

Supplementary Materials: Ciprofloxacin-Loaded Mixed Polymeric Micelles as Antibiofilm Agents

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Supplementary information:

1. Preparation of mixed micelles

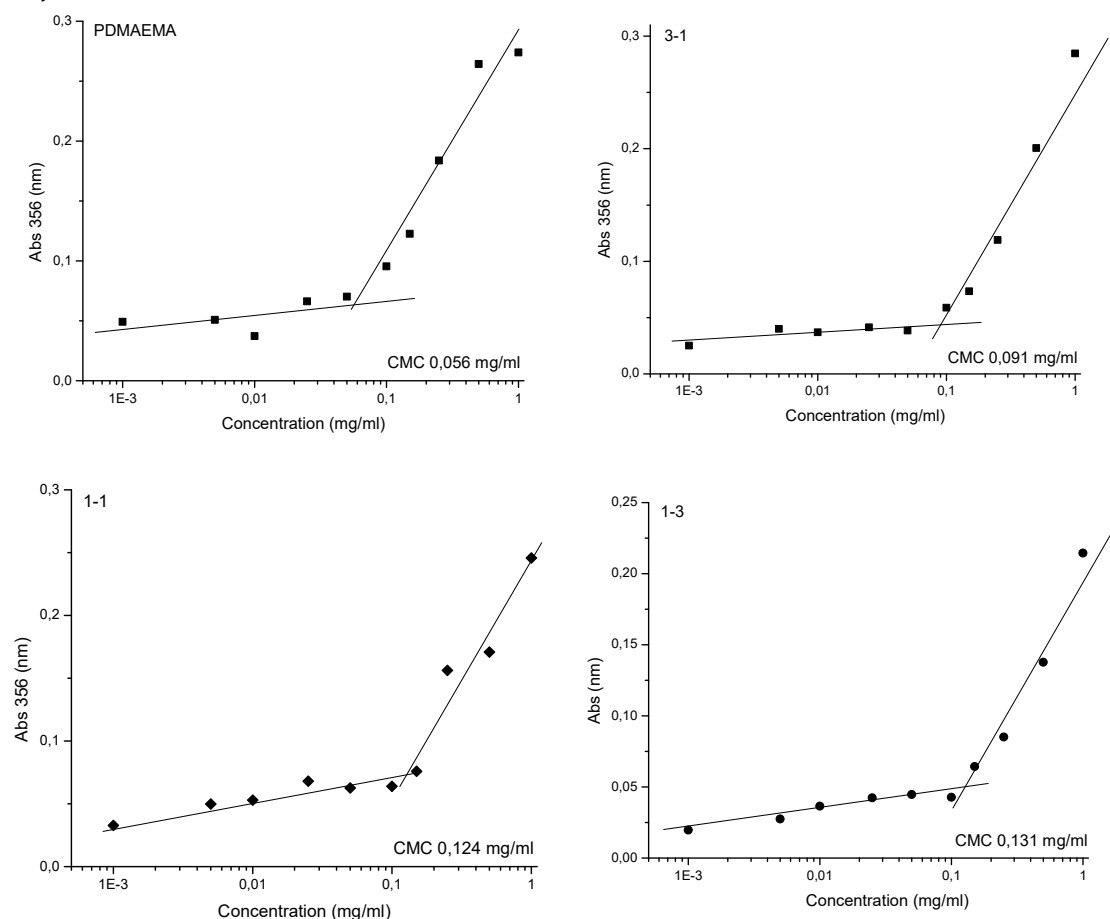
Calculation of hydrophilic-lipophilic balance (HLB) of copolymers

For HLB calculation the Griffin method [1] was used following the equation:

$$HLB = 20 * M_h/M$$

where M_h is the molar mass of the hydrophilic block of copolymer, and M is the molecular mass of the whole macromolecule.

Determination of critical micellization concentration (CMC)



