Supporting Information

Programmed pH-Responsive Microcapsules for the Controlled Release of CdSe/ZnS Quantum Dots

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Table S1. The nucleic acid sequences used in this study.

No.	Sequence (5'→3')
1	TCGAAGCTTTGGAGGGGAGGGGAGGTTTACCTCCCCTCCCCTCCCT TTGCCTCCCCTCCCCTCCGTACTC
2	AAAGCTTCGAGGAGGGGAGGGGAGGTTTACCTCCCCTCCCCTCCCT TTGCCTCCCCTCCCCTCCGTACTC
3	GGAGGGGAGGGGAGGTTTACCTCCCCTCCCCTCCCTTTGCCTCCCCT CCCCTCCTCGACTTTCTGCTCTC
4	GGAGGGGAGGGGAGGTTTACCTCCCCTCCCCTCCCTTTGCCTCCCCT CCCCTCCTCGACTGAGAGCAGAA
5	AGTCGAGGAGGGT CGTCACTATCTGAGTACGGAGGG
6	CGTAAGCGTATTTTTTTTTTTTTTGATAGTGACG
7	AGTCGAGGAGGGTTACGCTTACGTGAGTACGGAGGG
8	ATGTCCTCAGCCTGTTTTTCTTTTCTTTTCTTTTCTTCACCAAGAAAA GAAAAGAAAAGAA
9	CTGAGGACATCCTGTTTTTCTTTTCTTTTCTTTTCTTCACCAAGAAAA GAAAAGAAAAGAA
10	AAGAAAAGAAAAGAAAAGAACACCTTCTTTTCTTTTCTTTTCTTTTC AGTCAAGCAGTCAC
11	AAGAAAAGAAAAGAAAAGAACACCTTCTTTTCTTTTCTTTTCTTTTC AGTCGTGACTGCTT
12	GACTGTTTTCTTTTCTTTTCTTTTCTTTCTTAGTCGACTTTCTTTTCTT TTCTTTTCTTTTATAGG
13	GATCAGCTAGCATTGTCTTGAAATCGTCGACTAAG
14	GACTGTTTTCTTTTCTTTTCTTTTCTTTCTAGCTGATCTTTCTTTTCTT TTCTTTTCTTTTATAGG

The triplex sequences are underlined.

Preparation of MPA-capped CdSe/ZnS QDs

The commercial CdSe/ZnS quantum dots were precipitated from toluene solution by adding 1 mL of methanol to 0.25 mL of QDs in toluene and followed by centrifugation for 10 min at 5000 rcf. The precipitated QDs were re-suspended in 0.5 mL chloroform. Subsequently, 100 μL of capping reagent solution composed of 40 μL mercaptopropionic acid, MPA, and 90 mg potassium hydroxide in 1 mL of methanol was added and shaken for 30 min. The MPA-capped QDs were transferred to water phase by adding 1 mL of 1 mM sodium hydroxide solution and separated from chloroform by centrifuging at 3000 rcf for 1 min. Two sequential QDs precipitation steps by the addition of NaCl and methanol, followed by a centrifugation at 3000 rcf for 1 min, were applied to remove the excess MPA. The resulting MPA-capped QDs were dissolved in 10 mM HEPES buffer (pH 7.4).

