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Supporting Information for

L-carnitine Derived Zwitterionic Betaine Materials

Wei Wang, Jianhai Yang, Ershuai Zhang, Yang Lu and Zhiqiang Cao*

Materials

L-carnitine hydrochloride (>98%), anhydrous N,N-Dimethylformamide (DMF, 99.8%), hydroquinone (99%), triethylamine (TEA, 99.5%), anhydrous dichloromethane (DCM, 99.8%), anhydrous Diethyl ether (99%), copper(I) bromide (99.9%), bromoisobutyryl bromide (BIBB 98%), 11-mercapto-1-undecanol (97%), 2,2'-bipyridine (BPY 99%), and activated charcoal (DARCO, -100 particle size) were purchased from Sigma-Aldrich Chemical Company, Milwaukee. Anhydrous Methanol (99%) and acryloyl chloride (>96%) were obtained from Alfa Aesar, Thermo Fisher Scientific. Alcohol (200 proof) was purchased from Decon Laboratories, Inc.

Characterization of L-carnitineMA

The Nuclear magnetic resonance (NMR) spectrum of L-carnitineMA was measured using a Varian Mercury-400 MHz NMR, using the D_2O as the solvent. The areas under the peak were calculated using mestrec23 software. The mass spectrometry was conducted on a Water Micromass ZQ ion trap mass spectrometer in methanol as the solvent.

Preparation of L-carnitineMA polymer brush coating on gold substrate

SPR glass chip coated with a surface plasmon active gold layer was purchased from Institute of Photonics and Electronics, Czech Academy of Sciences, Czech Republic. The gold surface was washed with DI water and pure ethanol, and then it was placed in an O-zone cleaner and heated for 20 mins. After cleaning, the chip was soaked overnight in the pure ethanol containing 1 mM ATRP initiator ω-mercaptoundecyl bromoisobutyrate. The gold substrate was then rinsed with pure ethanol followed by THF and dried in a stream of nitrogen. The L-carnitineMA polymers were grown onto the gold substrate by atom transfer radical

polymerization (ATRP). Briefly, DI water and DMF were purged with nitrogen for 15 mins to remove oxygen. 21.4 mg CuBr and 46.25 mg 2,2'-Bipyridine (BPY), as well as the cleaned substrate, were added into a reaction tube treated with three vacuum/nitrogen cycles. 216 mg L-carnitineMA monomer (1 mmol) was added into another reaction tube treated with three vacuum/nitrogen cycles. 5 ml mixed solvent (DI water/DMF 2:1 volume ratio) was added to both tubes and the L-carnitineMA monomer solution was transferred to the tube containing the gold chip using a syringe. The tube was tightly sealed and the ATRP reaction was carried out with gentle stirring at room temperature for 3 h. After the reaction, the polymer coated substrate was immersed in DI water to remove unreacted monomer and other small molecules before the test.

Coating thickness characterization

After ATRP, the polymer brush thickness was measured by surface ellipsometry and atomic force microscope (AFM). The surface ellipsometry was performed on an ellipsometer (α-SETM, J.A. Woollam Co., Inc.) and the thickness was calculated by the average of three measurements of different coating sites with an assumed gold surface refractive index of 1.45. The AFM imaging of bare gold substrate and L-carnitineMA polymer brush-coated gold substrate were conducted on a Dimension 3100 AFM from VEECO. Samples were vacuum dried before imaging. An L-carnitineMA polymer brush coating was gently scratched with a sharp lancet to expose the coating section for thickness measurement. The coating thickness and morphology were measured in the air through the tapping mode using silicon probes (VEECO) with a nominal frequency of 150 kHz. The AFM images were analyzed using Nanoscope software version 5.12 (VEECO).

Measurement of protein adsorption by an SPR sensor

The Protein absorption on the bare gold substrate, the L-carnitineMA polymer brush-coated substrate, and the PCBMA polymer brush-coated substrate were measured using a surface plasmon resonance (SPR) sensor custom-built by Institute of Photonics and Electronics, Czech Academy of Sciences, Czech Republic. The tested chips were attached to the base of the prism.

