

LIGHT-SCATTERING STUDY OF POLYELECTROLYTE HOLLOW CAPSULES*

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Abstract Silver halide (AgX) microcrystal was used as template to synthesize hollow polyelectrolyte capsules. These hollow capsules were characterized by laser light scattering (LLS) used to measure the size of the capsules in solution. The ratio of hydrodynamic radius (R_h) from dynamic LLS to the radius of gyration (R_g) from static LLS is almost unity, revealing that the entities are hollow in solution. The results suggest that the LLS method can be regarded as a good complement to the confocal laser scanning microscopy (CLSM) method for the characterization of small hollow capsules, and it possesses the advantage of not needing fluorescence labeling.

Keywords Layer-by-layer (LbL) self-assembly, Hollow capsule, Silver halide, Laser light scattering

INTRODUCTION

The cell is the smallest structural unit of almost all organisms on earth. It is an entity that is enclosed by a semipermeable membrane. In recent years a large number of artificial cells has been synthesized by chemists. The wall of the hollow capsules can be semipermeable too. Different materials can be encapsulated or entrapped inside the capsules. Catalytic reactions may proceed inside these microreactors and it is possible to use them to simulate the biological reactions in a true cell.

Among the methods to prepare hollow capsules, the so-called LbL method has attracted more and more attention since it was first introduced into this field by Möhwald *et al.* in 1998^[1]. It is based on sequential electrostatic adsorption of oppositely charged species^[2], followed by removal of the template. This technique possesses the advantages of finely tuned polymer layer thickness and easily assembled multicomposite coatings. Very recently, we also introduced other LbL methods based on covalent and hydrogen bonding into this field^[3, 4].

The morphological characterization of these entities is an important basis for their preparation and application. Three methods are most frequently employed: transmission electron microscopy (TEM), atomic force microscopy (AFM) and confocal laser scanning microscopy (CLSM)^[5]. All of them have certain limitations. AFM and TEM can only reveal the morphology of the samples in a dried state. Usually it is difficult to identify whether the entity is a truly hollow one or not from the AFM or TEM images. For *in situ* observation in solutions, CLSM is the only method up to now to differentiate the hollow ones from the solid ones. Unfortunately like all optical microscopic methods, CLSM has a limiting lateral resolution in the order of 0.5 μm ^[6]. In order to visualize the nonfluorescent capsules, the samples should be labeled using proper

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fluorescent dyes^[5, 6].

In this paper laser light scattering (LLS) was used to study polyelectrolyte hollow capsules. LLS is an important technique to study polymer and colloid solutions^[7]. It is a powerful tool to measure the size of particles in a radius range from several nanometers to hundreds of nanometers, which is beyond the CLSM limit. Although LLS method can not give a real image of the particles, it can provide a lot of information about their morphology by combining the dynamic LLS and the static LLS. From dynamic LLS, one can obtain the hydrodynamic radius (R_h) of the particles. The radius of gyration (R_g) can be obtained from static LLS measurement. From the ratio of R_g and R_h , one can draw an image of the particles^[8].

EXPERIMENTAL

Synthesis of Polyelectrolyte Hollow Capsules

Silver halide photographic emulsion was used as template to fabricate the polyelectrolyte hollow capsules. The emulsion contains cubic AgX microcrystals with an average length of 0.12 μm and a narrow size distribution. The emulsion was first dissolved in hot water. After centrifugation the supernatant was discarded. The process was repeated at least 5 times to completely remove gelatin in the emulsion. Then 1 mL of 1 mg/mL poly(sodium styrenesulfonate) (PSS; M_w 70,000 g/mol) in 0.1 mol/L KBr aqueous solution was added. After the solution was allowed to stand for 10 min for adsorption, the excess PSS was removed by three repeated centrifugation/washing/redispersion cycles. Then 1 mL of 1 mg/mL poly(dimethyl diallylammonium bromide) (PDDA; M_w 70,000 g/mol) in 0.1 mol/L KBr aqueous solution was added. Excess PDDA was removed by the above same washing procedure. After the desired number of polyelectrolyte layers was achieved, the AgX core was dissolved by adding $\text{NH}_3 \cdot \text{H}_2\text{O}$. The resulting capsule solution was then dialyzed against water until the pH value reached about 7.

