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Supporting Information

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Colorless Multifunctional Coatings Inspired by Polyphenols Found in Tea, Chocolate, and Wine**

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Experimental Section:

Materials. TiO_2-coated silicon wafers (~4 nm thickness on SiO_2) and silicon wafers were cut into 1 cm x 1 cm pieces and subsequently cleaned by sonication in the following media: acetone, 2-propanol, ultrapure water for 20 min each. PC, PS, PTFE, Au, PDMS, and SS were cleaned by sonication in 0.12 M HCl and 2-propanol for 20 min, washed with DI water 3 times and dried with nitrogen gas. Epigallocatechin-3-gallate, epigallocatechin, epicateching-3-gallate, pyrogallol and tannic acid were used as acquired without any further purification (Sigma-Aldrich).

Polyphenol film deposition from tea and red wine. Green tea sachets (Tazo®) were steeped for 5 min in boiling tap water, after which the hot infusion was transferred into clean porcelain cups (Pier 1 Imports), covered to prevent evaporation, and allowed to stand undisturbed at room temperature for 2, 4 or 6 h. Likewise, clean wine glasses were partially filled with red wine (Malbec), covered to prevent evaporation, and allowed to stand undisturbed at room temperature for 8 h. All beverage containers were then rinsed with tap water after discarding the beverage solutions. The adherent polyphenol film was visualized by filling the dried beverage containers with 100 mM $AgNO_3$ for 24 h, followed by rinsing with DI water. Tea cups, tea sachets and wine glasses were photographed before and after modifications, and the tea sachets were further characterized by SEM (Hitachi).

Crude extracts. Green Tea Extract (GTE): 1 g of dry green tea leaves (Lipton) was covered with 3 mL of 80°C DI water for 5 minutes, followed by addition of 7 mL of methanol. The mixture was sonicated (Branson) for 30 min, centrifuged at 3,000 rcf for 10 min, the supernatant collected, and the pellet subjected to 2 additional cycles of extraction after which the extracts were pooled. Cacao Bean Extract (CBE): To defat raw cacao, 5 g of raw cacao nibs (Navitas Naturals) were homogenized (Kinematica) in 50 mL of hexane for 5 min, sonicated for 5 min, centrifuged at 3,000 rcf for 10 min, and the supernatant was discarded. The cacao pellet was subjected to an additional cycle of defatting prior to extraction. Defatted cacao nibs were homogenized in a 70/29.8/0.2 mixture of acetone/water/acetic acid for 5 min, sonicated for 10 min, centrifuged at 3,000 rcf for 10 min, the supernatant collected, and the pellet subjected to 2 additional cycles of extraction after which the extracts were pooled. Dark Chocolate Extract (DCE): polyphenol extraction from dark chocolate followed the same defatting and extraction protocol used for CBE. Red Wine Extract (RWE): 410 mL of red wine (Malbec) were concentrated to approximately 100 mL using a rotary evaporator (Buchi) and mild heating (40°C). Expended Green Tea Extract (EGTE): 2 g of dry green tea leaves were steeped in hot water for 10 min to make a morning cup of tea, after which the expended solids were subjected to 5 min of homogenization and 10 min of sonication in 20 mL of 0.12 M acetic acid. After centrifugation, the supernatant was collected. All crude extract solutions prepared using organic solvents were concentrated using a rotary evaporator and the aqueous fractions passed through a 0.22 μm filter prior to freezing and lyophilization.

Polyphenol content of crude extracts. A previously established protocol using Folin-Ciocalteu (FC) reagent[1] was modified for use with a plate reader (BioTek). Gallic acid standard solutions were prepared from 0 to 0.5 mg/mL in water. Stock solutions of polyphenol-containing extracts and modified fumed silica particles were prepared at a concentration of 0.5 mg/mL. For the assay, 790 μL of DI water were combined with 10 μL of stock solution in a 1.5 μL centrifuge tube (Eppendorf). To individual tubes, 50 μL of FC reagent were added, the contents were vortexed and allowed to react for 5 minutes. 150 μL of 20% sodium carbonate were added to each tube, the contents mixed and allowed to react for 2 h. Of the resultant solutions, 250 μL

were transferred to wells of a 96-well tissue culture plate (Greiner) and absorbance was measured at 765 nm. Based on the standard curve, gallic acid equivalent (GAE) polyphenol content was determined for 1 g of extract or fumed silica-based antioxidant material.

