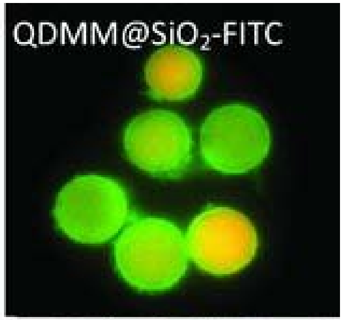
# Stable Encapsulation of Quantum Dot Barcodes with Silica Shells

**Shang-Hsiu Hu and Xiaohu Gao**

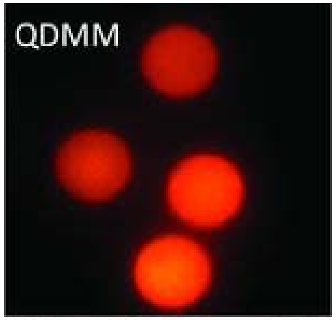


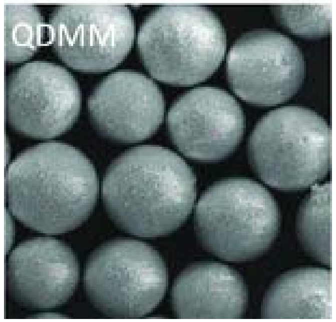


*Department of Bioengineering, University of Washington*
*Seattle, WA 98195 (USA)*

ADVANCED FUNCTIONAL MATERIALS

***Adv. Funct. Mater.*** 2010. ASAP



# Introduction

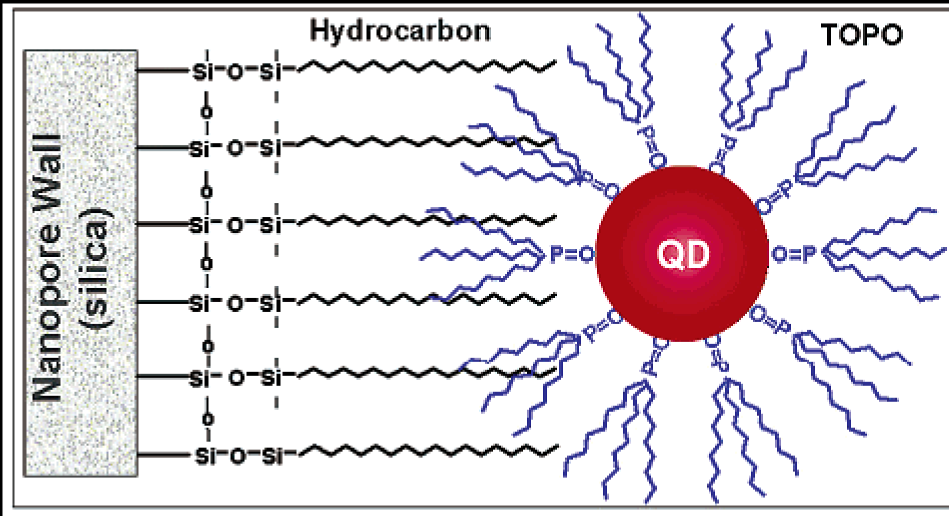
- **Optical barcoding technology based on fluorophore-encoded microbeads has become an important alternative to planar chip-based assays for highly multiplexed gene and protein screening.**

- **Quantum dot (QD)-doped microbeads with unique optical properties offer key advantages over microspheres labelled with traditional organic fluorophores, such as a dramatically increased number of possible barcodes, and single light source excitation of multiple colors.**

- **Microbead barcodes are produced and resolved based on spectral information.**

