JACS
JOURNAL OF THE AMERICAN CHEMICAL SOCIETY
Communication
pubs.acs.org/JACS

Facile Surface Modification of Hydroxylated Silicon Nanostructures Using Heterocyclic Silanes

Dokyoung Kim,[†,∥] Jonathan M. Zuidema,[†,∥] Jinyoung Kang,[‡,∥] Youlin Pan,[§] Lianbin Wu,[†,⊥] David Warther,[†] Barry Arkles,*[,§] and Michael J. Sailor*[,†,‡]

[†]Department of Chemistry and Biochemistry, University of California, San Diego, La Jolla, California 92093, United States
[‡]Department of Nanoengineering, University of California, San Diego, La Jolla, California 92093, United States
[§]Gelest Inc., 11 East Steel Road, Morrisville, Pennsylvania 19067, United States
[⊥]Key Laboratory of Organosilicon Chemistry and Material Technology, Hangzhou Normal University, Hangzhou 311121, China

Supporting Information

ABSTRACT: Heterocyclic silanes containing Si−N or Si−S bonds in the ring undergo a ring opening reaction with −OH groups at the surface of porous Si nanostructures to generate −SH or −NH functional surfaces, grafted via O−Si bonds. The reaction is substantially faster (0.5−2 h at 25 °C) and more efficient than hydrolytic condensation of trialkoxysilanes on similar hydroxy-terminated surfaces, and the reaction retains the open pore structure and photoluminescence of the quantum-confined silicon nanostructures. The chemistry is sufficiently mild to allow trapping of the test protein lysozyme, which retains its enzymatic activity upon release from the modified porous nanostructure.

Electrochemical or chemical syntheses of porous silicon (pSi) or silicon nanoparticles usually produce hydrogen-terminated surfaces.[1,2] Driven by the desire for a functional and stable interface, the reactive Si−H surface is often modified postsynthesis using silicon−carbon (Si−C) or silicon−oxygen (Si−O) bond forming reactions.[3] The most common means to functionalize these materials is to graft organotrialkoxy-silanes[4] such as R-Si(OEt)$_3$ or R-Si(OMe)$_3$ to the Si−O modified surfaces.[2,5] These organotrialkoxysilane reagents react with hydroxyl-rich surfaces via hydrolytic condensation, and they provide convenient routes into amine- (R = 3-aminopropyl) or thiol- (R = 3-mercaptopropyl) functionalized surfaces[6,7] that are used in various sensor, energy, and biomedical applications.[1]

Despite their utility, trialkoxysilane reagents have limitations. They can require long reaction times or elevated temperatures to obtain efficient coverage, the alcohol or water byproducts can be deleterious to the performance of the final product, and they can undergo cross-linking reactions that result in overly thick coatings or clogging of micro- or mesopores.[2,8] Monoalkoxysilanes (such as (3-aminopropyl)-dimethylmethoxysilane) are not susceptible to cross-linking, but their reaction rates, coupling efficiency, and reaction byproducts can still be limiting for many purposes.[2]

In this work, we describe the use of 5-membered heterocyclic compounds containing silicon−sulfur (Si−S) or silicon−nitrogen (Si−N) motifs in the ring, which undergo a facile ring-opening reaction to modify hydroxyls[9−11] at the surface of silicon nanostructures (Figure 1). The reaction is demonstrated on films, microparticles, and nanoparticles of porous silicon (pSi). We refer to the method as a "ring-opening click" reaction due to its combination of simplicity, high yield, wide scope of applicability, lack of byproducts, and use of easily removable solvents.