A four-channel flow cell with four independent parallel flow channels was used to contain liquid samples during experiments. PBS buffer at 50 µl/min and 25°C was first used to obtain a stable baseline. Fibrinogen (from bovine plasma, Sigma) and lysozyme (from chicken egg white, Sigma) solutions of 1.0 mg/mL in PBS flowed over the surfaces at a flow rate of 0.05 mL/min for 10 min followed by flowing PBS to remove any loosely bound proteins. A surface-sensitive SPR detector was used to monitor protein-surface interactions in real time. Wavelength shift between the baselines before and after protein injection was used to measure the surface protein concentration (mass per unit area). For bare-gold SPR substrates, a 1 nm wavelength shift starting at a resonant wavelength of 750 nm represents 17 ng/cm^2 of absorbed proteins[1]. For L-carnitineMA polymer brush surface of 23 nm, a calibrator factor of 1.31 was calculated based on existing protocol[2]. Thus, for this specific L-carnitineMA polymer coated chip, a 1 nm shift in resonant wavelength represents 22.3 ng/cm^2 of protein coverage.

Preparation of L-carnitineMA hydrogel sample

The L-carnitineMA hydrogel was fabricated by UV initiation of L-carnitineMA monomer solution of 50 % (weight ratio) in the presence of 0.2% UV initiator 2-Hydroxy-4'-(2-hydroxyethoxy)-2 -methylpropiophenone (I-2959) and 5% crosslinker N,N'-Methylenebisacrylamide (MBAA). The pre-gel solution was filled in the space between two glass slides separated by a Teflon spacer with 1 mm thickness. The hydrogels were equilibrated in DI water to remove unreacted small molecules. Hydrogel samples were tailored into a disc shape (5 mm in diameter and 1 mm in thickness) using a punch for further evaluation.

Protein absorption on L-carnitineMA hydrogel

To measure the human fibrinogen (Fg, Sigma-Aldrich) adsorption on PCBMA (positive control), PHEMA (negative control) and L-carnitineMA hydrogel, the hydrogel samples were incubated with 1mg/ml Fg in a 24-well plate for 10 minutes at room temperature, followed by 5 washes with PBS buffer. L-carnitineMA hydrogels were then incubated with 1 mg/ml bovine serum albumin solution for 10 minutes at room temperature with 5 times wash again with PBS buffer. The tested hydrogel samples were then transferred to new wells. They were next incubated

with a 1:200-dilution of horseradish peroxidase (HRP)-conjugated anti-fibrinogen in PBS for 10 minutes, followed by another 5 washes with the same buffer. After the fifth wash, the tested hydrogels were transferred to new wells and SIGMAFAST OPD was added to each well at 30-second intervals. The samples were incubated in the OPD solution for 30 minutes away from light. The supernatant was removed from each test well, transferred to a 96-well plate, and its absorbance at 490 nm was measured. All samples were measured in triplicate.

Bacteria culture

The bacteria were cultured based on our prior experience[3]. *E.* coli K12 was cultured overnight at 37 °C on Luria-Bertani (LB) agar plates. One colony was picked and transferred to the culture in 25 mL of LB medium (20 g/L) overnight under 37 °C at 300 rpm shaking rate. The bacteria culture was used to inoculate a second culture in 150 ml of LB medium. The new culture was continuously shaken under 37 °C until it reached an optical density of 0.9 at 600 nm. The obtained bacteria were collected by centrifugation at 4400 rpm for 5 min. The bacteria were then washed with sterile PBS for three times and diluted to a concentration of 10^8 cell/ml for the adhesion experiments. The bare gold surface, gold surface coated with L-carnitineMA polymer and PCBMA polymer were placed in the bacterial suspension for 0.5 h.

SEM imaging and bacteria adhesion density

After bacteria incubation with the substrates, the substrates were removed and immersed in a fix solution of 2.5% glutaraldehyde, 2% paraformaldehyde in 0.1 M sodium phosphate buffer. Then the substrate was dehydrated in a gradient ethanol series and dried in vacuum. Before SEM imaging, the substrate sample was coated with a nano-gold layer for 20 s using an SEEVac Conductive IV sputter coater. The adhered bacteria were imaged using a JSM - 6510LV SEM at 5 μm magnification and bacteria adhesion density was calculated by counting the bacteria number per 900 μm^2.

References

1. J. Homola, *Surface Plasmon Resonance Based Sensors*, Springer-Verlag, **2006**
2. Cao Z, Brault N, Xue H, et al. Manipulating Sticky and Non-Sticky Properties in a Single Material, Angew. Chem. Int. Ed., 2011, **50**, 6102-6104.
3. Wang W, Lu Y, Xie J, et al. A zwitterionic macro-crosslinker for durable non-fouling coatings, Chem. Comm., 2016, **52**, 4671-4674.