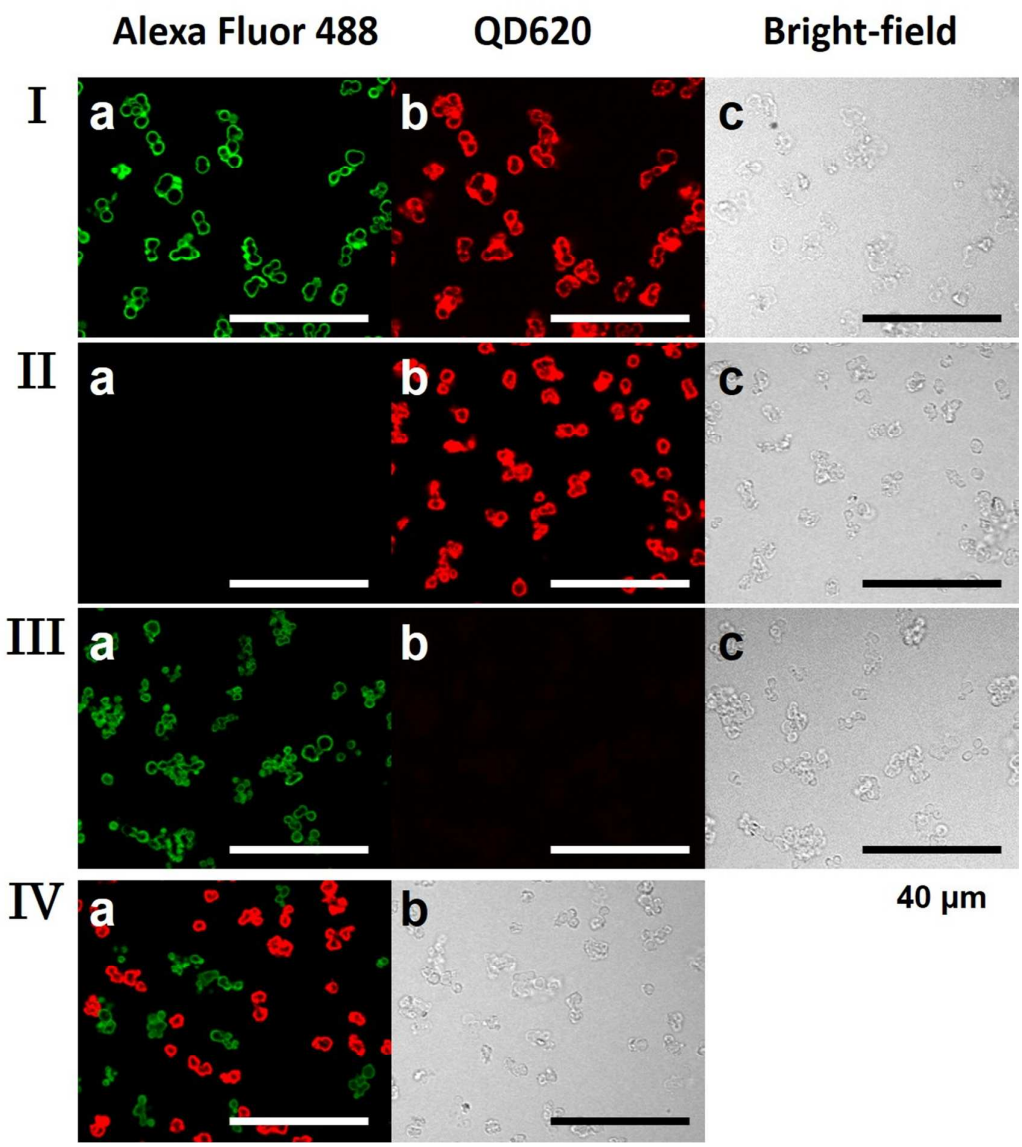

Figure S1. Confocal fluorescence microscopy images and bright-field microscopy images of different microcapsules. Panel I: The QDs-loaded Alexa Fluor 488-labeled DNA microcapsules. Panel II: The shell unlabeled QDs-loaded microcapsules. Panel III: Alexa Fluor 488-labeled DNA microcapsules that are not loaded with the QDs. Panel IV: Mixture of the shell unlabeled QDs-loaded microcapsules and the Alexa Fluor 488-labeled unloaded microcapsules. Figure a in panel I, II, and III show the fluorescence generated from Alexa Fluor 488. Figure b in panel I, II, and III show the fluorescence generated from QDs. Panel IV, a, depicts a merged fluorescence image of Alexa Fluor 488 and QDs emitting at 620 nm. Figure c in panel I, II, III and figure b in panel IV show the bright-field images of microcapsules. Scale bar is 40 μm.

Control experiments of non-pH-responsive DNA microcapsules

Figure S2 demonstrates the microcapsules that include DNA shells that lack the C-G·C$^+$ or T-A·T triplex-responsive bridges, and loaded with CdSe/ZnS QDs (λ_{em} = 620 nm) are not affected and not unlocked upon subjecting to pH = 5.0 or pH = 9.0. Toward this goal we loaded ATP-responsive microcapsules with CdSe/ZnS QDs, λ_{em} = 620 nm. (For the preparation of this microcapsules see *ACS Nano* **2015**, *9*, 9078−9086). Figure S2 shows the fluorescence spectra of the "released" QDs upon subjecting the microcapsules to pH = 7.0, curve a, pH = 5.0, curve b, and pH = 9.0, curve c, only a minute background luminescence is observed, implying that the different pH values have no effect on the release properties of nucleic acid-stabilized microcapsules that lack the pH-responsive elements. For comparison Figure S2, curve d, shows the fluorescence spectrum of the released CdSe/ZnS from the C-G·C$^+$ triplex-responsive microcapsules at pH = 5.0. The results indicate the selectivity of the C-G·C$^+$ or T-A·T triplex-responsive microcapsules towards the pH-induced release of the QDs loads.

