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Supporting Information

Direct Formation of Giant Unilamellar Vesicles from Microparticles of Polyion Complexes and Investigation of Their Properties Using a Microfluidic Chamber

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1. Detachment of satellite vesicles inside and outside the mother vesicles

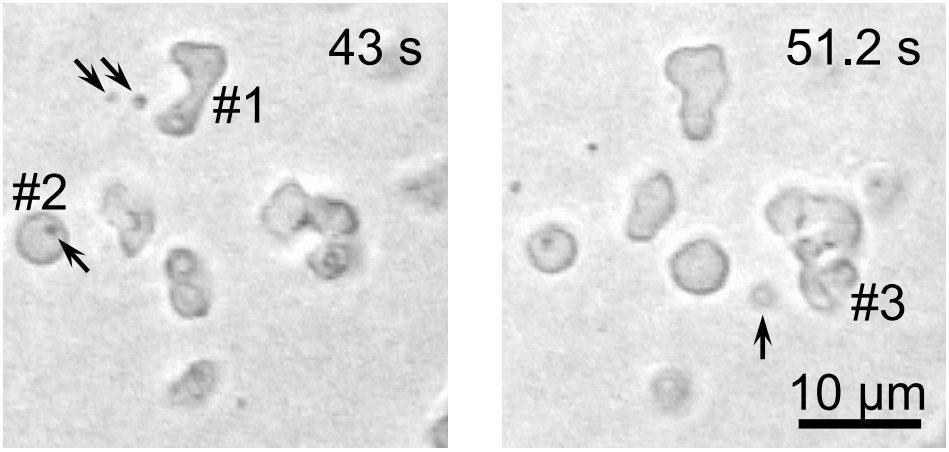

Fig. S1 Example images of satellite vesicles (indicated by black arrows) generated during the morphological changes in PIC microparticles. The time at which ultrapure water was introduced into the main channel is set at 0 s. Left (43 s): two PIC satellites detached from #1 and one satellite remained inside #2. Right (51.2 s): one satellite detached from #3.

2. Relationship between radius of initial PIC microparticles in 150 mM NaCl and radius of the main spherical vesicles

From Fig. S2, it is seen that the radii of the main spherical vesicles are proportional to the 1.5^{th} power of the radii of the initial PIC microparticles. This indicates that the surface areas of the formed main vesicles are proportional to the volumes of the initial PIC microparticles.

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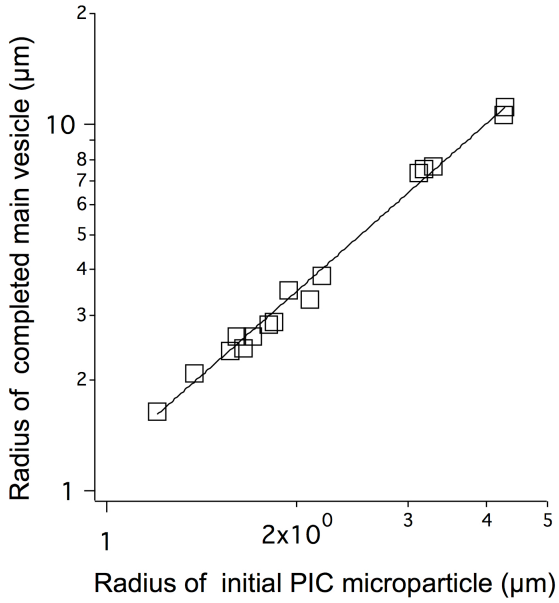

Fig. S2 Plot of the radius of initial PIC microparticles vs. the radius of the main spherical vesicles (N = 16). The solid line is a guide to the eye; $y \propto x^{1.5}$.