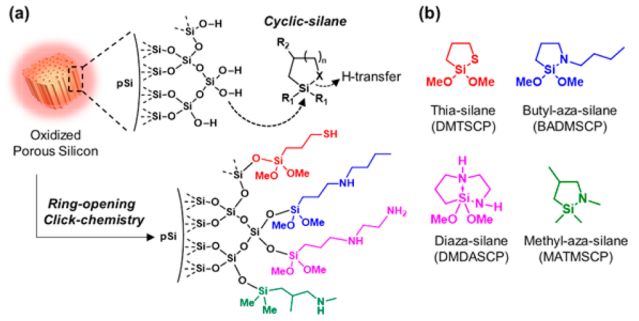


Figure 1. (a) Schematic illustration of the ring-opening click-reaction of cyclic-silanes with the silanol-terminated pore walls of oxidized porous silicon (pSi). (b) Structures of the reagents used in this study: thia-silane (DMTSCP, 2,2-dimethoxy-1-thia-2-silacyclopentane), butyl-aza-silane (BADMSCP, *N*-*n*-butyl-aza-2,2-dimethoxy-silacyclopentane), diaza-silane (DMDASCP, 2,2-dimethoxy-1,6-diaza-2-silacyclooctane), and methyl-aza-silane (MATMSCP, *N*-methyl-aza-2,2,4-trimethyl-silacyclopentane). R_1 = OMe, Me. R_2 = H, Me.

The pSi used in these experiments was a frame-sheath type of structure, consisting of a crystalline Si framework coated with a hydrophilic, hydroxylated SiO_2 sheath, prepared by treating as-etched pSi with oxidant (deionized water or aqueous hydrogen peroxide) at room temperature. Reaction of dichloromethane (DCM) solutions of the cyclic silanes with the hydroxylated-surface pSi nanoparticles (pSiNPs)[12] proceeded to completion within 1−2 h. The reaction was monitored by observing the change in surface charge on the nanoparticles. The pSiNP starting material displayed a negative ζ-potential (in the range −24 to −28 mV, Figure 2a, Table S1) consistent with a hydroxylated surface, and the particles displayed average hydrodynamic diameters of ∼200 nm (Figure 2b).

Received: August 17, 2016
Published: November 7, 2016

DOI: 10.1021/jacs.6b08614
J. Am. Chem. Soc. 2016, 138, 15106−15109

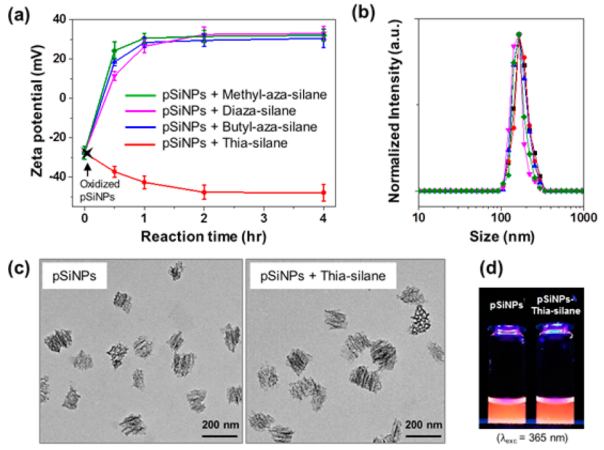


Figure 2. (a) ζ-potential and (b) mean hydrodynamic diameter (intensity distribution) of surface-oxidized pSiNPs, measured by dynamic light scattering (DLS), as a function of time of reaction with the indicated cyclic silanes (diluted 1:4 v:v into dichloromethane) at 25 °C. Particles were isolated at the indicated time points, rinsed, and redispersed in deionized water for the measurement. Means and standard deviations calculated from triplicate measurements. (c) Transmission electron microscopy (TEM) images of pSiNPs before and after reaction with thia-silane at 25 °C for 2 h. (d) Photograph of nanoparticles from (c) dispersed in ethanol and viewed under 365 nm illumination.