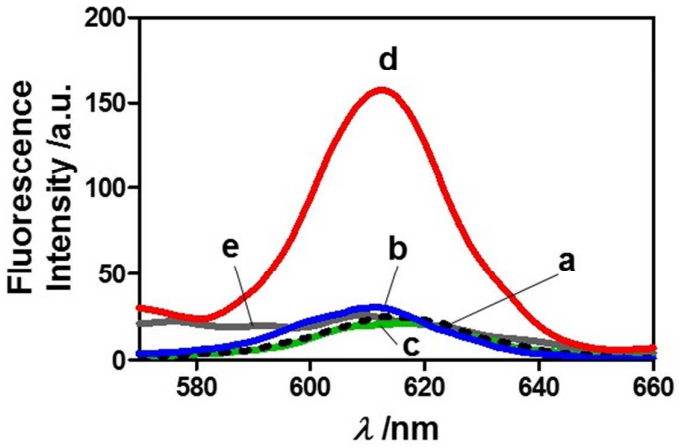

Figure S2. Fluorescence spectra corresponding to the released QDs from the microcapsules subjecting to different conditions for a fixed time-interval of 30 min: (a) ATP-responsive microcapsules subjecting to pH = 7.0, (b) ATP-responsive microcapsules subjecting to pH = 5.0, (c) ATP-responsive microcapsules subjecting to pH = 9.0, (d) C-G·C$^+$ triplex-responsive microcapsules subjecting to pH = 5.0, and (e) C-G·C$^+$ triplex-responsive microcapsules subjecting to pH = 7.0.

Effect of pH on the fluorescence of the QDs

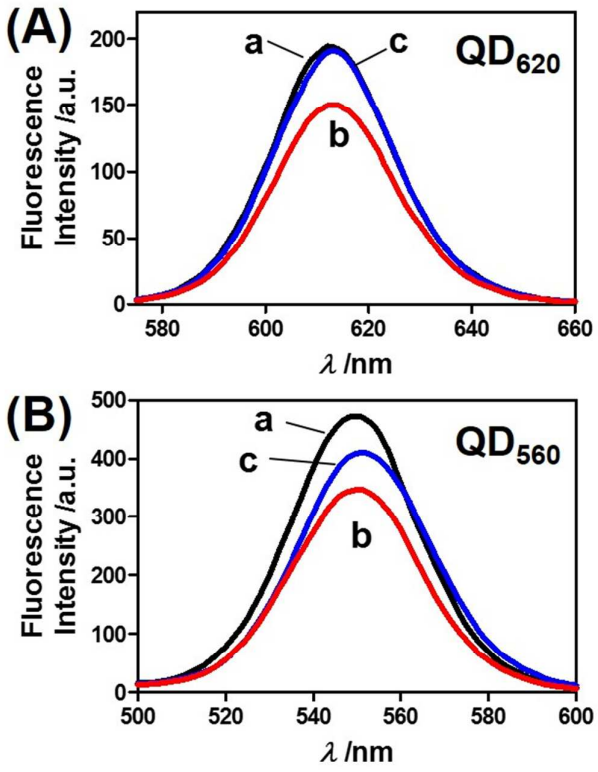

Figure S3. Effects of pH on the fluorescence intensities of the CdSe/ZnS quantum dots. (A) 620 nm-emitting QDs subjecting to pH = 7.2 (a), pH = 5.0 (b), and pH = 9.0 (c). (B) 560 nm-emitting QDs subjecting to pH = 7.2 (a), pH = 5.0 (b), and pH = 9.0 (c).

