Supporting Information

Facile silicification of plastic surface for bioassays

Seonki Hong, Ki Soo Park, Ralph Weissleder, Cesar M. Castro, Hakho Lee

Experimental section

Polydopamine-assisted silica coating: Polyethylene (PE, 11-13 μ m) and polytetrafluoroethylene beads (PTFE, 13-21 μ m) were kindly donated by Micro Powders, polystyrene beads (PS) were purchased from Sigma Aldrich (15 μ m), and poly(methyl methacrylate) beads (PMMA) microbeads were purchased from Bangs Laboratories (14.74 μ m, copolymerized with 5% divinylbenzene (DVB)). PMMA and PS microbeads dispersed in 1XPBS (40 mg/mL, pH 7.4) were mixed with the same volume of 1xPBS containing 4 mg/mL of dopamine (pH 8.3, adjusted by adding 1 M NaOH), then the mixture was stirred by hula-mixer for 1 hr at room temperature. PE and PTFE were dispersed in ethanol instead of 1XPBS (40 mg/mL), then mixed with the same volume of 1XPBS in pH 8.3 containing 4 mg/mL of dopamine (DA, Sigma Aldrich), then stirred by hula-mixer for overnight at room temperature. After the reaction, polydopamine-coated beads (pDA-beads) were collected by centrifuge at 1,500 g for 5 min, and washed with distilled water (PMMA, PS) or ethanol (PE, PTFE) for five-times, then lyophilized. For silica coating, monosilicic acid solution was prepared by adding 40 μ L of 1 M HCl (final 1 mM) and 595.2 µL of Tetramethyl orthosilicate (TMOS, final 100 mM, Sigma Aldrich) to 40 mL of 1XPBS, then stirred for 15 min at room temperature prior to use. 500 mg of pDA-beads were re-dispersed in 18 mL of 1XPBS (PMMA, PS) or ethanol (PE, PTFE), and mixed with 18 mL of prepared monosilicic acid solution, followed by stirred for 1 hr at room temperature. After the reaction, silica-coated beads were collected and washed with distilled water (PMMA, PS) or ethanol (PE, PTFE) for five-times by centrifuge (300 g for 3 min), then lyophilized.

Characterization of silica coated beads: The morphological change of PMMA after pDA and silica coating was determined by using HR-SEM (high-resolution scanning electron microscope, Zeiss Merlin). Beads were coated with 20 nm of gold or 10 nm of carbon (for EDS) using a Denton Vacuum DV- 502A evaporator before imaging. Elemental analysis on top layer (~10 nm) of unmodified and modified beads was performed using a Model PHI5000 Versaprobe II X-ray photoelectron spectrometer, manufactured by ULVAC-PHI, which has a monochromated aluminum source and hemispherical analyzer. The operating pressure during analysis was approximately 6.0e-9 Torr, the Al X-ray source power was 50 W, the X-ray energy was 1486.6 eV, and the analysis area was 200 μ m. The take-off angle to the analyzer was 45 degree, and the pass energy for high energy resolution spectra was 23.5 eV. As these samples were insulating, spectra were acquired using PHI's dual-beam neutralizer. For FT-IR analysis, bare, pDA-, and pDA-Si coated PMMA was measured using a Thermo Fisher Continuum FT-IR spectrometer after lyophilization. For BET surface area and BJH pore distribution analysis, nitrogen adsorption-desorption isotherms were obtained by a Tristar 3020 Micromeritices (USA) analyzer at 77K. Spectroscopic ellipsometry (Gaertner Scientific Co., IL) was

used to determine the thickness of pDA and SiO₂ coated on a Si wafer with a HE-Ne laser (632.8 nm) at a 70° angle of incidence.

RNA Extraction column preparation: 50 mg of silica-coated beads (dispersed in ethanol) were added to a filter column (PierceTM spin cups-paper filter, 10 μ m pore) followed by centrifuged at 9,000 g for 30 sec. Beads packed in the column were serially washed with RNase-awayTM decontamination reagent (Life Technologies), RNase-free water, and ethanol by centrifuge at 9,000 g for 30 sec, respectively.