The cyclic azasilanes (butyl-azasilane, diazasilane, and methyl-azasilane) generate primary and secondary amines upon ring-opening, and these reagents increased the ζ-potential of the nanoparticles to between +30 and +35 mV (Figure 2a, Table S1). By contrast, ring-opening click of the cyclic thia-silane exposes a thiol functionality, and a decrease in ζ-potential, to −50 mV, was observed from particles subjected to this grafting reaction. The chemistry did not result in a significant increase in nanoparticle size, and no aggregates were observed (Figure 2b). Similar trends were observed when the reaction was performed under mild heating conditions (37 °C) (Figure S1). Transmission electron microscopy (TEM) images indicated that the open pore structure of the nanoparticles was preserved after modification (Figure 2c, Figure S2).

We compared the extent of reaction of cyclic silanes to the extent of reaction of triethoxy-/monoethoxy-silanes[13] with porous Si particles, using ζ-potential, thermogravimetric analysis (TGA), and attenuated total reflectance Fourier transform infrared (ATR-FTIR) spectroscopy. The alkoxy-silane grafting reactions were carried out in either dichloro-methane or the more common ethanolic solvent conditions, and the results indicated that the hydrolytic condensation reaction generally required greater time and resulted in lower surface coverage relative to the ring-opening click reaction (Figure S3, S4, S5). The TGA data indicated the ring-opening reactions gave ∼8% attached silane by mass, whereas the alkoxysilane reactions gave 2−4% attached silane by mass (Table S2, Figure S6). We attribute the greater efficiency of the heterocyclic coupling reaction primarily to the lower energies of Si−N and Si−S bonds (relative to Si−O) and secondarily to the relief of ring strain from the cyclic reactant. Both of these factors provide a stronger driving force relative to hydrolysis, resulting in faster and more extensive surface coupling.

Next, we investigated the influence of solvent on the extent and rate of the reaction. The changes in ζ-potential were more pronounced in the polar aprotic solvents dichloromethane and dimethyl sulfoxide than in less polar diethyl ether (Figure S7). A ^{1}H NMR study (Figure S8) indicated that the ring-opening click reactions proceed cleanly, with no significant side-products. The ζ-potential values of the pSiNP products displayed little sensitivity to concentration of cyclic silane used in the reaction (Figure S9).

The attenuated total reflectance Fourier-transform infrared (ATR-FTIR) spectrum of the pSi starting material displayed strong bands at 1020 cm^{-1} associated with Si−O stretching modes from the oxide layer of the Si−SiO_2 frame−sheath structure, and stretching and bending modes at 3300 and 1640 cm^{-1}, respectively, assigned to a combination of surface O−H and adsorbed water (Figure S10). After reaction with the cyclic silane reagents, the modified pSi material displayed bands that could be assigned to C−H stretching and bending modes of the grafted reagents. These spectral bands persisted even after extensive rinsing of the materials, indicating that the species were chemisorbed to the pSi surface. The modes associated with surface O−H and adsorbed water on the starting pSi substrate were significantly decreased after the grafting chemistry, indicating high surface coverage of the particle surface including the inner pore walls. Control experiments using samples that were prepared by thermal oxidation of pSi at high temperature (to dehydrate surface Si−OH to Si−O−Si) showed no substantial grafting (Figure S11), demonstrating the importance of surface Si−OH for the reaction to proceed. Additionally, controls using as-prepared pSi samples that contained little surface oxide showed a much lower degree of grafting, and the organosilane species were removed by briefly rinsing the treated surface with ethanolic aqueous HF solution (Figure S12). This type of rinse readily removes Si−O bonded or physisorbed species, whereas it does not remove Si−C bonded surface species.[2] These results also support the importance of surface Si−OH for the ring-opening click reaction.