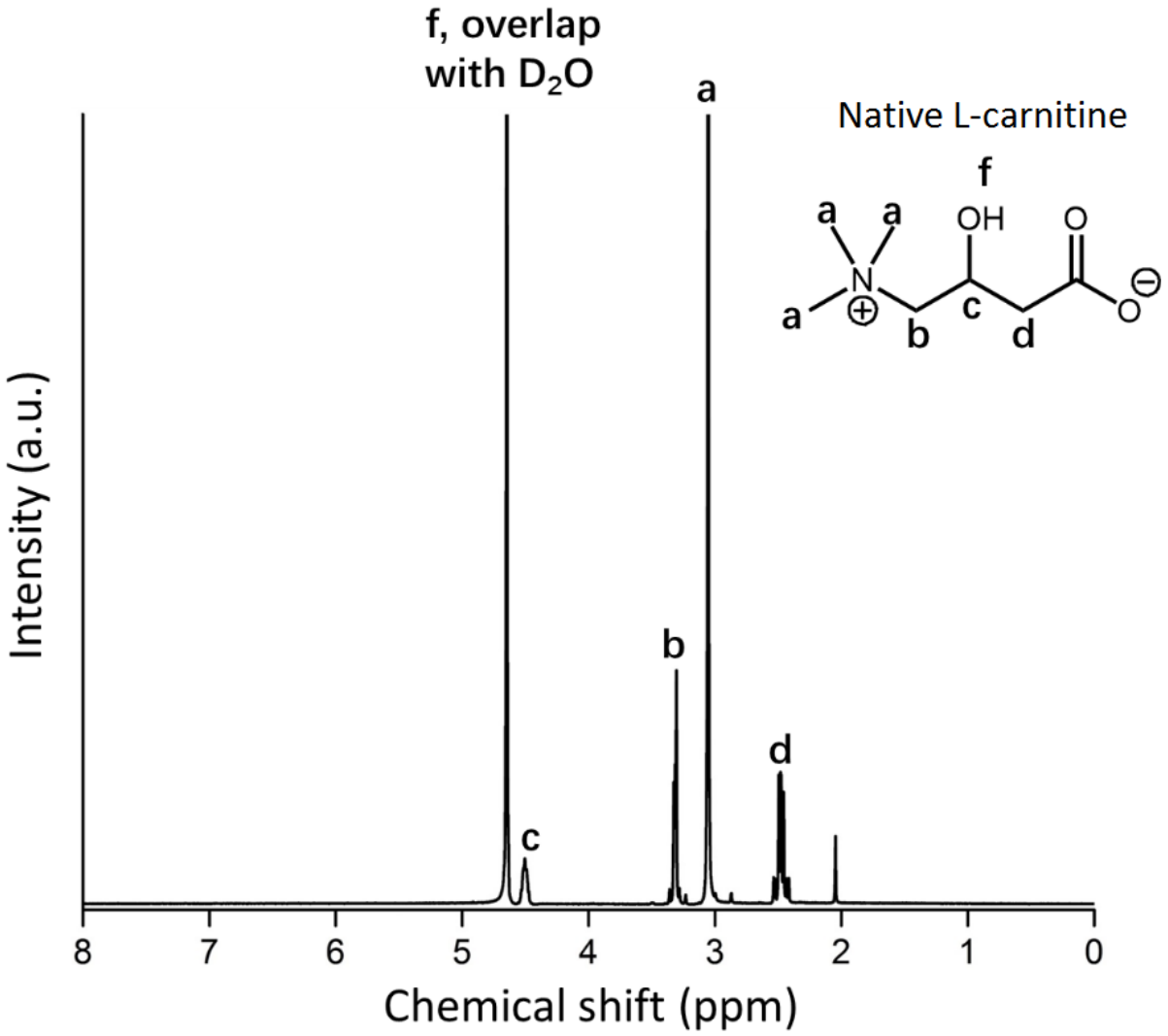

Figure S1, H^1 NMR characterization of L-carnitine

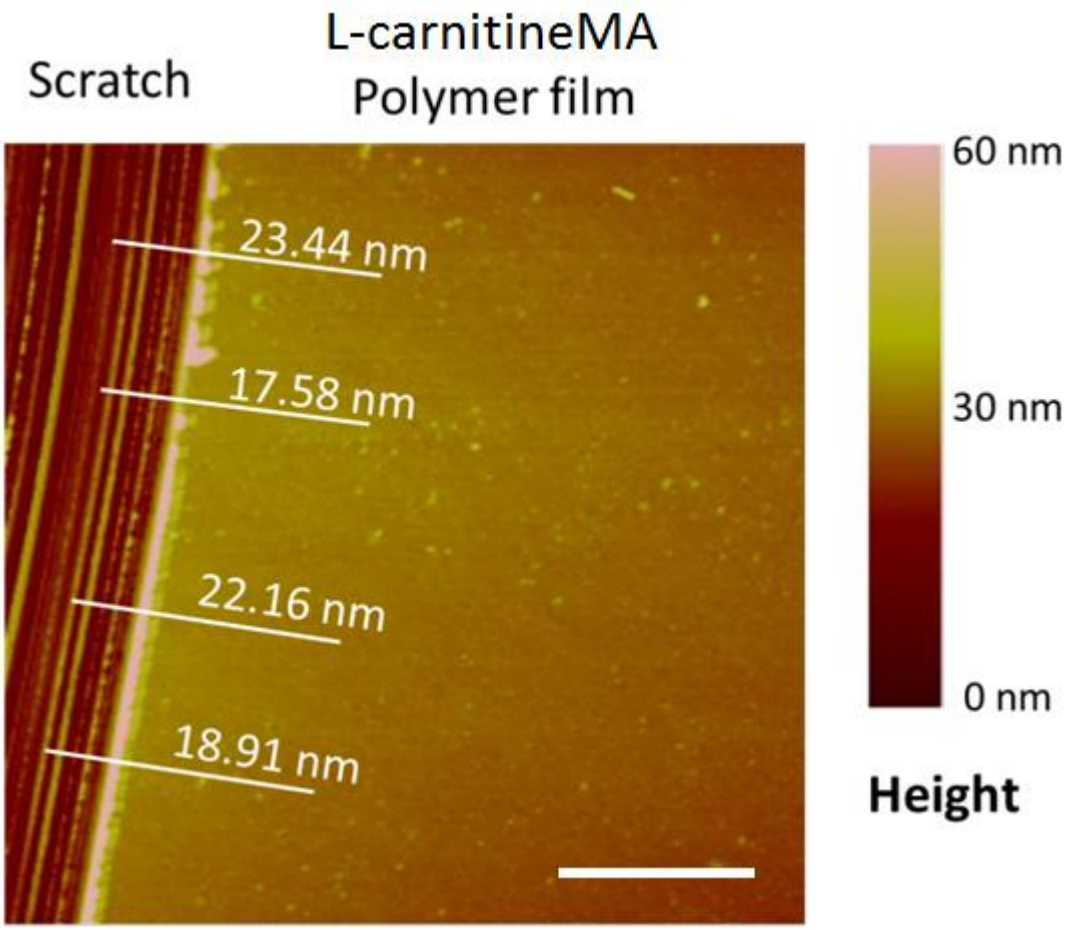

Figure S2 measurement of the scratch depth on the L-carnitineMA polymer brush surface, scale bar = 10 µm

Chemicals	T(°C)	DCM	THF	EA	$CHCl_3$	Acetone	ACN	DMF	DMSO
L-carnitine	40	No	No	No	No	No	No	No	partial
hydrochloride	80	No	No	No	No	No	No	partial	Yes
L-carnitine	40	No	No	No	No	No	No	No	No
inner-salt	80	No	No	No	No	No	No	No	partial

Table S1 Solubility of L-carnitine hydrochloride and L-carnitine inner-salt at 80 mg/ml in different solvent at 40 °C and 80 °C (No: not soluble; partial: partially soluble; Yes: soluble)

solvent	Water	DMF	Methanol	Film thickness (nm)
Volume ratio	1	0	0	8.5 ± 1.3
	1	1	0	16.4 ± 2.8
	1	0	1	5.7 ± 1.2
	0	0	1	2.3 ± 0.5
	2	1	0	23.0 ± 3.8
	2	0	1	7.3 ± 1.9
	1	1	1	6.5 ± 2.2

Table S2 L-carnitineMA polymer coating thickness on gold substrate obtained from different ATRP reaction solvent (average ± standard deviation, n=3)