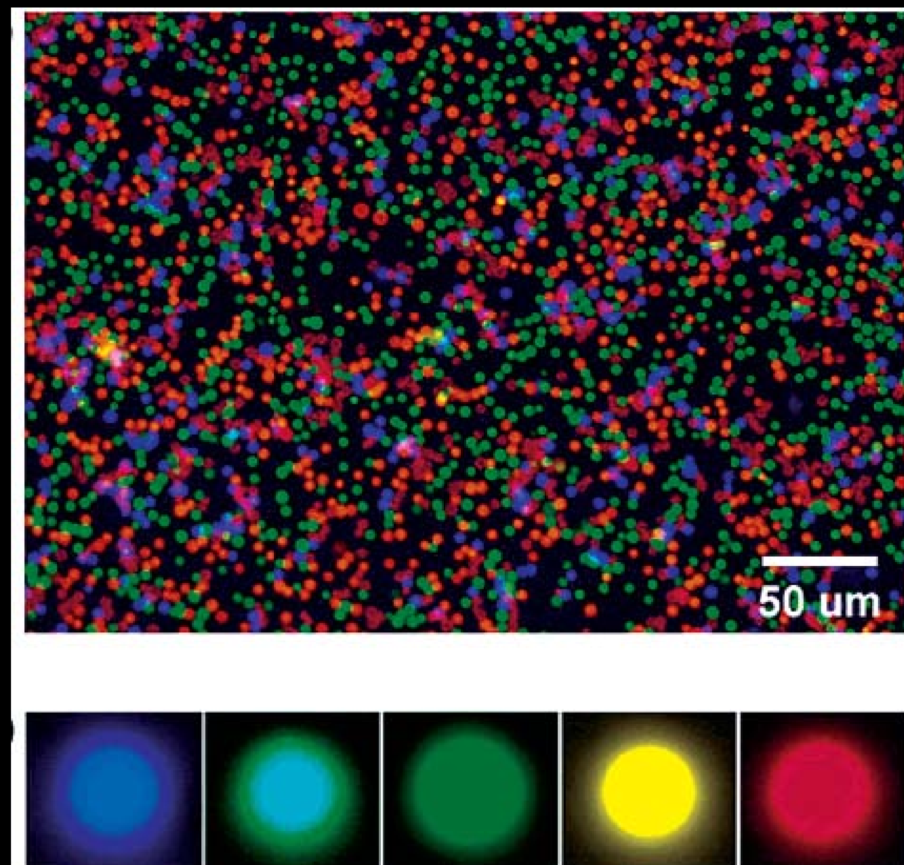
- **An improved brightness of about 50-100 times is observed in case of mesoporous materials due to their large surface area (150 $m^2$ per gram of beads) when compared to nonporous beads of the same size.**

- **Mesoporous silica have an ordered structure of nanopores in the size range of 2-100 nm diameter and it provides a stable and accessible surface for immobilizing a variety of functional molecules and small particles.**




Schematic representation of multivalent hydrophobic interactions between the surfactant (TOPO) molecules on the QD surface and the C-18 hydrocarbon molecules on the pore walls.

True-color fluorescence images of mesoporous silica beads (5 µm diameter, 32 nm pore size) doped with single-color quantum dots emitting light at 488 (blue), 520 (green), 550 (yellow), 580 (orange), or 610 nm (red). (a) Wide-field view of a large population of doped beads that were prepared in batches and then mixed, and (b) enlarged views of single monochromatic beads. Each bead contains up to 2 million dots of the same color.




X. Gao and S. Nie. J. Phys. Chem. B, Vol. 107, No. 42, 2003

## Limitations of the quantum dot-doped mesoporous microbeads (QDMMs) in optical barcoding technology

- ❖ Since the microbeads are mesoporous, potential leaching and chemical-induced degradation (e.g., spectral shift and intensity variation) of the embedded QDs are of major concern. i.e. QDs slowly diffuse out of the pores in organic solvents, resulting in a loss of barcode fluorescence intensity.

- ❖ Fluorescence fluctuation is often observed in different biological buffers or in reactions with bioconjugation crosslinking reagents.

- ❖ High hydrophobicity of the QDMMs leads to poor water solubility and lack of functional groups for bioconjugation.