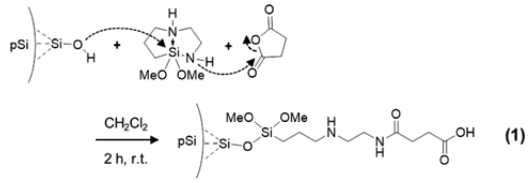
Nitrogen adsorption−desorption isotherm analysis of the modified pSiMPs revealed slight decreases in surface area, total pore volume, and average pore diameter relative to the unmodified material (Figure S13, Table S3). The BJH surface area and TGA results yielded surface coverage values of 1.1 × 10^{14} molecules cm^{-2} for thia-silane, and 1.3 × 10^{14} molecules cm^{-2} for methyl-aza-silane. Powder X-ray diffraction (XRD) measurements showed that the crystallinity of the silicon skeleton was retained in the modified particles; Debye−Scherrer analysis indicated a mean crystallite size of ∼2.2 nm, Figure S14). Elemental analysis confirmed the presence of sulfur and nitrogen in the thia-silane and methyl-aza-silane-modified materials, respectively (Table S4).

The surface wettability of the modified pSi material was analyzed by water contact angle measurements made on suitably modified pSi films (Figure S15). The products of reaction with thia-silane or diaza-silane, which generate terminal thiol and primary amine moieties, respectively, displayed low contact angles (<16°) characteristic of hydrophilic surfaces. The butyl-aza-silane and methyl-aza-silane reagents generated a more hydrophobic surface (water contact angles of 88° and 73°, respectively), as expected from the more hydrophobic *N*-methyl and *N*-butyl terminal species that were generated.[14]

The cyclic thia-silane and aza-silane reagents have an advantage over alkoxysilane reagents in that the thiol or primary amine functional species are only exposed when the reagent undergoes ring-opening with surface −OH groups. The cryptic nature of these functional groups presents the possibility

DOI: 10.1021/jacs.6b08614
J. Am. Chem. Soc. 2016, 138, 15106−15109

for a one-pot tandem synthesis involving common thiol- or amine- coupling agents. To demonstrate this feature, we performed the diaza-silane surface modification reaction in a mixture containing succinic anhydride (eq 1). NMR measure-



(1)

ments confirmed that the diaza-silane reagent does not react with succinic anhydride on its own under the reaction conditions used. However, when hydroxylated-surface pSiNPs were present, the ring-opening click reaction generated a primary amine at the surface, which then coupled to succinic anhydride *in situ* to form the tandem product shown in eq 1. The reaction was confirmed by ζ-potential measurements and ATR-FTIR analysis (Figure S16, S17), and the data are consistent with the formation of the expected amide linker and terminal carboxylic acid functionality.

To test the effect of the surface modification on the photoluminescence (PL) of quantum-confined pSi, we prepared a pSiNP formulation that displayed strong PL (λ_{ex} = 365 nm, λ_{em} = 780 nm, external quantum yield ~ 23%, Figure 2d, S18).[15] Silicon quantum dots are useful for bioimaging,[16−19] and there is a need for mild surface modification reactions that do not destroy the PL properties.[20] The surface modification reaction was found to preserve more than 90% of the original PL intensity (based on an equivalent mass of nanoparticles), with no substantial shift in the emission wavelength.

Finally, we tested the compatibility of the surface modification chemistry with a protein payload. pSiNPs have shown utility as delivery vehicles for therapeutic cargos[21−25] that can protect sensitive biologics from denaturing *in vitro* or *in vivo*,[26,27] and there is a need for chemistries that can be used to attach targeting or biocompatibility agents while preserving the integrity of the therapeutic payload.[28] The model protein lysozyme was used (Table S5), and it was loaded into the pSiNPs from aqueous solution (0.1 mg of lysozyme-loaded pSiNPs, mass loading of lysozyme 41%). The particles were separated from the aqueous loading solution by centrifugation, but no attempt was made to remove residual water from the mesopores. The particles were then dispersed into either *n*-hexane or DCM solutions containing the thia-silane reagent (Figure 3a, S19). The surface modification was confirmed by ζ-potential measurements (Table S1), where the relatively large positive charge on the lysozyme-loaded particles dropped to negative values after thia-silane grafting due to the generation of surface thiol species. The net change in surface charge was comparable to that observed on empty particles before and after thia-silane modification. The surface chemistry did not significantly impede the rate of lysozyme release from the pSiNPs (Figure 3b); 30−40% of the protein payload was released into 37 °C aqueous PBS solution within 72 h. The rate of lysozyme release did not depend strongly on the solvent used (*n*-hexane or DCM) for the chemical modification step. However, the modification solvent exerted a strong influence on activity of the released protein; lysozyme released from material modified in DCM retained 98% of its activity, whereas when the chemistry was performed in *n*-hexane, the released lysozyme showed only 72% activity. This is consistent with