General procedure for coating of substrates by crude extracts, plant polyphenols, PG, and TA. Except where noted, clean substrates were immersed into 0.1-2.0 mg/mL solutions of PG, TA, or crude extracts in buffered saline (100 mM bicine, 0.6 M NaCl, pH 7.8) for 0.5-24 h at R.T. with mild agitation on a rocking platform. Subsequently, substrates were removed from the modification solution, rinsed with DI water and dried with nitrogen gas. TA-modified substrates were further sonicated in DI water for 10 sec, followed by rinsing with DI water and drying with nitrogen gas. Metallization of silver was achieved by incubation of polyphenol-modified substrates in 100 mM $AgNO_3$ for 0.5-24 h. Incorporation of mPEG-SH and mPEG-NH_2 (5k MW, Laysan Bio) was achieved by incubating polyphenol-modified substrates in 1 mM mPEG solution in pH 7.8 buffered saline for 10 minutes. Chemical composition of resultant surfaces was confirmed by XPS (Thermo) survey scans collected between 0-1,100 eV binding energies, with detailed scans of C1s, O1s, and characteristic substrate electrons performed in triplicate for element composition evaluation. Coating thickness was determined by optical ellipsometry (J.A. Woollam). Surface morphology was characterized by SEM (Hitachi) in secondary electron mode, with the electron beam operating at 2,000 V and 10 μA, without the use of additional conductive coating.

The effect of excluding molecular oxygen during coating formation was determined by modifying Au, TiO_2 and PC substrates under oxygen-depleted conditions. Buffered saline, along with PG and TA powders, was allowed to acclimate in an anaerobic glove box (molecular oxygen concentration ≤ 5 ppm, Coy Lab Products) overnight. The powders were combined with buffered saline at 2.0 mg/mL and agitated using a rocker for 24 h.

PG modification and plasmon tuning of gold nanorods. CTAB-stabilized gold nanorods were prepared using a standard protocol.[2] For deposition of adherent PG layer, 0.5 mL of Au-CTAB NR suspension were centrifuged and the resultant pellet suspended in 0.1 mg/mL PG, pH 7.8 (100 mM bicine, no NaCl supplementation). The suspension was sonicated for 20 min, followed by addition of 10 μL of 12 mM acetic acid, centrifugation and washing with 1 ml water. For silver-shell deposition, sonication in PG solution was immediately followed by addition of 0-4,000 μM $AgNO_3$, sonication for 10 min, addition of acetic acid, centrifugation, and washing with water. UV-Vis spectra were collected between 450-950 nm for the resultant NR constructs. STEM (Hitachi) was used to visualize the PG coatings and gold core-silver shell morphologies of the NR constructs, with twin EDS channels utilized to collect characteristic X-rays for elemental mapping. Solutions of modified nanorods were photographed with the light source positioned in the background.

Bacterial cell attachment and viability. For attachment studies, PC substrates modified with PG (2 mg/mL in buffered saline, 8 h) and mPEG-SH, as described above, were exposed to $1x10^8$ CFU/mL of *Pseudomonas aeruginosa* (7700, ATCC) or *Staphylococcus aureus* (29213, ATCC) in 0.85% NaCl for 24 h at 37°C. Attached cells were stained with a live/dead stain (Invitrogen), washed and imaged using a fluorescence microscope (Leica) using a 20x objective lens. Quantification of surface area coverage was performed using ImageJ software (NIH), considering the total of both live and dead cells, with nine live/dead image sets collected per sample type. For viability assessment, modified PC surfaces were exposed to 20 μL of $1x10^8$ CFU/mL of bacteria premixed with live/dead stains in saline, hermetically sealed using VALAP (1:1:1 mixture of Vaseline:lanolin:paraffin) against a glass microscope slide, and incubated for

24 at 37°C.[3] During incubation, cells settled under the influence of gravity alone into direct contact with the coating and therefore this configuration probed on-surface antibacterial effects of the coating. In parallel, the experiment was performed in an inverted configuration to test for off-surface antibacterial effects. The bacteria were imaged using a fluorescence microscope and the fraction of live cells quantified using ImageJ software.