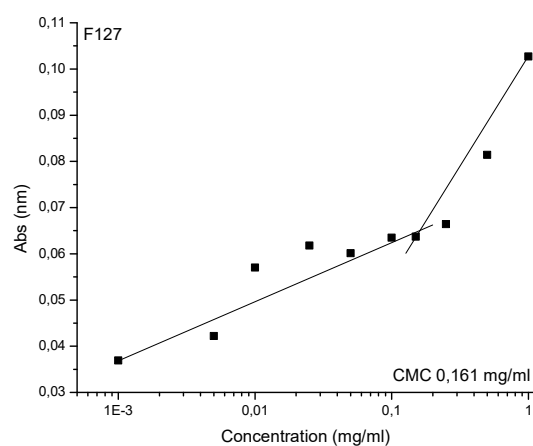
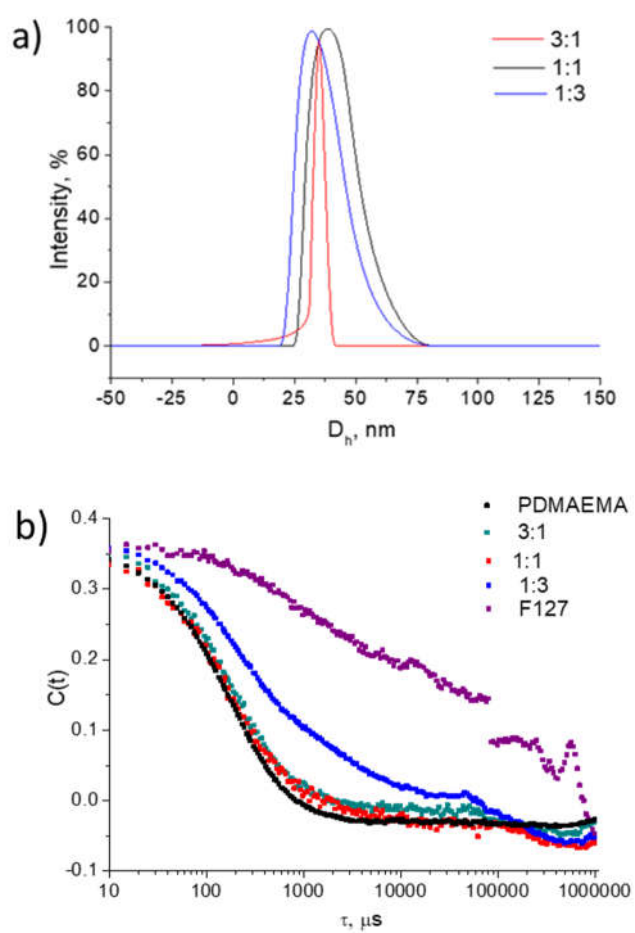


Figure S1. Critical micelle concentration plot of SCPMs and MPMs based on PDMAEMA-b-PCL-b-PDMAEMA and Pluronic F127 triblock copolymers.