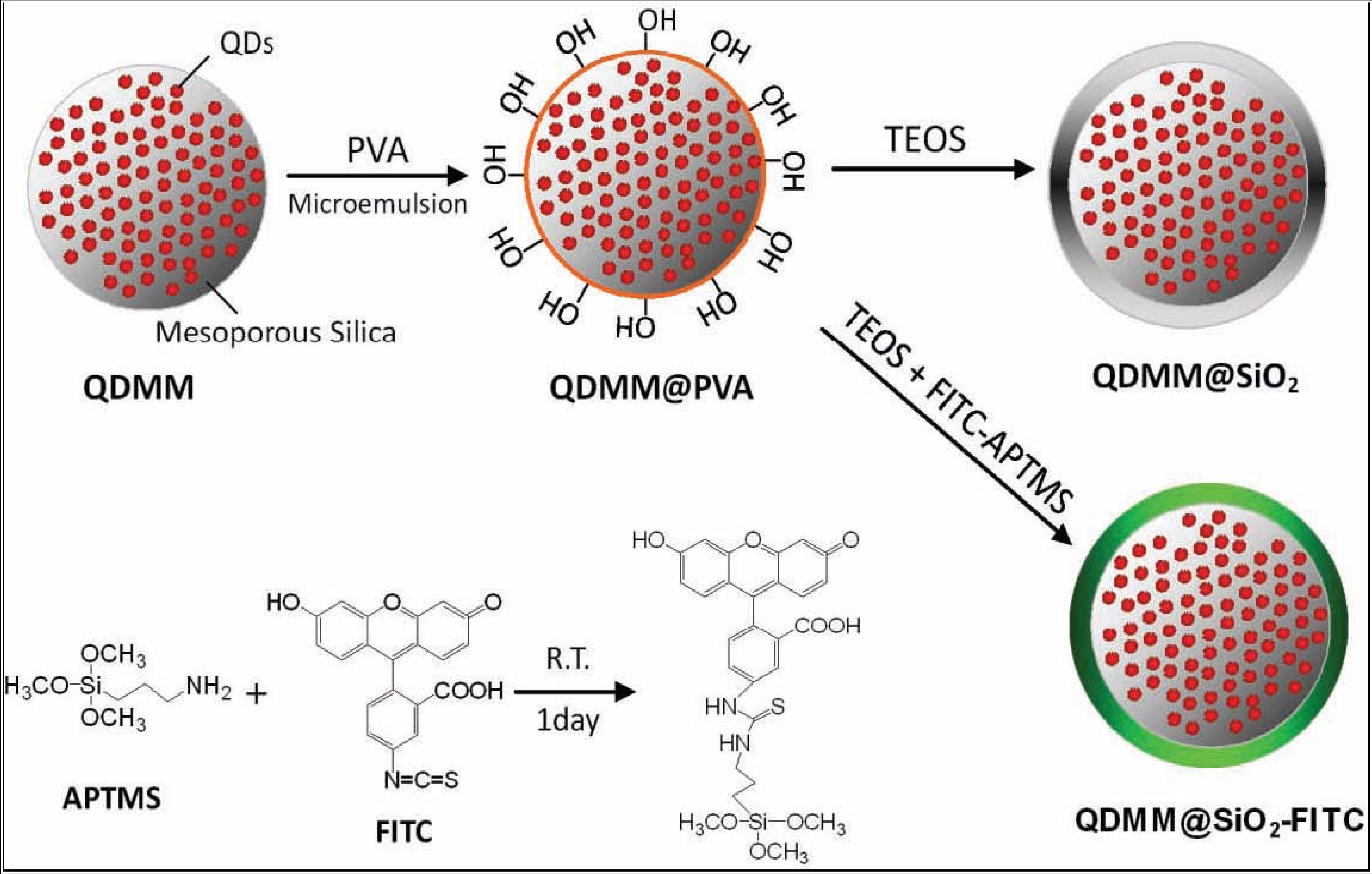
## In this paper

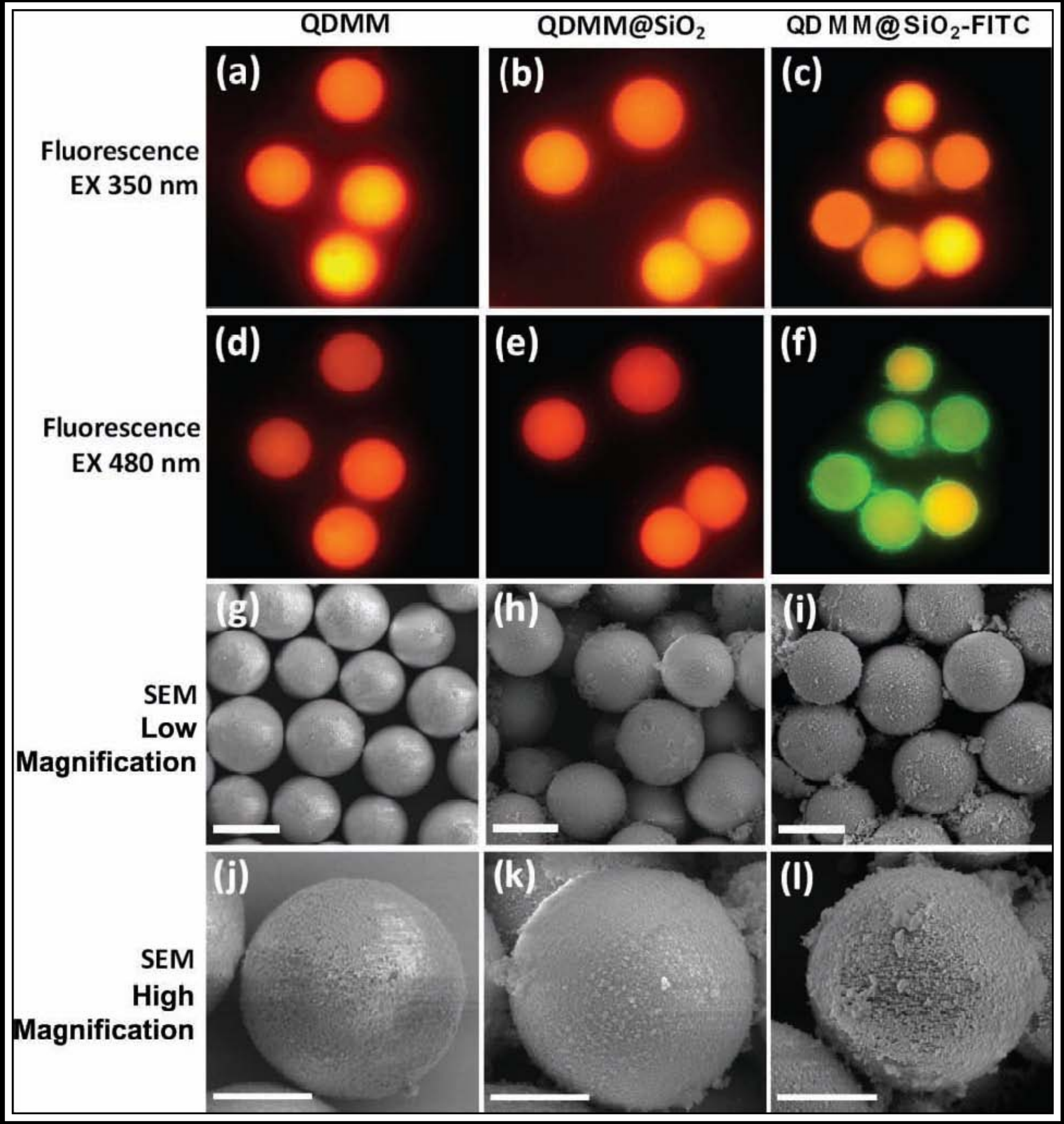
- ❖ A simple method for encapsulation of quantum dot-doped mesoporous microbeads with silica shells to stabilize the embedded QDs.

- ❖ Encapsulation using fluorescent silane precursors help in microscopic examination of the silane shells on the QDMMs.

- ❖ Studies were conducted to compare the physical and chemical stability of the QDMMs before and after silica coating.