Characterization

TEM measurements were carried out on a JEM-100CX microscope (Japan Electronic Company). Specimens were prepared by dropping a suspension onto Formvar-coated copper grids. Laser light scattering was carried out on a commercial light scattering spectrometer (ALV/SP-125) equipped with an ALV-5000 multi- τ digital time correlator and an ADLAS DPY425II solid-state laser (output power: 400 mW at 532 nm).

RESULTS AND DISCUSSION

Many templates, such as melamine formaldehyde particles^[1, 5], polystyrene latex^[3, 9], gold nanoparticles^[10], and biological cells^[11], have been used for the fabrication of hollow polyelectrolyte capsules using the LbL method. To the best of our knowledge, the present study is the first one to use silver halide photographic emulsion as template, and we believe it is a versatile template for hollow polyelectrolyte capsule fabrication. First, the surface of silver halide microcrystals can bear positive or negative charges, which makes them suitable templates for electrostatic self-assembly. Secondly, it can be dissolved in a number of solvents, so the AgX core can be removed easily to obtain empty capsules. In addition, AgX microcrystals with various sizes and shapes and narrow size distribution are available. Using these templates, it is possible to synthesize hollow capsules with various sizes and shapes.

We first used TEM to examine the morphology of the dried capsules. As shown in Fig. 1, the capsule is round in shape, although the AgX template is cubic in shape. The result indicates that the polyelectrolyte capsule can adjust its shape to a certain extent. The diameter of the capsule is about 130 nm, which is in accordance with the size of the AgX template. In contrast to previous observations reported in the references^[1, 5], no clear fold and crease were found in our case. A possible explanation is that folding is difficult for the small capsules.

TEM observations can only reveal the morphology of dried samples. Although this information is valuable, the information about the capsule in solution will be more important. Unfortunately CLSM can't "see" these entities, because their small size is beyond the limit of this method, so we resort to LLS. To prepare samples for LLS experiments, the solution was filtered twice through a 0.5 μm Millipore filter to remove dust. In static LLS,

the angular dependence of excess absolute time-averaged scattered intensity, known as the excess Rayleigh ratio $R_{vv}(q)$, was measured. In the limit of a dilute solution, the reciprocal $R_{vv}(q)$ follows the relationship:

$$\frac{Hc}{R_{vv}(q)} \approx \frac{1}{M_w} \left(1 + \frac{\langle R_g^2 \rangle}{3} q^2 \right) + 2A_2c \quad (1)$$

where $H = 4\pi^2 n^2 (dn/dc)^2 / (N_A \lambda^4)$, $q = 4\pi n \lambda^{-1} \sin(\theta/2)$, n is the solvent refractive index, dn/dc is the specific refractive index increment, N_A is Avogadro's number, λ is the wavelength of the incident beam in vacuum, θ is the scattering angle, M_w is the weight-average molar mass, and A_2 is the second virial coefficient. This relationship reveals that there is a linear relationship between $Hc/R_{vv}(q)$ and q^2 . When the concentration of the capsule is small (5.35×10^{-5} mol/L, calculated in terms of styrenesulfonate monomer), the second term in Eq. (1) can be omitted. From the slope and the intercept of the $Hc/R_{vv}(q)$ - q^2 plot, we can get the radius of gyration of the capsule, which is the only concern about the system, without the exact value of dn/dc . Figure 2 shows the $Hc/R_{vv}(q)$ - q^2 plot, from which a radius of gyration of 72.6 nm was obtained.

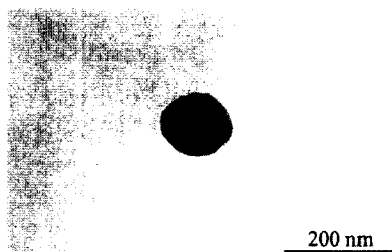


Fig. 1 TEM micrograph of a (PAH/PSS)₅ hollow capsule

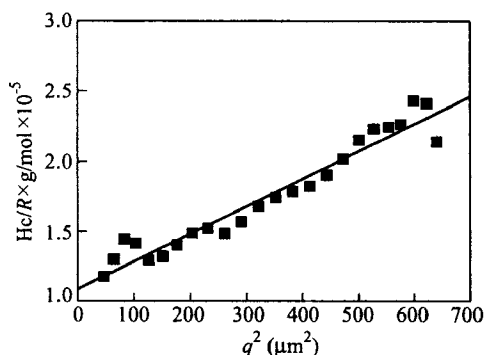


Fig. 2 $Hc/R_{vv}(q)$ - q^2 plot of (PAH/PSS)₅ hollow capsule in water, obtained at $T = 298$ K, $dn/dc = 0.1111$, $\theta = 30^\circ$ - 145°