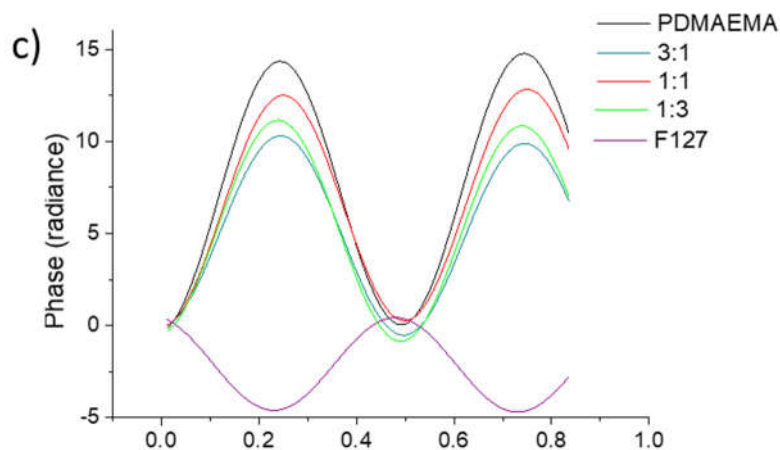
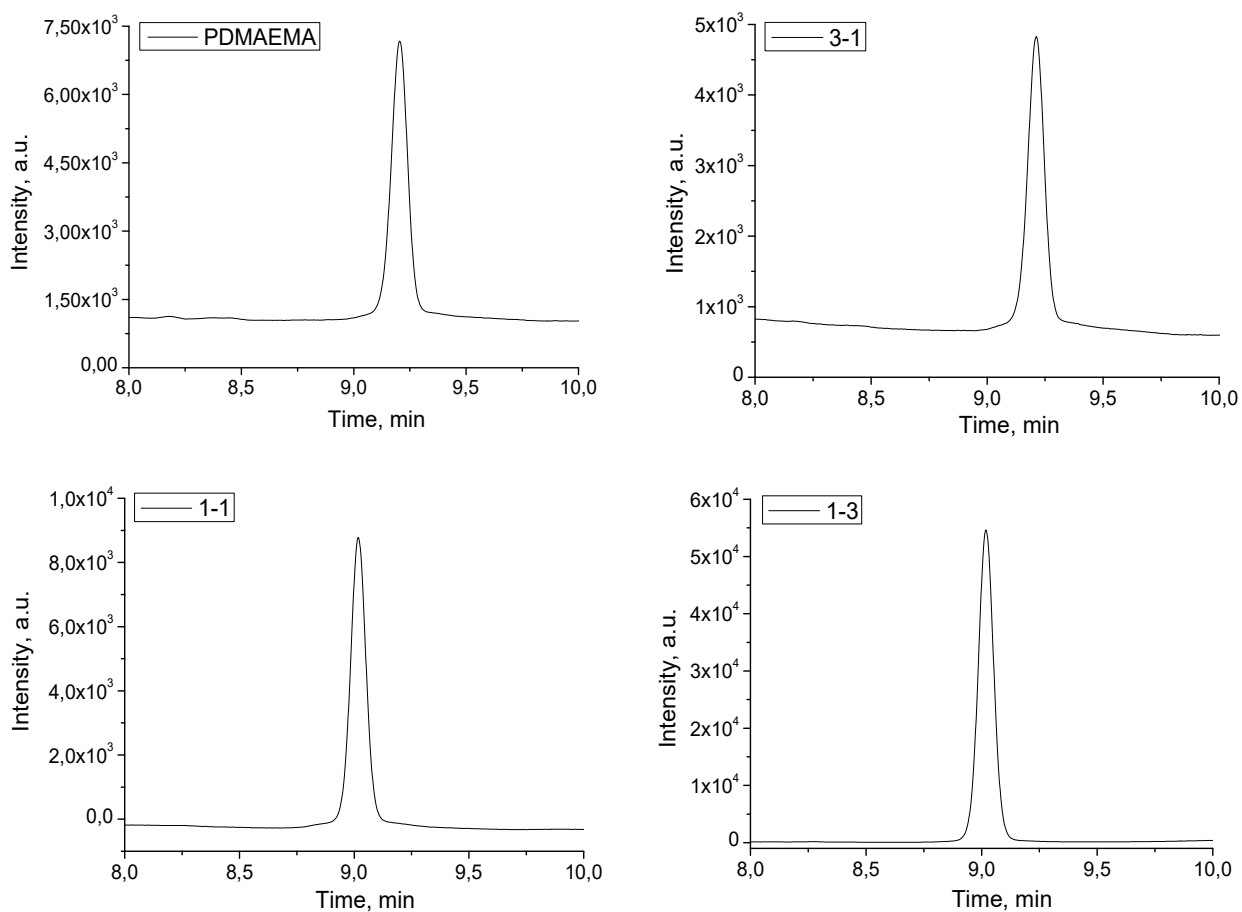


Figure S2. Size distribution curves (a), correlation curves (b) and phase plots (c) of SCPMs and MPMs based on PDMAEMA-*b*-PCL-*b*-PDMAEMA and Pluronic F127 triblock copolymers at concentration 0.01 mg.ml⁻¹.