Mammalian cell attachment and viability. TiO_2, PC and polystyrene substrates were modified with PG or TA (2 mg/mL, 8 h) and mPEG-SH or mPEG-NH_2 as described above, and covered with a solution of 3T3 fibroblasts (NIH) in DMEM supplemented with 10% NBCS (Gibco), with a final concentration of 10,000 cells per sample. The samples were incubated for 24 h at 37°C with 5% CO_2, stained with live/dead stain (Invitrogen), imaged with a fluorescence microscope, and attachment and viability quantified using ImageJ software.

Antioxidant and mammalian cell protective assays of PG and TA coatings. Polyphenol Coating of Fumed Silica: 2 g of fumed silica (Aerosil® 150, a generous gift from Evonik Industries) and 80 mg of either PG or TA were added to a 50-mL centrifugation tube, which was then filled with pH 7.8 buffered saline. The solution was shaken for 30 min and centrifuged at 15,000 rcf for 10 min. After decanting the supernatant, the fumed silica was washed and centrifuged repeatedly (4x in 50 mL water, 2x in 50 mL isopropanol) and then dried under reduced pressure. Radical Scavenging Assay: A previously described radical scavenging assay[4] was modified for use with a plate reader. A scintillation vial was charged with 3 mL of DPPH solution (29 μM) in methanol and 12.5, 25.0, 37.5, 50.0, 75.0, or 100.0 μL of bare, PG-coated, or TA-coated fumed silica suspension (0.6 mg/mL) were added. Methanol with no fumed silica was used as a control. At 15 and 60 min, 250 μL of each solution were added to wells in a 96-well plate, and the absorbance at 515 nm was recorded with a plate reader. All measurements were taken in triplicate. Antioxidant Assay: The assessment of antioxidant performance was performed as described,[5] modified for use with a plate reader. Ethanol was used to disperse bare, PG-coated, or TA-coated fumed silica (0.5 mg/mL). This ethanolic mixture (40 μL) was added to 1 mL of a water-based suspension of β-carotene, linoleic acid, and Tween® 40. Ethanol with no fumed silica was used as a control. The resulting solutions (250 μL) were added to a 96-well plate and incubated at 50°C. All non-experimental wells were filled with water in order to prevent evaporation from experimental samples. Absorbance at 470 nm was monitored over 3 h with a plate reader. All measurements were made in triplicate. Cell protective assay (nanoparticles): Mammalian cell protective assay was performed as previously described with slight modifications.[6] 40,000 NIH 3T3 fibroblasts per well were grown overnight in a 96-well tissue culture plate. The cells were preloaded with dichlorofluorescin in growth media for 1 h. After washing with Hank's balanced salt solution (HBSS), the wells were incubated with 2,2'-azobis(2-amidinopropane) dihydrochloride in parallel with polyphenol-modified and unmodified fumed silica nanoparticles. Fluorescence was monitored over 1.5 h at 530 nm with excitation at 485 nm using a plate reader. Cell protective assay (TCPS): Wells of a 96-well tissue culture plate were modified with 2.0 mg/mL solutions of PG and TA in buffered saline for 8 h at R.T., followed by rinsing with DI water and drying under ambient conditions. 40,000 NIH 3T3 fibroblasts per well were grown overnight in unmodified and PG- or TA- modified wells. After rinsing with HBSS, the cells were preloaded with dichlorofluorescin in growth media for 1 h. Lastly, after a final washing with HBSS, the cells were incubated with 2,2'-azobis(2-amidinopropane) dihydrochloride in HBSS. Fluorescence was monitored over 1.5 h at 530 nm with excitation at 485 nm using a plate reader. Fluorescence intensity was directly correlated with intracellular reactive oxygen species levels.

