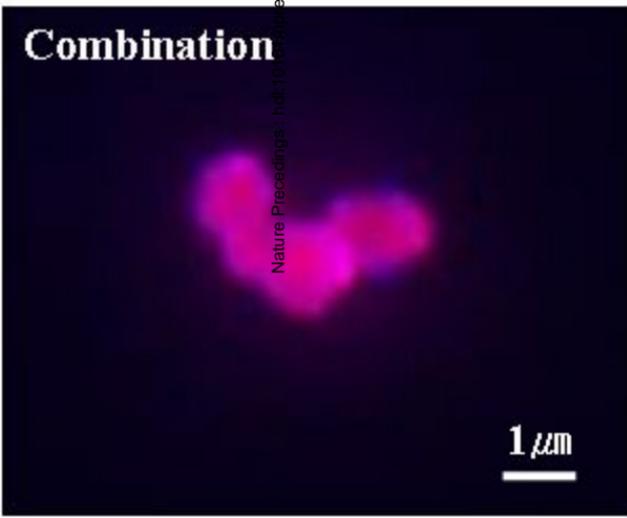




1.4  $\mu\text{m}$



200 nm



Nature Precedings : hdl:10101/npre.2008.1621.1 : Posted 22 Feb 2008