3. Morphological change of PIC microparticles exposed to 10 mM NaCl

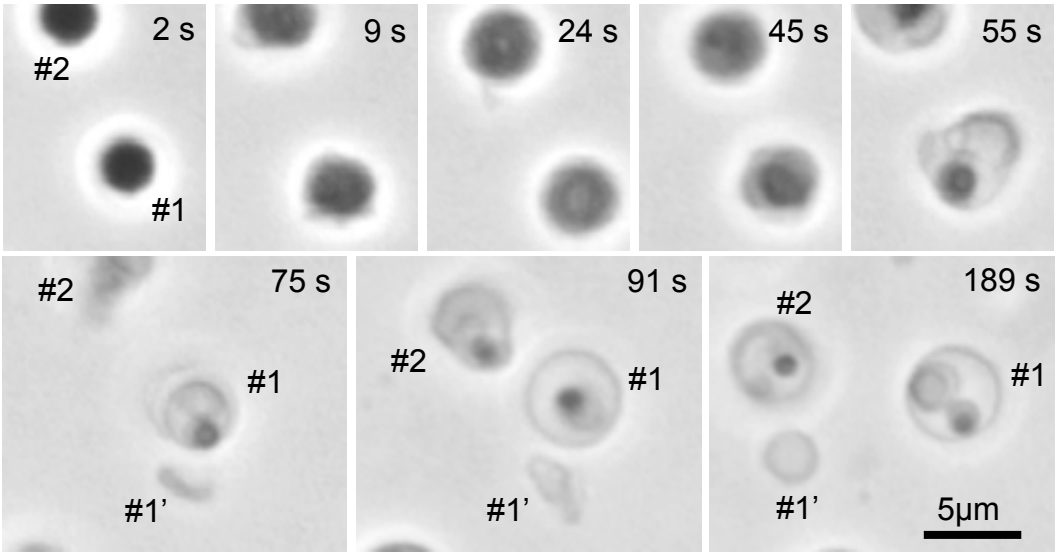

Fig. S3 Time development of the morphological changes in PIC microparticles during solution exchange of 150 mM NaCl with 10 mM NaCl. The time at which 10 mM NaCl was introduced into the main channel is set at 0 s. Here, vesicle #1' is detached from #1. A movie file is available in the Supporting Information (Movie 2).

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4. Morphological changes of a PICsome exposed to 1 M NaCl

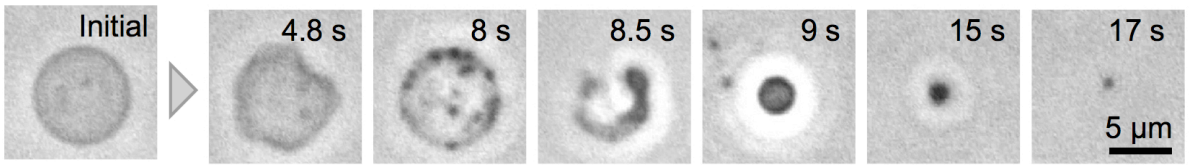

Fig. S4 Example phase-contrast images of the morphological changes in the PICsome during solution exchange of ultrapure water with 1 M NaCl. Initially, micropockets were filled with ultrapure water and tight spherical PICsomes were observed therein. The time at which 1 M NaCl solution was introduced into the main channel is set at 0 s. A movie file is available in the Supporting Information (Movie 3).

5. Encapsulation of a fluorescence-labeled antibody into PICsomes

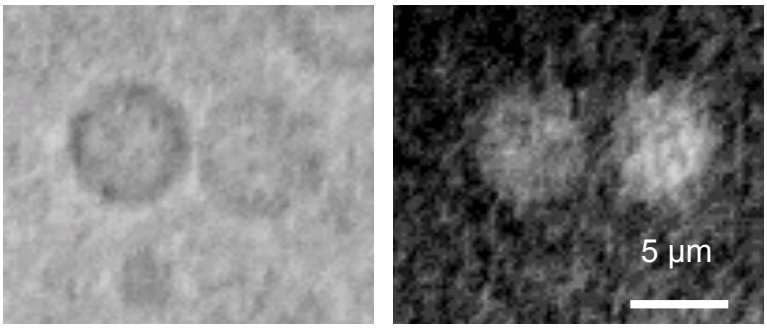

Fig. S5 Left: a phase-contrast image. Right: a fluorescence image. Images were captured 40 min after vesicle formation, i.e., solution exchange to ultrapure water.

6. Exposure of formed PICsomes to FITC-Dex40k in the micropockets

Initially, the micropockets were filled with ultrapure water, and spherical PICsomes were observed therein. Thereafter, ultrapure water was replaced with 1 μM FITC-Dex40k solution (without salt) and kept for 20 min. After that, FITC-Dex40k solution was flushed with ultrapure water, and then, encapsulation of the guest polymers in the PICsomes was

examined using fluorescence microscopy and phase-contrast microscopy (Fig. S6). When the FITC-Dex40k solution was introduced, the PICsomes were recognized as spherical dark shadows against a high-fluorescence background (A). Then, when the surrounding solution was changed to ultrapure water, the fluoresence intensity decreased. Meanwhile, the fluorescence intensity of the inner PICsomes was still low, making their recognition by fluorescence microscopy difficult (B). After 3 h of flushing with ultrapure water, the fluorescence background further decreased, and the PICsomes could not be recognized by fluorescence microscopy (C). This result shows that FITC-Dex40k does not enter the preformed PICsomes.