Any Additional Author notes: This study was conceptualized by T.S.S. and P.B.M. T.S.S., R.Z., D.G.B., K.H.A.L, and P.B.M. designed and performed experiments. The manuscript was drafted by T.S.S. and P.B.M. and was reviewed and edited by all of the authors.

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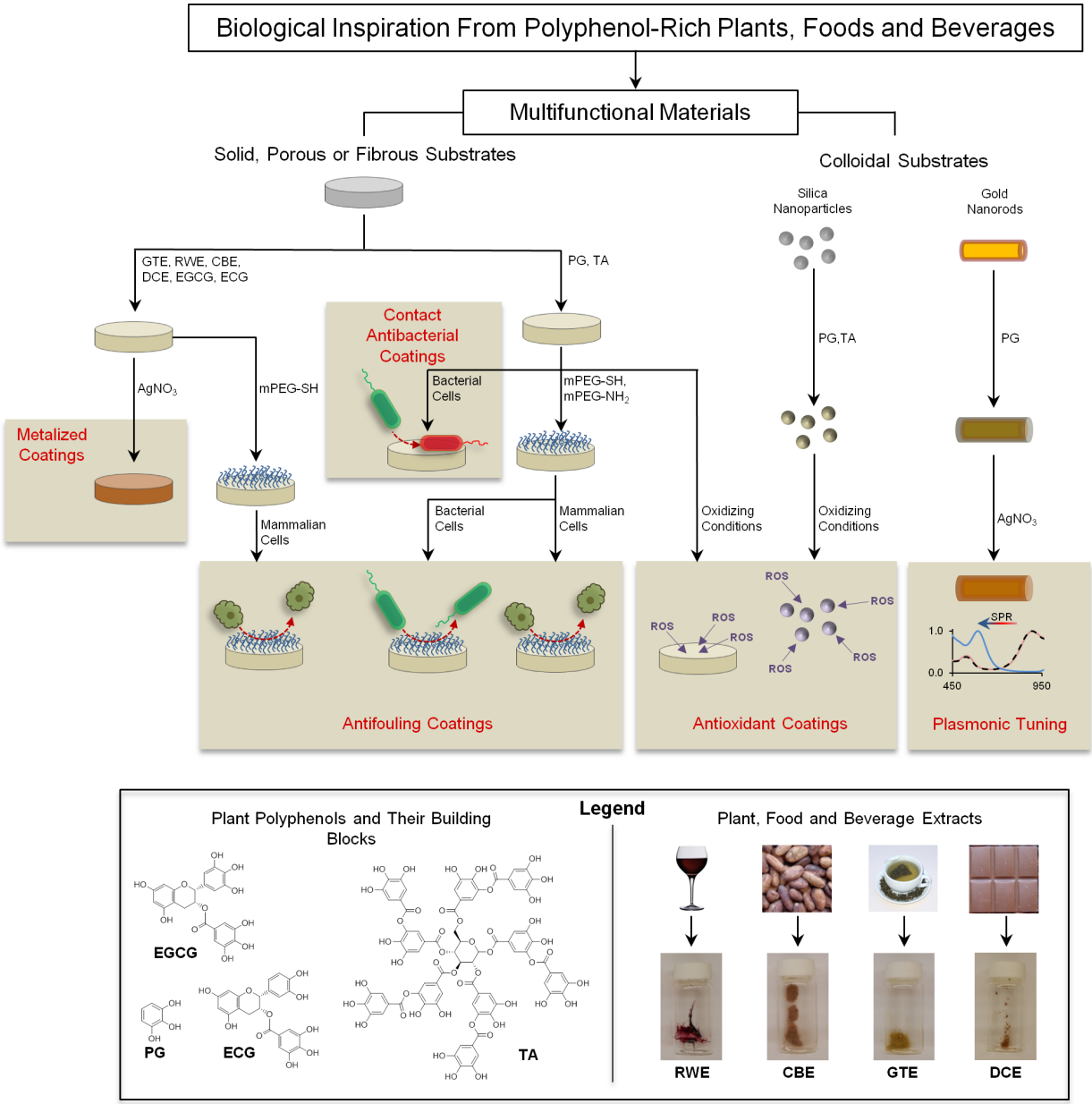
[2] Y.-F. Huang, K.-M. Huang, H.-T. Chang, *Journal of colloid and interface science* **2006**, *301*, 145-154.
[3] T. S. Sileika, H.-D. Kim, P. Maniak, P. B. Messersmith, *ACS Applied Materials & Interfaces* **2011**, *3*, 4602-4610.
[4] aW. Brand-Williams, M. Cuvelier, C. Berset, *LWT-Food Science and Technology* **1995**, *28*, 25-30; bY. Deligiannakis, G. A. Sotiriou, S. E. Pratsinis, *ACS Applied Materials & Interfaces* **2012**, *4*, 6609-6617.
[5] H. Miller, *Journal of the American Oil Chemists' Society* **1971**, *48*, 91-91.
[6] H. Wang, J. A. Joseph, *Free Radical Biology and Medicine* **1999**, *27*, 612-616.