Cell culture and exosome isolation: A2780, OVCAR3, OV420, and TIOSE4 cells were cultured in RPMI 1640 medium (Cellgro) with L-glutamine, 10% (v/v) fetal bovine serum (FBS, Cellgro) and 1% (v/ v) penicillin-streptomycin (Cellgro). Cells were incubated at 37 °C in a humidified atmosphere of 95% room air and 5% CO₂ until they reached 70 to 80% confluency, and trypsinized followed by washed with PBS. The cell pellets (1 x 107 cells) were then stored at -80 °C until RNA extraction. For exosome collection, culture medium was removed, and cells in the culture dish (150 mm in diameter) were gently washed with 1XPBS, and then RPMI 1640 with 5% (v/v) exosome-depleted FBS was added when cells reached 70 to 80% confluency. After 48 hr incubation, culture medium from 12 culture dishes was collected, filtered through a 0.22 μ m filter (cellulose acetate membrane, Corning), and concentrated by ultracentrifugation (24,200 rpm for 70 min at 4 °C). After the supernatant was removed, the exosome pellet was washed with 1x PBS and centrifuged at 24,200 rpm for 70 min 4 °C. The resulting exosome pellet was stored at -80 °C until RNA extraction. All bacteria were purchased from the American Type Culture Collection (ATCC). Bacterial cultures were grown to mid-log phase in vendor-recommended medium: S. aureus (#25923) in Staphylococcus broth (BD Biosciences); E.coli (#25922) in LB medium (BD Biosciences): MRSA (#BAA-1720) and *P. aerugionsa* (#142) in tryptic sov broth (BD Biosciences). Bacteria were collected via centrifugation (6000 g, 10 min) and washed with 1x PBS. The pellets (10⁸ CFU) were stored at -80 °C until RNA extraction.

RNA extraction and quantification: Cells (1 × 10⁷ cells) were lysed in 700 μ L of RLT lysis buffer from Qiagen RNeasy mini kit and then mixed with 490 μ L of ethanol before added to a spin-column containing 50 mg of silica-coated beads. 120 μ L of prepared lysates was added/filtered on the column by centrifuge (9,000 rpm for 30 sec) to remove cellular debris. Then, the beads were washed with commercial buffer set from Qiagen RNeasy mini kit (once with 400 μ L of RW1 buffer, twice with 300 μ L of RPE buffer) by centrifuge at 9,000 rpm for 30 sec, then completely dried by centrifuge at 14,000 g for 2 min. RNAs captured on silica-coated beads were finally collected in a fresh tube by adding 50 μ L of RNase-free water followed by centrifuge at 12,000 g for 1 min (repeated twice if total RNA was over 30 μ g). Extracted RNA was quantified by using a Nanodrop 1000 spectrophotometer (Thermo Scientific) and further investigated with 2100 Bioanalyzer (Agilent) using a RNA 6000 Nano chip.

mRNA analysis by quantitative reverse transcription PCR (RT-qPCR): Single-stranded cDNA was synthesized by using High-capacity cDNA reverse transcription kit (Applied Biosystems) from 300 ng of extracted RNA. PCR products were quantitatively amplified from cDNA by using a Taqman gene expression master mix (Applied Biosystems) and two unlabeled oligonucleotide primers with a FAM[™] dye-labeled probe with a minor groove binder (MGB) moiety (Taqman gene expression assays, Applied Biosystems) following the manufacturer's protocol. qPCR was performed by 1 cycle of 95 °C for 10 min

and 40 cycles of 95 °C for 15 sec followed by 60 °C for 1 min with an ABI 7500 Fast Real-Time PCR system (Applied Biosystems). All analysis were triplicated, and mRNA levels were normalized against the GAPDH expression.

Reverse transcription loop-mediated isothermal amplification (RT-LAMP) of RNA: The primer sequences specific to *nuc* and *mecA* genes were previously designed¹ as shown in **Table S1**. The 5' end of the forward and backward loop primers (FLP and BLP) were labeled with fluorescein isothiocyanate (FITC) and biotin, respectively, for rapid detection on lateral flow immunoassay strips (PCRD nucleic acid immunoassay, TwistDX) after RT-LAMP of RNA. RT-LAMP reaction was performed following the manufacturer's procedure (All chemicals except for primers were purchased from New England Biolabs). Briefly, 25 μ L of the reaction mixture was first prepared with 50 pg of RNA, 1× isothermal amplification buffer, 1.4 mM dNTPs, 8 mM MgSO₄, 0.32 U Bst 2.0 DNA polymerase, 0.5 μ L WarmStart RTx reverse transcriptase, and the primer set (custom synthesized from Integrated DNA Technologies, mixture of 1.6 μ M each of forward and backward inner primers (FIP and BIP), 0.2 μ M each of forward and backward outer primers (F3 and B3) and 0.4 μ M each of loop primers (FLP and BLP)). The mixture was then kept at 65 °C for 15 min. For the visualized detection of the amplified products on the test strip, 5 μ L of amplicons were diluted into 70 μ L of PCRD extraction buffer (TwistDX), followed by applied on the sample port of the test strip. The assay was finally read-out after 10 min incubation at room temperature.