2. Loading of micelles with CF



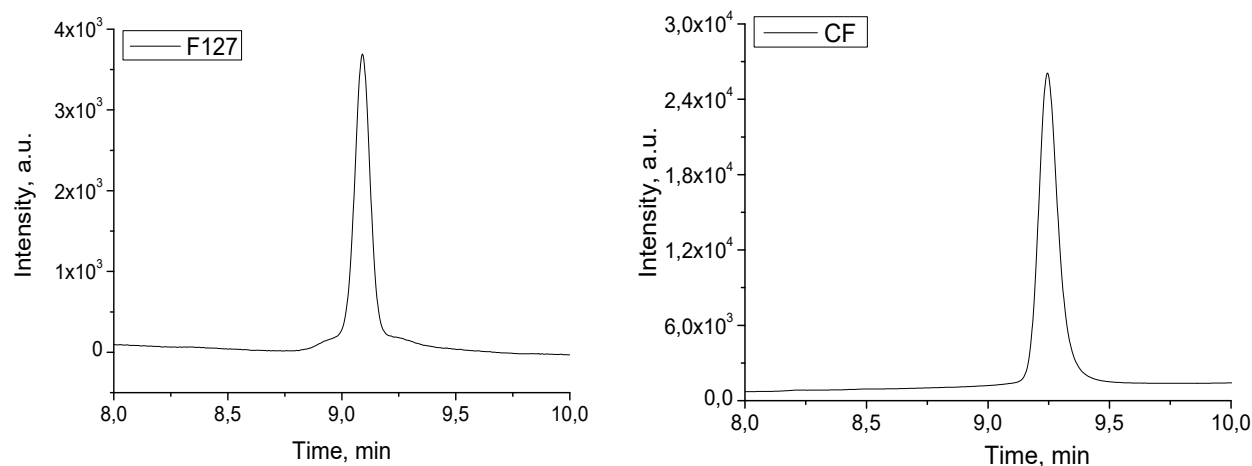


Figure S3. HPLC chromatograms of non-loaded (free) CF extracted from SCPMs and MPMs based on PDMAEMA-b-PCL-b-PDMAEMA and Pluronic F127 triblock copolymers. HPLC chromatogram of pure CF used as standard is given as well. The samples were prepared at polymer to drug mass ratio 10:1. The integrated area of the peaks was used for calculation of EE and DLC.

Table S1. Encapsulation efficiency (%) towards CF, determined by HPLC and UV spectrophotometry, of SCPMs and MPMs based on PDMAEMA-b-PCL-b-PDMAEMA and Pluronic F127 triblock copolymers prepared at different polymer to drug mass ratio. Each value represents the arithmetic mean of three separate experiments.

Polymeric micelles Polymer to drug ratio	PDMAEMA		3:1		1:1		1:3		F127	
	HPLC	UV	HPLC	UV	HPLC	UV	HPLC	UV	HPLC	UV
50:1	85.6 ±4.3	90.9 ±4.6	98.9 ±2.9	97.7 ±3.3	87.3 ±5.2	90.2 ±4.5	97.9 ±2.4	97.0 ±2.8	99.0 ±1.8	96.4 ±3.8
10:1	98.1 ±4.1	98.7 ±3.6	98.8 ±2.5	99.0 ±2.1	99.2 ±1.6	99.2 ±2.2	99.4 ±2.1	99.1 ±1.9	98.6 ±1.8	91.2 ±2.2
5:1	74.8 ±4.8	80.1 ±3.6	86.9 ±4.3	92.5 ±4.1	84.9 ±3.8	90.7 ±4.1	92.1 ±4.5	94.2 ±4.9	82.2 ±4.3	73.0 ±4.2
2.5:1	48.9 ±5.0	59.13 ±5.1	51.2 ±5.2	64.1 ±5.9	52.9 ±5.2	69.6 ±6.1	62.0 ±6.0	70.3 ±5.8	31.0 ±4.6	30.2 ±4.8
1:1	42.8 ±4.3	68.0 ±5.9	54.4 ±5.2	74.6 ±6.9	63.0 ±6.0	70.6 ±6.2	57.7 ±6.1	78.6 ±6.9	43.4 ±4.3	41.0 ±4.0

Table S2. Drug loading content (%) towards CF, determined by HPLC and UV spectrophotometry, of SCPMs and MPMs based on PDMAEMA-b-PCL-b-PDMAEMA and Pluronic F127 triblock copolymers prepared at different polymer to drug mass ratio. Each value represents the arithmetic mean of three separate experiments.

Polymeric micelles Polymer to drug ratio	PDMAEMA		3:1		1:1		1:3		F127	
	HPLC	UV	HPLC	UV	HPLC	UV	HPLC	UV	HPLC	UV
50:1	2.1 ±0.2	2.2 ±0.1	2.0 ±0.2	2.0 ±0.3	1.8 ±0.1	1.9 ±0.1	2.2 ±0.1	2.1 ±0.2	2.0 ±0.1	1.9 ±0.2
10:1	10.4 ±0.8	10.5 ±0.4	11.0 ±0.5	10.9 ±0.5	10.9 ±0.4	10.9 ±0.3	10.3 ±0.4	10.3 ±0.9	9.9 ±0.8	9.3 ±0.7
5:1	15.6 ±1.1	17.0 ±0.9	17.9 ±0.6	18.7 ±0.9	17.5 ±0.8	18.7 ±0.6	19.2 ±0.8	19.6 ±0.9	16.4 ±0.9	14.6 ±0.7
2.5:1	20.6 ±1.9	25.0 ±1.0	21.2 ±1.0	26.6 ±0.9	21.9 ±0.9	28.8 ±1.1	26.3 ±0.7	29.8 ±1.2	12.4 ±1.3	12.4 ±1.2
1:1	44.5 ±1.3	70.1 ±1.2	56.6 ±1.3	74.4 ±1.5	65.5 ±1.2	73.8 ±1.2	59.4 ±1.4	80.3 ±1.5	43.4 ±1.8	41.0 ±1.6