Supplementary Materials:



<u>Figure S1</u>: Schematic illustration of compositions, deposition conditions and potential applications of plant polyphenol inspired multifunctional coatings.

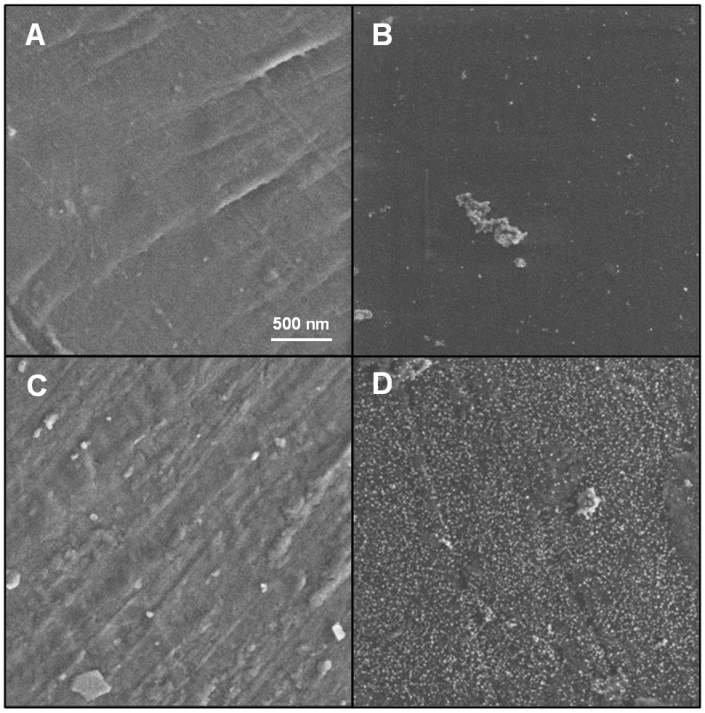


Figure S2: Modification of tea bag surface by green tea infusion. (A) High magnification SEM images of unmodified teabag fiber and (B) teabag fiber following exposure to aqueous $AgNO_3$. (C) Tea bag fiber after green tea infusion and (D) following exposure to aqueous $AgNO_3$. Silver nanoparticles are observed on the fiber surface only after green tea infused tea packets are incubated in $AgNO_3$, coinciding with the visible discoloration of the tea bag as shown in Figure 1C.