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		BJH desorption		BJH desorption	Ý- B
Samples	BET surface area (m²/g)	Average pore diameter (nm)	Surface area (m²/g) (pores: 1.7 - 300 nm)	⁽ ^c uuu) 0 2-	
PMMA	0.259	n.d.	n.d.		
PMMA@pDA	0.282	n.d.	n.d.	ore	
PMMA@pDA/Si	2.188	11.717	0.501	<u> </u>	
				Pore diamete	

Fig. S1. Surface area and pore size analysis. (a) Surface area and pore size were measured by Brunauer-Emmett-Teller (BET) and Barret-Joyner-Halenda (BJH) methods, respectively. The pores on bare and pDA-coated PMMA beads were not detectable (n.d.). (b) The pore size distribution (BJH desorption cumulative pore volume) of PMMA@pDA/Si microbeads shows two portions of nanopores, below 10 nm and around 45 nm in diameter.



Fig. S2. Elemental analysis of pDA/Si-coated PMMA microbeads. (a) Si element was detected at 1.74 keV in energy-dispersive X-ray spectrum. **(b)** Elemental mapping of C, O, Si, and N.



Fig. S3. High resolution X-ray photoelectron spectroscopy (XPS) data of bare, pDA-coated, and pDA/ Si-coated PMMA beads.



Fig. S4. Fourier transform infrared spectra of bare, pDA-coated, and pDA/Si-coated PMMA. Characteristic peaks of SiO₂ were detected after the pDA/Si coating.



Fig. S5. A disposable in-flow column for use with a bench-top centrifuge



Fig. S6. Comparison of mRNA level isolated by pDA/Si-coated PE and a commercial column from Qiagen.



Table S1. Assay procedures of centrifugal filters (RNeasy mini kit from Qiagen and the column device) and a cartridge with syringe.

RNA extraction method	Cost per sample	Washing steps	Time required	A260/A280	A260/A230	Yield (10 ⁶ cells)
RNeasy mini kit	6	15 sec centrifuge	30 min	2.14 ± 0.058	2.16 ± 0.056	5.4 ± 0.76 μg
Syringe device (50 mg PE@pDA/Si)	\$3 (device \$2)	1 sec simple injection	10 min	2.14 ± 0.047	2.11 ± 0.147	4.9 ± 0.50 μg

Table S2. Comparison of a commercial kit from Qiagen (RNeasy mini kit) and a syringe device.

	nuc	mecA
F3	TCGCTTGCTATGATTGTGG	GGTACAAGATGATACCTTCGTT
В3	ACATACGCCAATGTTCTACC	ATAGCAGTACCTGAGCCAT
FIP	GTACAGTTTCATGATTCGTCCCGCCATCATTATTGTAGGTGT	TCTTCAGAGTTAATGGGACCAAACAGAAAGTCGTAACTATCCTC
BIP	TGTTCAAAGAGTTGTGGATGGTGTACAGGCGTATTCGGTT	AAGCTCCAACATGAAGATGGCTTGTATGTGCGATTGTATTGC
FLP	5'FAM-TTGAAAGGACCCGTATGATTCA	5'FAM-ACCTAATAGATGTGAAGTCGCT
BLP	5'Biotin-GATACGCCAGAAACGGTGA	5'Biotin-CGTGTCACAATCGTTGACG

Table S3. Primer sequences specific to *nuc* and *mecA* genes.¹ 5' end of FLP and BLP were modified by FITC and Biotin, respectively. (F3:Forward outer primer, B3: Backward outer primer, FIP: Forward inner primer, BIP: Backward inner primer, FLP: Forward loop primer, BLP: Backward loop primer)

Reference

1. S. Sudhaharan, L. Vanjari, N. Mamidi, N. EDE, L. Vemu, J. Clin. Diagn. Res., 2015, 9, DC06.