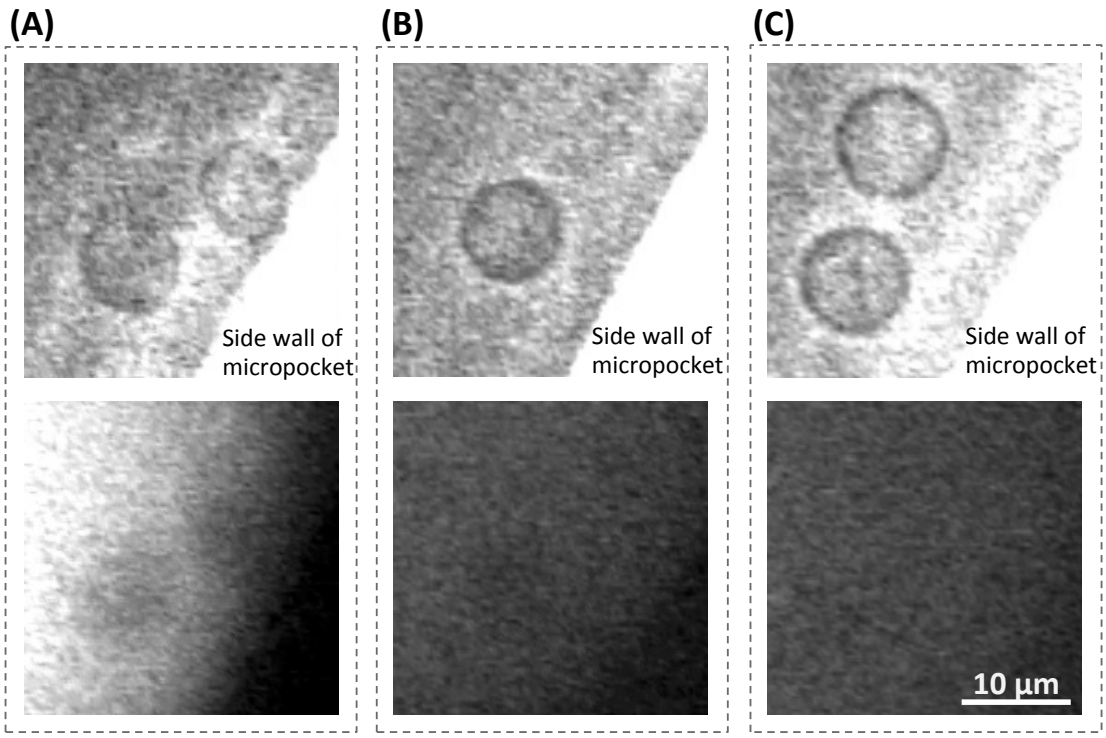

Fig. S6 Examples of phase-contrast images (top row) and fluorescence images (bottom row) of PICsomes exposed to FITC-Dex40k solution, followed by flushing with ultrapure water. (A) PICsomes 15 min after exposure to the FITC-Dex40k solution. Images recorded (B) 1 h and (C) 3 h after flushing with ultrapure water.

7. Deformation of cross-linked vesicles exposed to 1 M glucose

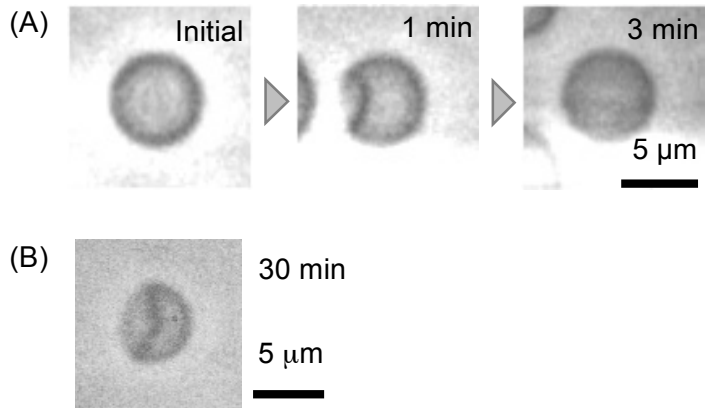

Fig. S7 (A) Partial deformation and retrieval of the cross-linked PICsome during solution exchange of ultrapure water with 1 M glucose in the micropockets. The time at which the glucose solution was introduced into the main channel is set at 0 s. (B) An example image of an overly deformed and irreversible bowl-like structure.

8. List of Movies

Movie files are available on the WWW under

http://www.bntl.t.u-tokyo.ac.jp/oana/PICsomes.html

Movie 1:

Formation of spherical vesicles from PIC microparticles during solution exchange of 150 mM NaCl with ultrapure water (played at 2× speed) corresponding to Fig. 3. The movie starts 5 s after introduction of ultrapure water into the main channel.

Movie2:

Example morphological change of PIC microparticles during solution exchange of 150 mM NaCl with 10 mM NaCl (played at 2× speed) corresponding to Fig. S3. The movie starts 2 s after the introduction of 10 mM NaCl into the main channel.

Movie 3:

Breaking and dissolving of a PIC spherical vesicle exposed to 1 M NaCl (a real-time movie) corresponding to Fig. S4. The movie starts 4 s after the introduction of 1 M NaCl into the main channel.

Movie 4:

Fusion of PICsomes in the presence of 150 mM NaCl using optical tweezers (a real-time movie) corresponding to Fig. 5.

Movie 5:

Micromanipulation of PICsomes in ultrapure water (a real-time movie). The PICsomes are never fused as described in section 2.3.

Movie 6:

Micromanipulation of PICsomes in the presence of 1 M glucose (a real-time movie). The PICsomes are never fused as described in section 2.3.

Movie 7:

Partial deformation and retrieval of the cross-linked PICsomes during solution exchange of ultrapure water with 1 M NaCl in the micropocket (a real-time movie) corresponding to Fig. 8A. The movie starts 5 s after the introduction of 1 M NaCl into the main channel.