Dynamic LLS measures the intensity-intensity time correlation function $G^{(2)}(t, q)$ in the self-beating mode, which has a relationship with the electric field time correlation function $g^{(1)}(t, q)$ as follows:

$$G^{(2)}(t, q) = A[1 + \beta |g^{(1)}(t, q)|^2] \quad (2)$$

where A is the baseline, β is a parameter depending on the coherence of the detection, and t is the delay time. $g^{(1)}(t, q)$ is further related to the characteristic linewidth (Γ) distribution $G(\Gamma)$ by

$$g^{(1)}(t, q) = \int_0^\infty G(\Gamma) \exp(-\Gamma t) d\Gamma \quad (3)$$

$G(\Gamma)$ can be calculated by a Laplace inversion of $g^{(1)}(t, q)$ using the CONTIN program. The first and second

moments of $G(\Gamma)$ are $\langle \Gamma \rangle = \int_0^\infty \Gamma G(\Gamma) d\Gamma$ and $\mu_2 = \int_0^\infty (\Gamma - \langle \Gamma \rangle)^2 G(\Gamma) d\Gamma$, respectively. The value of $\mu_2 / \langle \Gamma \rangle^2$ is a measure of polydispersity. If the relaxation is diffusive, Γ can be related to the translational diffusion coefficient D as

$$\Gamma/q^2 = D(1 + k_d C)(1 + f + \langle R_g^2 \rangle q^2) \tag{4}$$

where k_d is the diffusive second virial coefficient, R_g is the radius of gyration, and f is a dimensionless number related to hydrodynamic draining, internal motion, polydispersion, and solvent quality. If the translational diffusion coefficient D is known, the hydrodynamic radius R_h can be obtained from the Stokes-Einstein equation

$$R_h = \frac{k_B T}{6\pi \eta D} \tag{5}$$

where k_B is the Boltzmann constant, T is absolute temperature, and η is the solvent viscosity.

The apparent hydrodynamic radius $R_{h, app}$ distribution obtained from dynamic LLS at a temperature of 298 K and different scattering angles is shown in Fig. 3. For all cases only one large peak with a hydrodynamic radius $R_{h, app}$ of about 70 nm was observed. The size and the size distribution show a small angular dependence. The value of $R_{h, app}$ increases slightly from 67.8 nm to 74.5 nm when the scattering angle decreases from 90° to 30°. By extrapolating linearly to angle 0°, a hydrodynamic radius R_h of 76.4 nm was obtained.

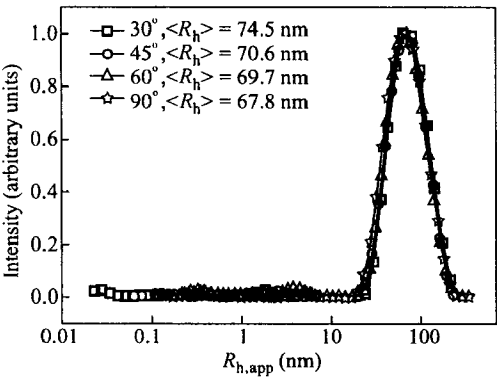


Fig. 3 Apparent hydrodynamic radius distributions of (PAH/PSS)₅ hollow capsule in water

Besides the large peak centered at about 70 nm, two small peaks were also observed at all scattering angles (Table 1). The leftmost peak falls out of the range of LLS measurement and will not be discussed further. The second peak, which centers from 1.25 nm to 4.71 nm, indicates that besides the larger capsules there are also many small particles present in the solution. These particles should be polyelectrolyte complex particles dispersed in the solution, their existence is a result of incomplete removal of the polyelectrolyte during the capsule preparation.