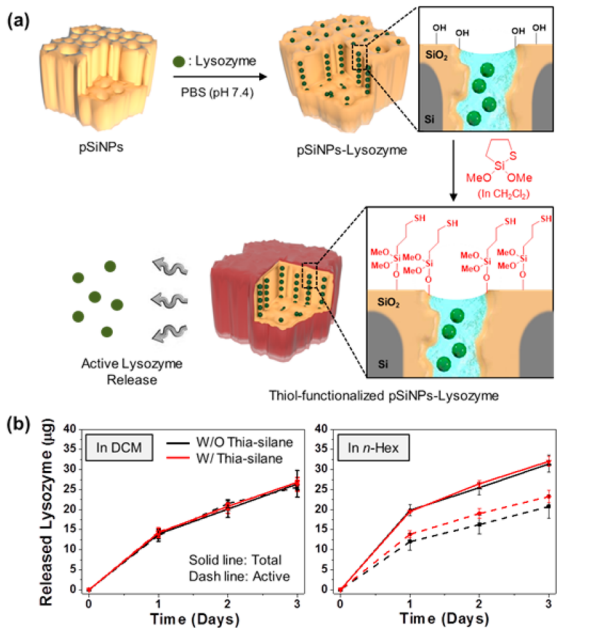


Figure 3. (a) Procedure used to load lysozyme into pSiNPs and modify the resulting particles using the ring-opening click reaction. The pSiNPs are loaded with lysozyme from aqueous solution, and the click reaction is run in DCM or *n*-hexane to decorate the outer perimeter of the nanoparticles. Immersion in aqueous buffer at 37 °C induces slow release of the protein payload. (b) Micrograms of total lysozyme released (BCA assay) from unmodified pSiNPs (black line) and thia-silane functionalized pSiNPs (red line). Dashed lines show micrograms of active lysozyme released (lysozyme activity assay). Total protein loaded was ~76 μg. "In DCM" and "In *n*-Hex" denote solvent in which the ring-opening click reactions were run. Release experiments were performed in PBS buffer at 37 °C.

control experiments in which lysozyme-loaded pSiNPs were exposed to the two solvents; after removal of the solvents, the lysozyme released into PBS showed 96% and 66% activity for DCM and *n*-hexane, respectively (Figure 3b). The results demonstrate that the modification chemistry does not react with or otherwise denature the protein payload, presumably due to the immiscibility of the organic solvents with the hydrated protein payload contained within the particles.

By contrast, when alkoxysilane chemistry (3-mercaptopropyltriethoxysilane, MPTES) was used to modify lysozyme-loaded pSiNPs (using ethanol solvent), the lysozyme released from the MPTES-modified pSiNPs retained only 68 ± 4% activity (Table S5). Furthermore, the MPTES chemistry substantially impeded the rate of release of the protein (by a factor of 2 relative to unmodified pSiNPs).

In summary, ring-opening click chemistry with cyclic silanes provides an efficient, mild, and experimentally convenient method to modify the surface of hydroxylated porous silicon films, microparticles, or nanoparticles. The chemistry proceeds via attack of surface −OH species at the cyclic silane, inducing ring-opening with no byproducts. The room-temperature reaction retains the pore structure and intrinsic photoluminescence of the Si nanomaterial, and it is amenable to tandem functionalization, where a cryptic thiol or primary amine revealed during the ring-opening reaction can couple *in situ* with a second functional species such as succinic anhydride. This nonaqueous chemistry is sufficiently mild that it does not alter the observed activity of a protein contained within the hydrated porous interior of the material.