3. CF release from micelles

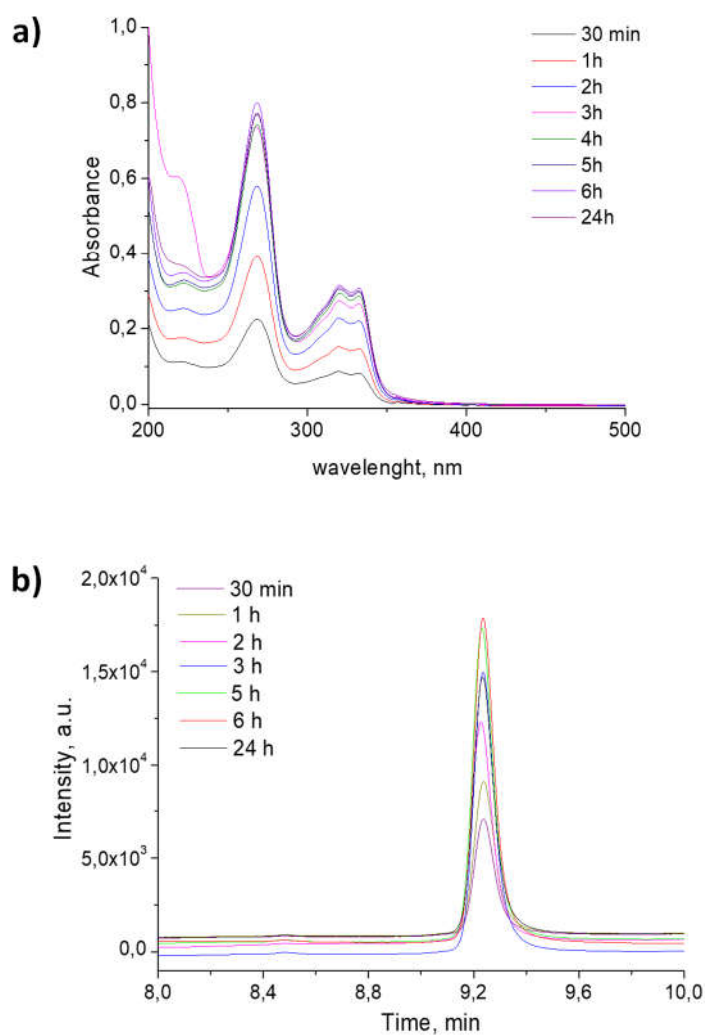


Figure S4. UV-Vis absorption spectra (a) and HPLC chromatograms (b) of CF released from SCPMs and MPMs based on PDMAEMA-b-PCL-b-PDMAEMA and Pluronic F127 triblock copolymers at 37 °C in phosphate buffer pH 7.4.

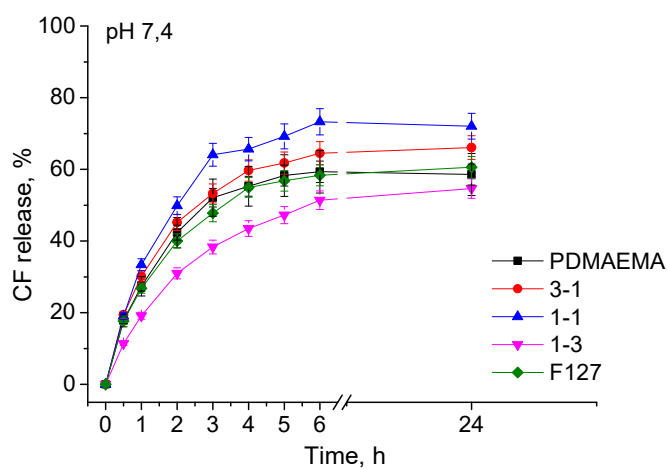


Figure S5. CF release profiles from SCPMs and MPMs based on PDMAEMA-b-PCL-b-PDMAEMA and Pluronic F127 triblock copolymers prepared at 10:1 polymer to drug mass ratio determined from UV-Vis spectrophotometry. The release was performed at 37 °C in phosphate buffer pH 7.4. Each data point in represents the arithmetic mean \pm SD of three separate experiments.

References:

1. Griffin WC: "Calculation of HLB Values of Non-Ionic Surfactants," Journal of the Society of Cosmetic Chemists 5 (1954): 259.