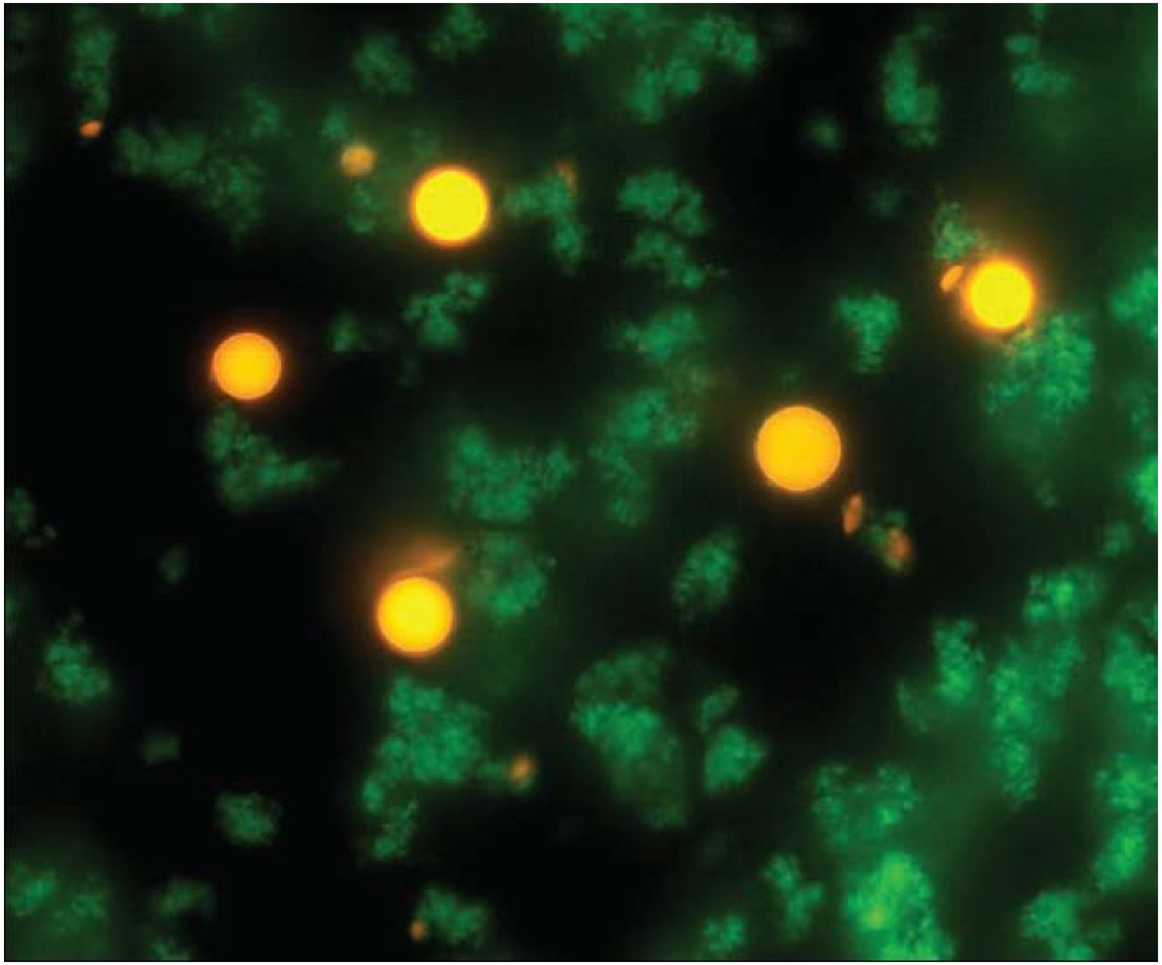
# Schematic illustration of key steps involved in silica encapsulation of QDMMs.