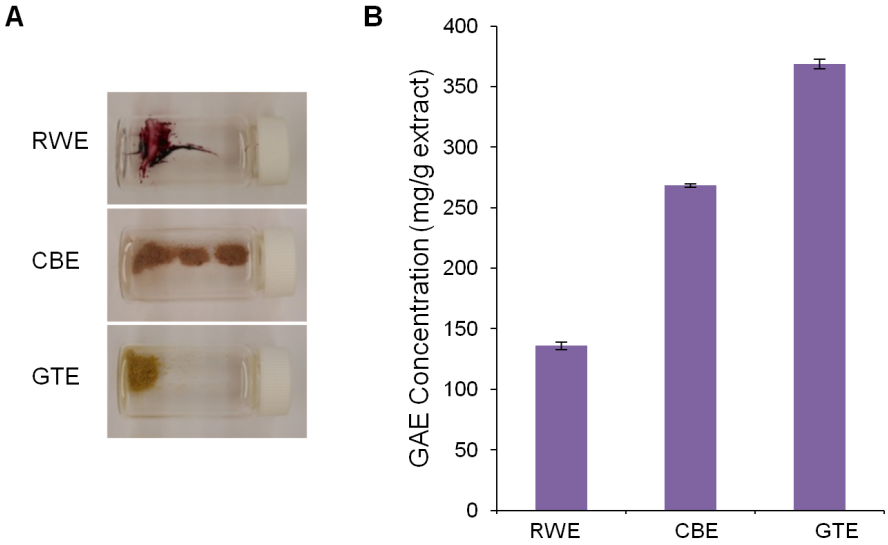


Figure S3: Polyphenol content of red wine extract (RWE), cacao bean extract (CBE) and green tea extract (GTE). (A) Photographs of crude extracts following lyophilization. (B) Polyphenol content of crude extracts, expressed in gallic acid equivalents (GAE).