DOI: 10.1021/jacs.6b08614
J. Am. Chem. Soc. 2016, 138, 15106−15109

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/jacs.6b08614.

Detailed experimental procedures, supporting figures and tables (PDF)

■ AUTHOR INFORMATION

Corresponding Authors

*M.J.S. msailor@ucsd.edu

*B.A. executiveoffice@gelest.com

ORCID

Dokyoung Kim: 0000-0002-7756-3560

Michael J. Sailor: 0000-0002-4809-9826

Author Contributions

‖These authors contributed equally.

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

This work was supported by the National Science Foundation under Grants No. DMR-1210417, CBET-1603177, and by the Defense Advanced Research Projects Agency (DARPA) under Cooperative Agreement HR0011-13-2-0017. The content of the information within this document does not necessarily reflect the position or the policy of the Government. D. Kim thanks the UCSD FISP program and the Basic Science Research Program of the Korean NRF, funded by the Ministry of Education (2016R1A6A3A03006343) for fellowships.

■ REFERENCES

(1) Canham, L. T. *Handbook of Porous Silicon*; Springer: Switzerland, 2014.

(2) Sailor, M. J. *Porous Silicon in Practice: Preparation, Characterization, and Applications*; Wiley-VCH: Weinheim, Germany, 2012; p 249.

(3) Buriak, J. M. *Chem. Rev.* **2002**, *102*, 1271.

(4) Pasternack, R. M.; Amy, S. R.; Chabal, Y. J. *Langmuir* **2008**, *24*, 12963.

(5) Riikonen, J.; Salomaki, M.; van Wonderen, J.; Kemell, M.; Xu, W.; Korhonen, O.; Ritala, M.; MacMillan, F.; Salonen, J.; Lehto, V. P. *Langmuir* **2012**, *28*, 10573.

(6) Nijdam, A. J.; Cheng, M. M. C.; Geho, D. H.; Fedele, R.; Herrmann, P.; Killian, K.; Espina, V.; Petricoin, E. F.; Liotta, L. A.; Ferrari, M. *Biomaterials* **2007**, *28*, 550.

(7) Williams, R. L.; Hadley, M. J.; Jiang, P. J.; Rowson, N. A.; Mendes, P. M.; Rappoport, J. Z.; Grover, L. M. *J. Mater. Chem. B* **2013**, *1*, 4370.

(8) Acres, R. G.; Ellis, A. V.; Alvino, J.; Lenahan, C. E.; Khodakov, D. A.; Metha, G. F.; Andersson, G. G. *J. Phys. Chem. C* **2012**, *116*, 6289.

(9) Arkles, B.; Pan, Y.; Larson, G. L.; Berry, D. H. Cyclic azasilanes: Volatile coupling agents for nanotechnology. In *Silanes and Other Coupling Agents*; Mittal, K. L., Ed.; CRC Press: Boca Raton, FL, 2004; Vol. 3, p 179.

(10) Ju, L.; Strandwitz, N. C. *J. Mater. Chem. C* **2016**, *4*, 4034.

(11) Maddox, A. F.; Matisons, J. G.; Singh, M.; Zazyczny, J.; Arkles, B. *MRS Online Proc. Libr.* **2015**, *1793*, 35.

(12) Qin, Z.; Joo, J.; Gu, L.; Sailor, M. J. *Part. Part. Syst. Charact.* **2014**, *31*, 252.

(13) Joo, J.; Liu, X.; Kotamraju, V. R.; Ruoslahti, E.; Nam, Y.; Sailor, M. J. *ACS Nano* **2015**, *9*, 6233.

(14) De Stefano, L.; Oliviero, G.; Amato, J.; Borbone, N.; Piccialli, G.; Mayol, L.; Rendina, I.; Terracciano, M.; Rea, I. *J. R. Soc., Interface* **2013**, *10*, 20130160.

(15) Park, J.-H.; Gu, L.; von Maltzahn, G.; Ruoslahti, E.; Bhatia, S. N.; Sailor, M. J. *Nat. Mater.* **2009**, *8*, 331.

