Supporting Information

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Hydrophobic Shell Loading of Biopolyelectrolyte Capsules

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Films characterization by quartz crystal microbalance with dissipation monitoring. The (QCHI/HA20C10) film buildup (where \( i \) denotes the number of layer pairs) was followed by in situ quartz crystal microbalance (QCM with dissipation monitoring, D300, Qsense, Sweden). The gold-coated quartz crystal was excited at its fundamental frequency (about 5 MHz, \( \nu = 1 \)) as well as at the third, fifth and seventh overtones (\( \nu = 3, 5 \) and 7 corresponding to 15, 25 and 35 MHz, respectively). Changes in the resonance frequencies \( \Delta f \) and in the relaxation of the vibration once the excitation is stopped \( \Delta D \) were measured at the four frequencies.

Atomic force microscopy. AFM images of the dried microcapsules were carried out in air with a PicoPlus AFM in tapping mode using tetrahedral tips (OMCL-AC240TM-E tip from Olympus) with a resonance frequency of 75 kHz and a spring constant of 2 N m\(^{-1}\). Capsules deposited on a mica substrate were imaged at line rates of 1 Hz. For surface roughness analysis, 1 × 1 µm\(^2\) AFM images were obtained and the arithmetic average roughness \( R_a \) was calculated according to:

\[
R_a = \frac{1}{N_x N_y} \sum_{i=1}^{N_x} \sum_{j=1}^{N_y} \left| z_{ij} - z_{mean} \right|
\]

(1)

where \( z_{ij} \) is the height of a given pixel, \( z_{mean} \) is the average height of the pixels, and \( N_x = N_y = 512 \) are the number of pixels in the \( x \) and \( y \) directions.
Measurement of NR release from capsules over time. The suspension of capsules (5 mL) in PBS was divided in 5 samples, corresponding to 5 different time periods from 0 to 8 days, noted D0, D2, D4, D6, and D8 respectively. The D0 sample, consisting in 200 µL of capsules suspension freshly prepared, was treated as described in the experimental section to determine the amount of NR incorporated. This process was performed in duplicate in order to confirm the values.

The D2, D4, D6 and D8 samples (each consisting of 1 mL of capsules suspension) were introduced in Spectra/Por® dialysis membranes (6-8 kDa cutoff) and the samples were immersed in PBS (200 mL). Each sample was washed four times with PBS (200 mL) during 2 days. For each sample (D2, D4, D6 and D8), 200 µL of capsules suspension were taken up and underwent a treatment similar to the D0 sample for determination of the amount of NR in the capsules. These processes were performed in duplicate.

The analysis of NR contained in the capsules as a function of time demonstrates only a minor release of the dye in spite of the washing steps with PBS, as shown in Fig. SI 4. This demonstrates the high stability of the entrapment of the hydrophobic molecule in the nanoshell.
Figure SI 1: (QCHI/HA20C10) film growth in PBS (pH 7.4) as measured by QCM-D on gold coated crystals. Differences in the QCM frequency shifts (A) and in the dissipation (B) measured at 15 MHz are represented as a function of the number of deposited layers. All QCHI concentrations and HA20C10 were of 2 mg/mL.
**Figure SI 2.** AFM topographical image of a microcapsule made of 4.5 layer pairs of (QCHI-4/HAl0C10)_{4.5} layers. Image size is 10 × 10 µm$^2$. 
Figure SI 3. Calibration curve for the absorbance of Nile Red in ethanol (measured at 590 nm) as a function of its concentration in solution.
Figure SI 4. Fluorescence intensity measurements of NR intensity remaining in (HA20C10/QCHI-4)$_3$ microcapsules over a 8 day period of immersion in PBS. The minor decrease in fluorescence indicates that the stability of NR in the hydrophobic nanodomains is very high.