**Fluorescence and SEM images of QDMMs before and after silica encapsulation. Scale bars for (g-i) 5 μ m, (j-l) 2 μ m.**

# Role of PVA in silica shell formation



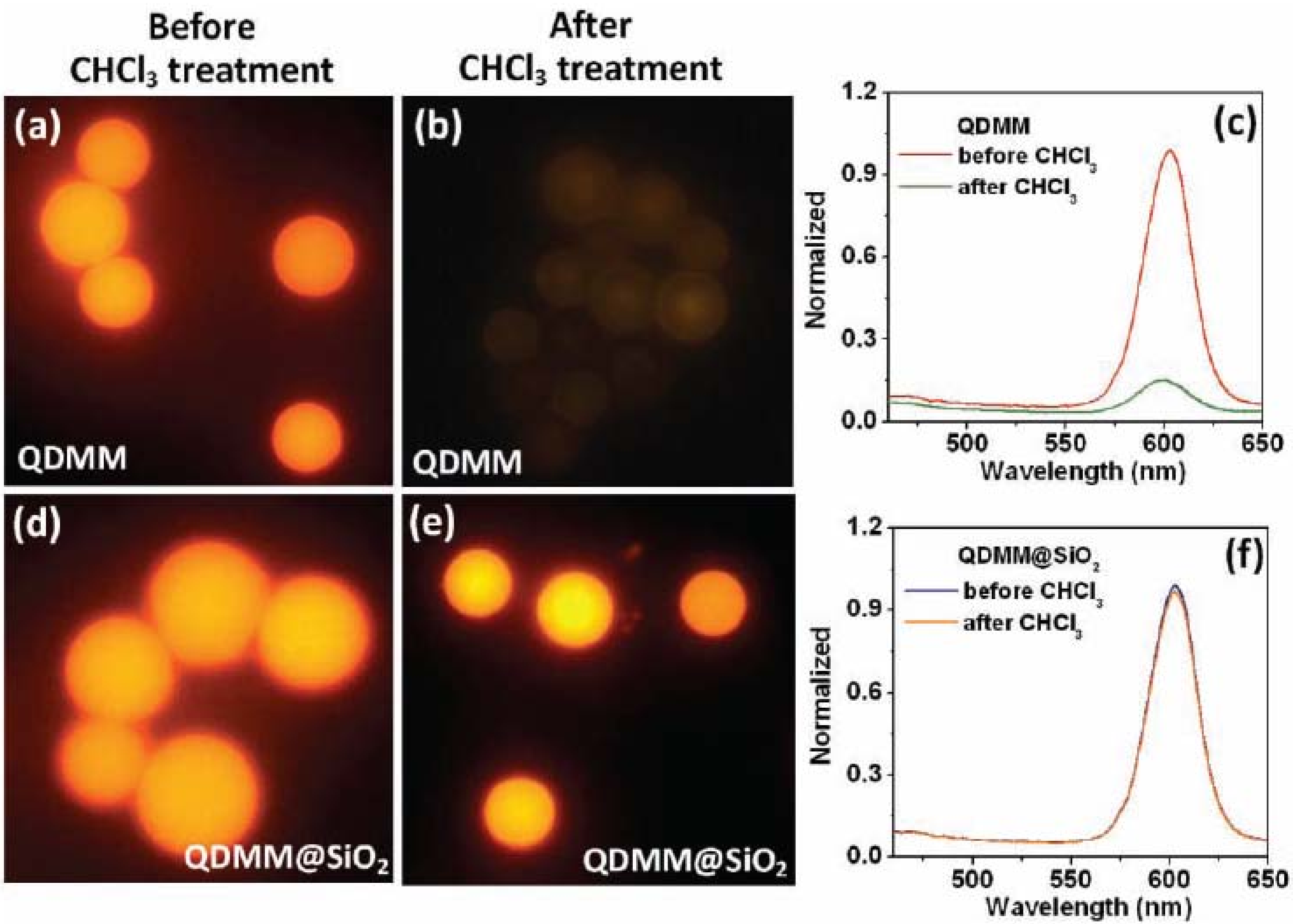
Fluorescence imaging of QDMM@$SiO_2$ synthesized without the microemulsion process.

Without PVA, the incompatibility between the hydrophobic hydrocarbons on microbead surface and hydrophilic silane compounds hinders nucleation of silane onto microbead surface.

They self-nucleate and condense into irregularly shaped aggregates (green aggregates in the background).