(16) Gongalsky, M. B.; Osminkina, L. A.; Pereira, A.; Manankov, A. A.; Fedorenko, A. A.; Vasiliev, A. N.; Solovyev, V. V.; Kudryavtsev, A. A.; Sentis, M.; Kabashin, A. V.; Timoshenko, V. Y. *Sci. Rep.* **2016**, *6*, 24732.

(17) Osminkina, L. A.; Sivakov, V. A.; Mysov, G. A.; Georgobiani, V. A.; Natashina, U. A.; Talkenberg, F.; Solovyev, V. V.; Kudryavtsev, A. A.; Timoshenko, V. Y. *Nanoscale Res. Lett.* **2014**, *9*, 463.

(18) Cheng, X.; Lowe, S. B.; Ciampi, S.; Magenau, A.; Gaus, K.; Reece, P. J.; Gooding, J. J. *Langmuir* **2014**, *30*, 5209.

(19) Liu, J. W.; Erogbogbo, F.; Yong, K. T.; Ye, L.; Liu, J.; Hu, R.; Chen, H. Y.; Hu, Y. Z.; Yang, Y.; Yang, J. H.; Roy, I.; Karker, N. A.; Swihart, M. T.; Prasad, P. N. *ACS Nano* **2013**, *7*, 7303.

(20) Kotkovskiy, G. E.; Kuzishchin, Y. A.; Martynov, I. L.; Chistyakov, A. A.; Nabiev, I. *Phys. Chem. Chem. Phys.* **2012**, *14*, 13890.

(21) Liu, D. F.; Bimbo, L. M.; Makila, E.; Villanova, F.; Kaasalainen, M.; Herranz-Blanco, B.; Caramella, C. M.; Lehto, V. P.; Salonen, J.; Herzig, K. H.; Hirvonen, J.; Santos, H. A. *J. Controlled Release* **2013**, *170*, 268.

(22) Tasciotti, E.; Liu, X. W.; Bhavane, R.; Plant, K.; Leonard, A. D.; Price, B. K.; Cheng, M. M. C.; Decuzzi, P.; Tour, J. M.; Robertson, F.; Ferrari, M. *Nat. Nanotechnol.* **2008**, *3*, 151.

(23) Henstock, J. R.; Canham, L. T.; Anderson, S. I. *Acta Biomater.* **2015**, *11*, 17.

(24) Canham, L. T. Porous silicon for medical use: from conception to clinical use. In *Porous Silicon for Biomedical Applications*; Santos, H. A., Ed.; Elsevier: Cambridge, 2014; p 3.

(25) Yong, K. T.; Law, W. C.; Hu, R.; Ye, L.; Liu, L. W.; Swihart, M. T.; Prasad, P. N. *Chem. Soc. Rev.* **2013**, *42*, 1236.

(26) Wu, C.-C.; Hu, Y.; Miller, M.; Aroian, R. V.; Sailor, M. J. *ACS Nano* **2015**, *9*, 6158.

(27) Tanaka, T.; Mangala, L. S.; Vivas-Mejia, P. E.; Nieves-Alicea, R.; Mann, A. P.; Mora, E.; Han, H. D.; Shahzad, M. M. K.; Liu, X. W.; Bhavane, R.; Gu, J. H.; Fakhoury, J. R.; Chiappini, C.; Lu, C. H.; Matsuo, K.; Godin, B.; Stone, R. L.; Nick, A. M.; Lopez-Berestein, G.; Sood, A. K.; Ferrari, M. *Cancer Res.* **2010**, *70*, 3687.

(28) Salonen, J. Drug Delivery With Porous Silicon. In *Handbook of Porous Silicon*; Canham, L. T., Ed.; Springer: Switzerland, 2014; p 909.

DOI: 10.1021/jacs.6b08614
J. Am. Chem. Soc. 2016, 138, 15106−15109