Table 1. The apparent hydrodynamic radius under different scattering angles

Scattering angle	$R_{h, app} 1$ (nm)	$R_{h, app} 2$ (nm)	$R_{h, app} 3$ (nm)
30°	0.0270	1.252	74.52
45°	0.1261	2.404	70.57
60°	0.3597	3.830	69.73
90°	0.3118	4.710	67.83

The size of the capsule measured from LLS (> 70 nm in radius) is larger than that from TEM (130 nm in diameter), indicating that the capsules in solution are swollen to a certain extent. The film materials are polyelectrolyte complex (PEC) in nature and it is well known that PEC can swell in water. From TEM

measurements we have known that the capsules in dried state are spherical in shape. If they are solid spheres, the ratio of radius of gyration to hydrodynamic radius, R_g/R_h , should be about 0.774. In contrast, the value of R_g/R_h is almost unity (0.95), suggesting the entities are hollow spherical shells.

In conclusion, the LLS method has been applied to the study of polyelectrolyte hollow capsules of small size, and the size of the capsules in solution was measured. More importantly, it clearly revealed that the entities are hollow in solution by combining static and dynamic LLS measurements. This method can be regarded as a complement to the CLSM method, and it possesses the advantage of no need for fluorescence labeling.

REFERENCES

- 1 Donath, E., Sukhorukov, G.B., Caruso, F., Davis, S.A. and Möhwald, H., *Angew. Chem. Int. Ed.*, 1998, 37: 2202
- 2 Decher, G., *Science*, 1997, 277: 1232
- 3 Zhang, Y., Yang, S., Guan, Y., Cao, W. and Xu, J., *Macromolecules*, 2003, 36: 4238
- 4 Zhang, Y., Guan, Y., Yang, S., Xu, J. and Han, C.C., *Adv. Mater.*, 2003, 15: 832
- 5 (a) Antipov, A.A., Sukhorukov, G.B., Fedutik, Y.A., Hartmann, J., Giersig, M. and Möhwald, H., *Langmuir*, 2002, 18: 66873
(b) Dai, Z., Möhwald, H., Tiersch, B. and Dähne, L., *Langmuir*, 2002, 18: 9533
(c) Gao, C., Leporatti, S., Donath, E. and Möhwald, H., *Langmuir*, 2000, 16: 7144
(d) Lulevich, V.V., Radtchenko, I.L., Sukhorukov, G.B. and Vinogradova, O.I., *Macromolecules*, 2003, 36: 2832
(e) Lulevich, V.V., Radtchenko, I.L., Sukhorukov, G.B. and Vinogradova, O.I., *J. Phys. Chem. B*, 2003, 107: 2735
- 6 Li, L., Sosnowski, S., Kumacheva, E., Winnik, M.A., Rajaram, S., Balke, S.T. and Chaffey, C.E., *Langmuir*, 1996, 12: 2141
- 7 (a) Niu, A., Liaw, D., Sang, H. and Wu, C., *Macromolecules*, 2000, 33: 3492
(b) Wang, X. and Wu, C., *Macromolecules*, 1999, 32: 4299
(c) Balsara, N.P. and Fetters, L.J., *Macromolecules*, 1999, 32: 5147
(d) Reinecke, H., Mijangos, C., Lopez, D. and Guenet, J.M., *Macromolecules*, 2000, 33: 2049
(e) Wang, X., Goh, S.H., Lu, Z.H., Lee, S.Y. and Wu, C., *Macromolecules*, 1999, 32: 2786
(f) Peng, S. and Wu, C., *Macromolecules*, 1999, 32: 585
- 8 Zhou, S., Burger, C., Chu, B., Sawamura, M., Nagahama, N., Toganoh, M., Hackler, U. E., Isobe, H. and Nakamura, E., *Science*, 2001, 291: 1944
- 9 Pastoriza-Santos, I., Schöler, B. and Caruso, F., *Adv. Funct. Mater.*, 2001, 11: 122
- 10 Gittins, D.I. and Caruso, F., *Adv. Mater.*, 2000, 12: 1947
- 11 Moya, S., Dahne, L., Voigt, A., Leporatti, S., Donath, E. and Möhwald, H., *Colloids and Surfaces, A*, 2001, 183: 27