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Supporting Information for:

PLA2-responsive and SPIO-loaded phospholipid micelles

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Materials

Hydrogenated soy phosphatidylcholine (HSPC) and 1-palmitoyl-2-{6-[(7-nitro-2-1,3-benzoxadiazol-4-yl)amino]hexanoyl}-sn-glycero-3-phosphocholine (NBD-PC) were purchased from Avanti Polar Lipids, Inc. Inhibitor 3-[3-(2-amino-2-oxoethyl)-1-benzyl-2-ethylindol-5-yl]oxypropylphosphonic acid (LY311727) was obtained from Santa Cruz Biotechnology, Inc. Phospholipase A_2 (PLA_2) from Naja mossambica and Triton X-100 were purchased from Sigma-Aldrich Co. Other chemicals and reagents were commercially available and were of analytical grade. Water was obtained by Millipore Milli-Q purification system.

Synthesis of Hydrophobic 13 nm SPIO NPs

Oleic acid coated SPIO NPs were prepared by thermal decomposition as previously described.[1]

Synthesis of SPIO-loaded phospholipid micelles

SPIO-loaded phospholipid micelles were prepared using an oil-in-water emulsion method.[2] A mixture (150 μL) containing HSPCs (0.88 mg in chloroform), NBD-PC (0.1 mg in chloroform), and SPIO NPs (1 mg in toluene) was injected into a glass vial containing 3 mL of water, and the sample was sonicated (Branson Ultrasonics, Danbury, CT, USA) until a homogenous mixture was obtained. The toluene was then allowed to evaporate overnight. The control micelles, i.e. without SPIO, were prepared using similar procedures as SPIO-loaded micelles.

Purification of SPIO-loaded phospholipid micelles

SPIO-loaded phospholipid micelles samples were first centrifuged (Eppendorf Microcentriguge 5418) at 1000 rpm for 30 minutes to remove large aggregates. In order to remove empty micelles, the resulting supernatant was then centrifuged at 10,000 rpm for half hour, and the pellet was resuspended in water. The stock solution of SPIO-loaded phospholipids micelles was stored in the dark at 4 °C.

Sensing of PLA_2

50 uL PLA_2 was added to 950 µL HEPES buffer (10 mM, pH 7.4) containing $CaCl_2$ and SPIO-loaded phospholipids micelles. Fluorescence was recorded every 60 s for 1 h.

Instrumentation

Fluorescence spectra measurements were done on a SPEX FluoroMax-3 spectrofluorometer (Horiba Jobin Yvon). The fluorescence spectra were recorded between 490 and 650 nm upon excitation at 464 nm. Dynamic light scattering (DLS) measurements were performed on a Zetasizer Nano from Malvern Instruments. Transmission electron microscopy (TEM, JEM-1010) was used to characterize the size and shape of the small hydrophobic SPIO and SPIO-loaded phospholipid micelles. T2 relaxation times were determined using a Bruker mq60 MR relaxometer operating at 1.41 T (60 MHz). The Fe concentration in samples was determined by ICP-OES analysis using a Genesis ICPOES (Spectro Analytical Instruments GMBH; Kleve, Germany).

1. A. Al Zaki, D. Joh, Z. L. Cheng, A. L. B. De Barros, G. Kao, J. Dorsey and A. Tsourkas, *Acs Nano*, 2014, **8**, 104-112.
2. J. Park, K. J. An, Y. S. Hwang, J. G. Park, H. J. Noh, J. Y. Kim, J. H. Park, N. M. Hwang and T. Hyeon, *Nat Mater*, 2004, **3**, 891-895.

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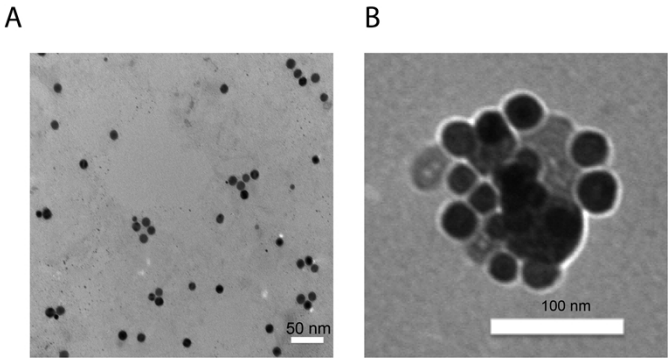

Figure S1. Transmission electron microscopy (TEM) image of individual hydrophobic SPIO (A) and a single SPIO-loaded phospholipid micelle (B).

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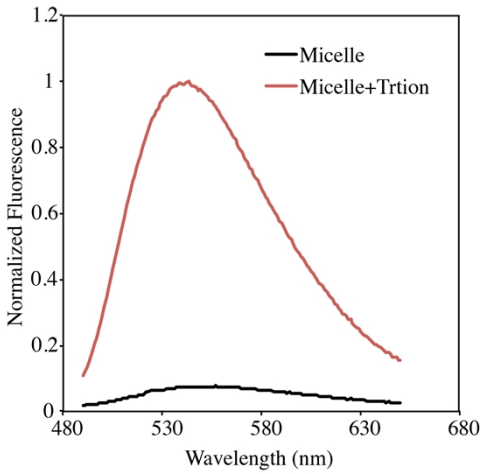

Figure S2. Fluorescence quenching of NBD-PC in a micellar formation. The study was done at the NBD-PC concentration of 1.67 µg/mL. The normalized fluorescence quenching was determined by subsequently lysing the micelles with Triton-X and measuring the total unquenched fluorescence intensity of the NBD-PC.