Figure S4 depicts the confocal microscopy images of the CdSe/ZnS QDs-modified microcapsules at pH = 7.2, panel I, and after treatment at pH = 5.0, and release of the QDs for a time-interval of 30 minutes. Evidently, the fluorescence intensities of the microcapsules treated at pH = 5.0 is substantially lower (*ca.* 70% decrease) as compared to the fluorescence intensities of the microcapsules at pH = 7.2. The pH has a low effect on the fluorescence intensities of the QDs, where only a decrease of 25% was observed upon switching the pH from 7.2 to 5.0 (*cf.* Figure S3(A)). Thus, the pronounced decrease in the fluorescence of the microcapsules after treatment at pH = 5.0 is attributed to the pH-stimulated release of the QDs from the microcapsules. Furthermore, the confocal microscopy image of the microcapsules treated at pH = 5.0, panel II, reveals a non-symmetric distribution of the residual QDs on the inner shell interface. Presumably, the QD-free regions represent pH-disrupted domains through which the QDs are released.

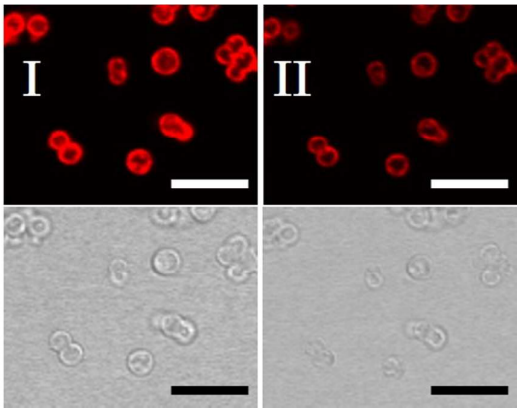

Figure S4. Fluorescence confocal microscopy images and bright-field microscopy images corresponding to: (I) C-G·C^+ triplex-coated QD^{620}-loaded $CaCO_3$ microcapsules after the dissolution of the core by EDTA. (II) pH-driven dissociated microcapsules under pH = 5.0 that resulted in the release of the QDs^{620}. Scale bar is 10 μm.

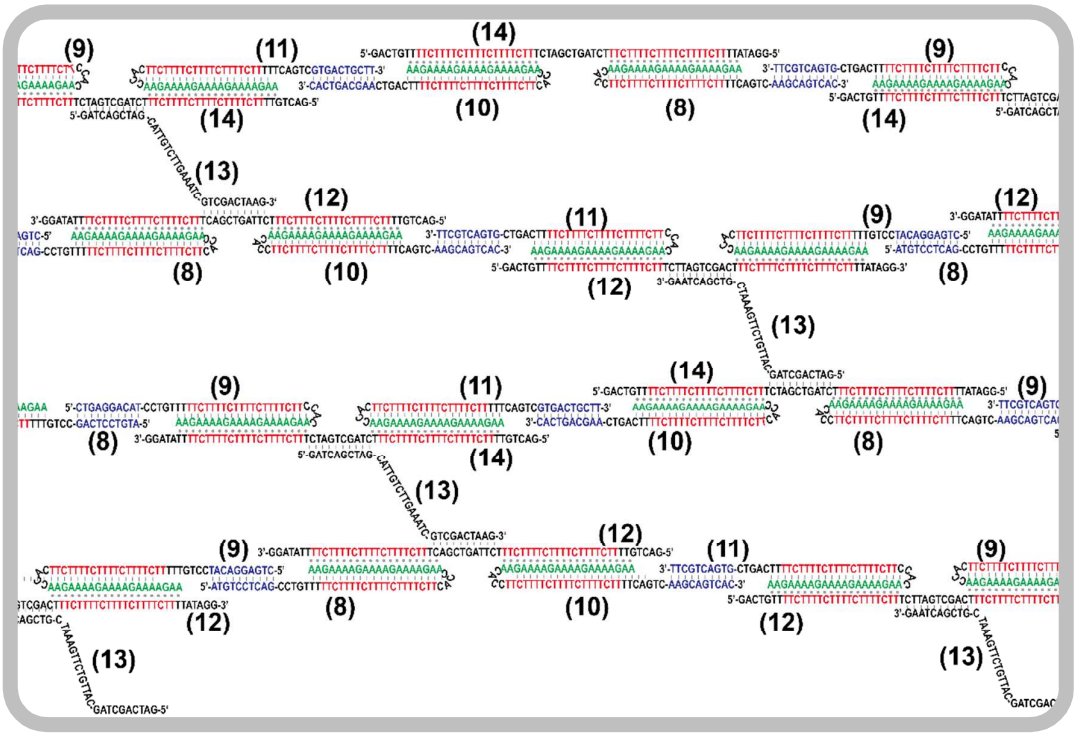

Figure S5. Detailed composition of four-layer constituents that build-up the T-A·T triplex pH-responsive shell of the microcapsules (Red-green-red sequences represent the T-A·T triplex structures).