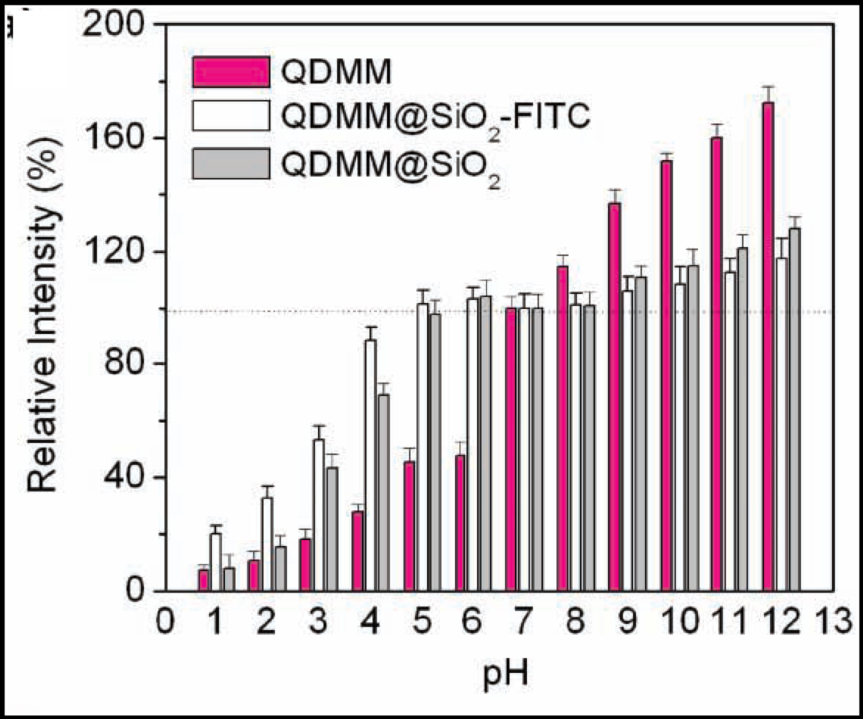
# Physical and chemical stability of the embedded QDs

## Solvent induced nanoparticle leaching - Study using Fluorescence imaging




**Fluorescence microscopic images of (a,b) QDMMs and (d,e) QDMM@$SiO_2$ before and after sonication for 2 min in chloroform, respectively.**

# Effect of pH on QDMMs and QDMM@$SiO_2$

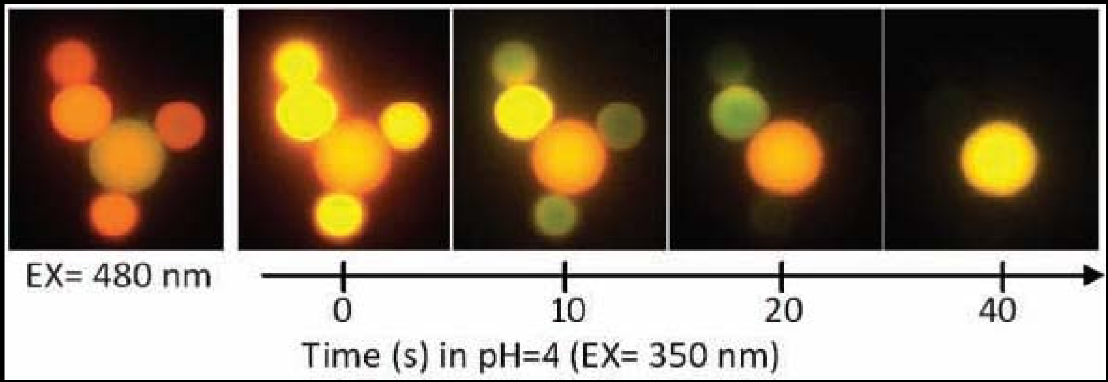



Microbeads were suspended in aqueous solutions with pH values ranging from 1 to 12 (fluorescence at neutral pH was set to 100%).

In absence of silica shell,

- Small changes in pH result in significant fluorescence fluctuation.
- Reducing pH from 7 to 5–6, fluorescence intensity of the embedded QDs is reduced by more than 50%.
- When the pH value is increased by 1 (to pH 8), the fluorescence signal is enhanced by approximately 20%.

In situ microscopic imaging of QDMM and QDMM@$SiO_2$-FITC at pH 4. The QDMMs are quenched significantly faster than the silica encapsulated microbeads.

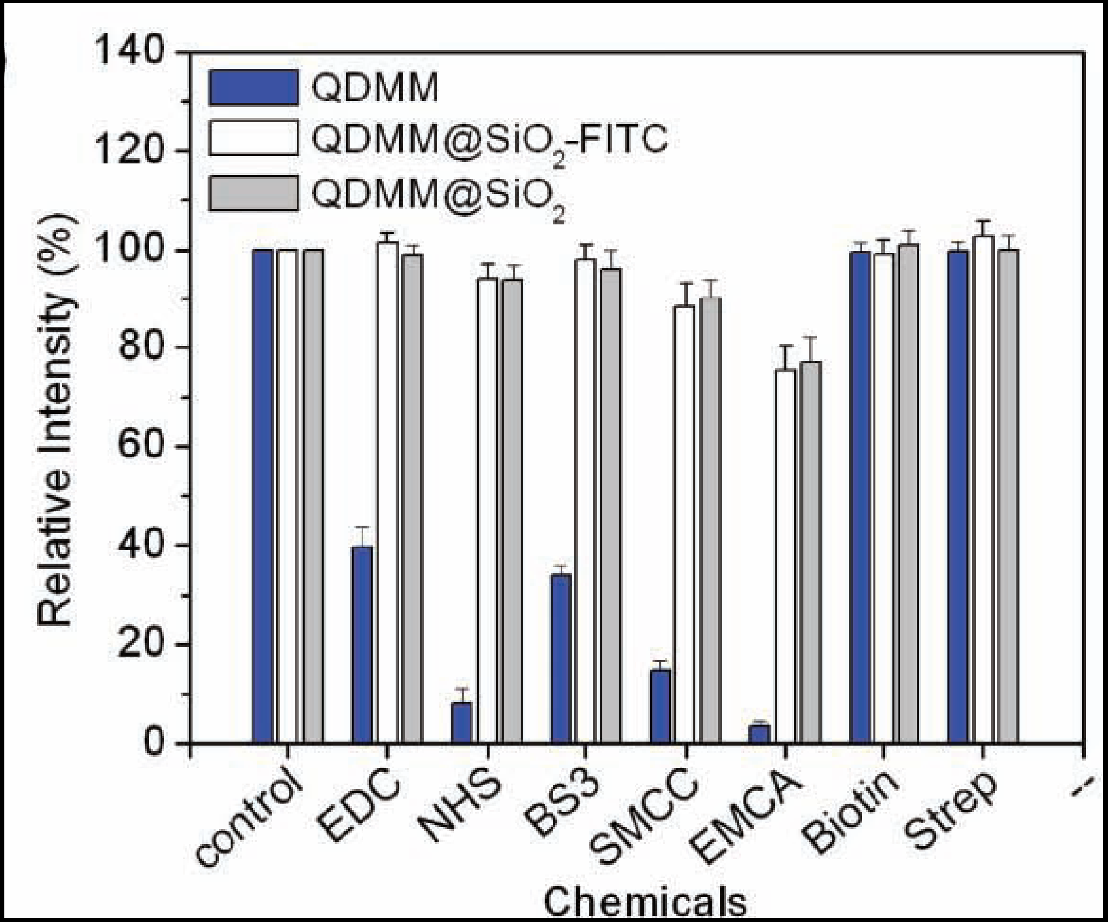


# Stability of the QDMMs and QDMM@$SiO_2$ against chemical treatment

- Cross linking reagents such as EDC, NHS and SMCC decreases the fluorescence of QDMM up to 90% after 24-hour incubation. In contrast that of QDMM@$SiO_2$ is nearly unaffected under the same condition.

- EMCA quenches the fluorescence of QDs in QDMM@$SiO_2$ by 20%. Thicker silica shell or decorating additional protection layers on top of the silica shell could possibly solve the problem.

**Chemical stability of QDMM, QDMM@$SiO_2$ and QDMM@$SiO_2$-FITC when treated with common bioconjugation reagents.**



# Summary

❖ A method for encapsulating QD barcodes with silica shells.

❖ To stabilize hydrophobic mesoporous microbeads in polar solvents, a microemulsion process was found highly effective in uniformly coating amphiphilic polymers onto the microbead surface. Subsequent condensation of silane compounds results in QDMM@$SiO_2$ with significantly improved chemical stability.

❖ This new method solved key problems in QD barcoding technology such as nanoparticle leaching. When QDMM@$SiO_2$ is suspended in organic solvents such as chloroform, in which hydrophobic QDs have excellent solubility, negligible nanoparticle leaching is detected.

❖ The barcode fluorescence remains stable when treated for 24 h with pH 5–8 aqueous solutions and a variety of common bioconjugation crosslinkers.

❖ Further optimization of this method such as fine tuning the silica shell thickness and linking silica surface with non-fouling polymers will allow clinical translation of the QD barcoding technology.

# Thanks