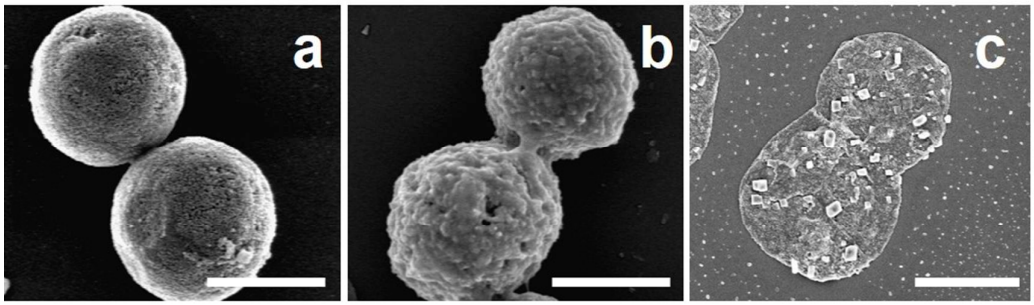

Figure S6. Scanning electron microscopy (SEM) images of uncoated quantum dot emitting at 560 nm (QD^{560})-loaded $CaCO_3$ microparticles (a), T-A·T triplex-coated QD^{560}-loaded $CaCO_3$ microparticles (b), and microcapsules after the dissolution of the $CaCO_3$ core (c). Scale bar is 2 μm.

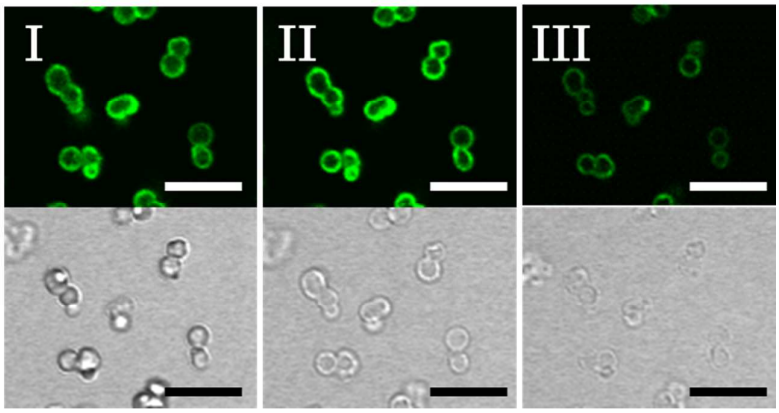

Figure S7. Fluorescence confocal microscopy images and bright field microscopy images corresponding to: (I) T-A·T triplex-coated QD^{560}-loaded $CaCO_3$ microparticles. (II) QD^{560}-loaded microcapsules after the dissolution of the core by EDTA. (III) pH-driven dissociated microcapsules under pH=9.0 that resulted in the release of the QDs^{560}. Scale bar is 10 μm.

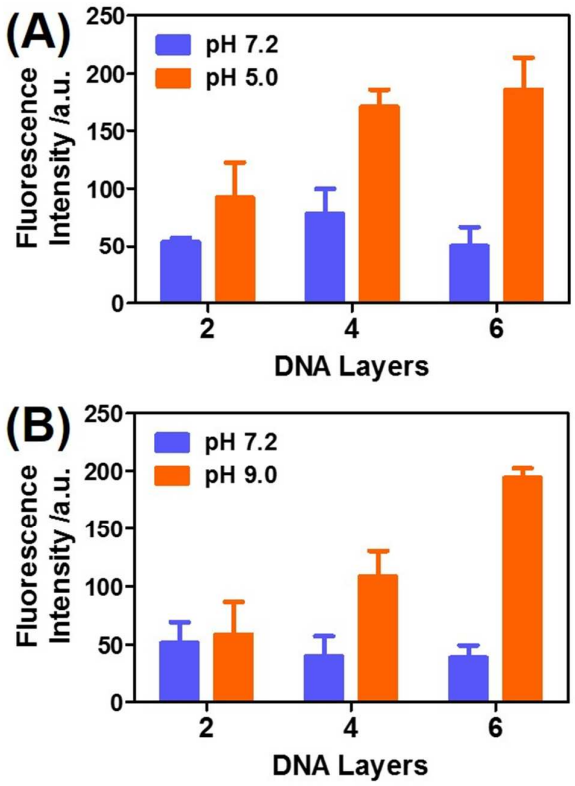

Figure S8. (A) Optimization of the number of C-G·C^+ triplex DNA coated layers. Comparisons of the released QDs^{620} from two, four, and six layers of DNA deposited microcapsules upon treatment with acidic solutions (pH 5.0). (B) Optimization of the coated T-A·T triplex DNA layer number. Comparisons of the released QDs^{560} from two, four, and six layers of DNA deposited microcapsules upon treatment with basic solutions (pH 9.0).

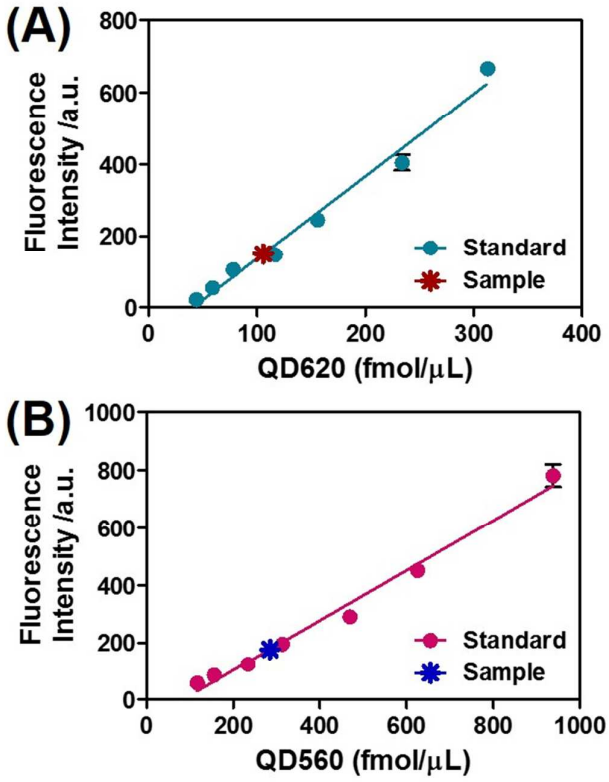

Figure S9. (A) Calibration curve of CdSe/ZnS QDs^{620} performed in pH 5.0 solution, which consists of 10 mM HEPES and 500 mM NaCl adjusting to pH 5.0 with 10% acetic acid. The fluorescence intensity of the released QDs^{620} upon subjecting the C-G·C^+ responsive microcapsules to acidic pH (pH = 5.0) for 30 min is indicated with a star. (B) Calibration curve of CdSe/ZnS QDs^{560} performed in pH 9.0 solution, which consists of 10 mM HEPES and 500 mM NaCl adjusting to pH 9.0 with 15% NH_4OH. The fluorescence intensity of the released QDs^{560} upon subjecting the T-A·T responsive microcapsules to basic pH (pH = 9.0) for 30 min is indicated with a star.

Figure S10 depicts the microcapsules precipitate of the C-G·C^+ (left) and T-A·T (right) QDs-loaded microcapsules after centrifugation of the respective microcapsules suspension at 500 rpm (g = 30 rcf). The precipitated microcapsules can be re-suspended upon shaking.

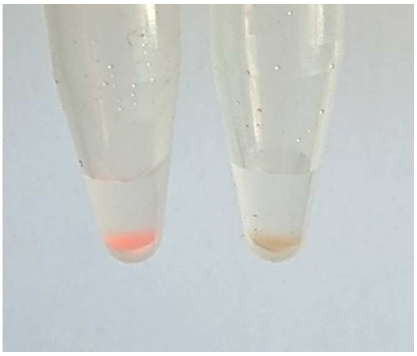

Figure S10. Photograph of the QD^{620}-loaded C-G·C^+ (left) and QD^{560}-loaded T-A·T (right) triplex-stabilized microcapsules after centrifugation at 500 rpm.