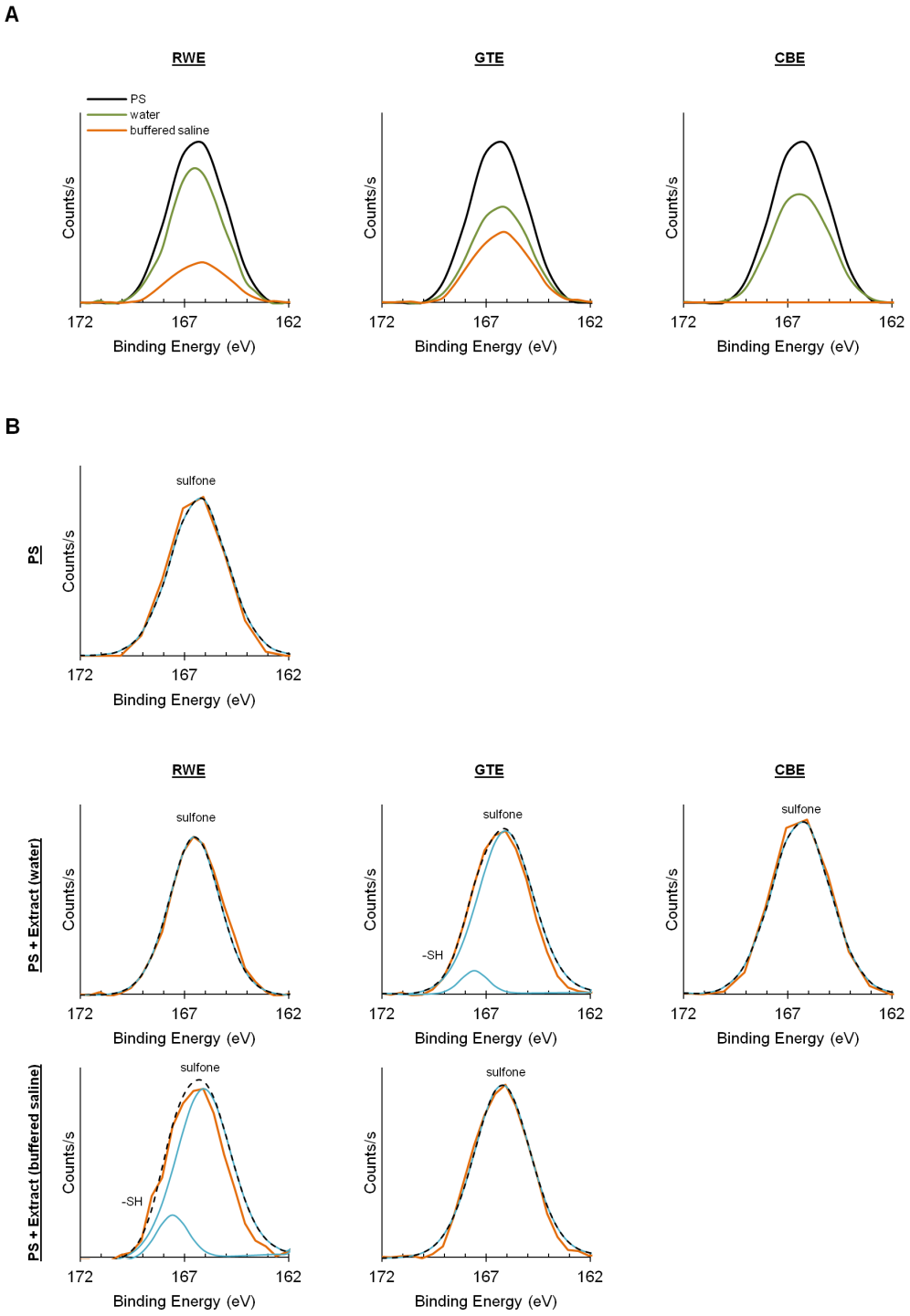


Figure S4: Detailed S2p XPS spectra of polysulfone (PS) modified with crude polyphenol-rich food extracts. (A) Combined S2p spectra of red wine (RWE), green tea (GTE) and cacao bean (CBE) extract. (B) S2p spectra of bare PS followed by modification by respective polyphenol-rich food extract in water and in buffered saline. The experimental signal (orange) intensities have been normalized and fitted (blue) to reflect the sulfur species present, with the overall fit (dashed black) also displayed. Thiol species (167.6 eV) were detected in addition to sulfone (166.1 eV) following certain incubations with RWE and GTE.

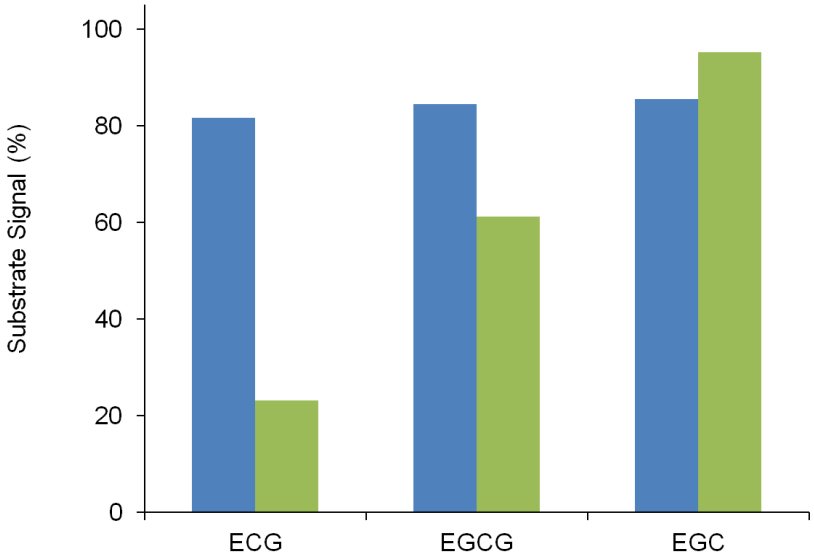


Figure S5: XPS analysis of TiO_2 surfaces modified with ECG, EGCG and EGC. Ti2p signal from the underlying TiO_2 substrate was attenuated following exposure to water (blue) and buffered saline (green) solutions of plant-derived polyphenols (8 h at R.T.). Coating formation was most effective for ECG and EGCG in buffered saline (0.6 M NaCl, pH 7.8).

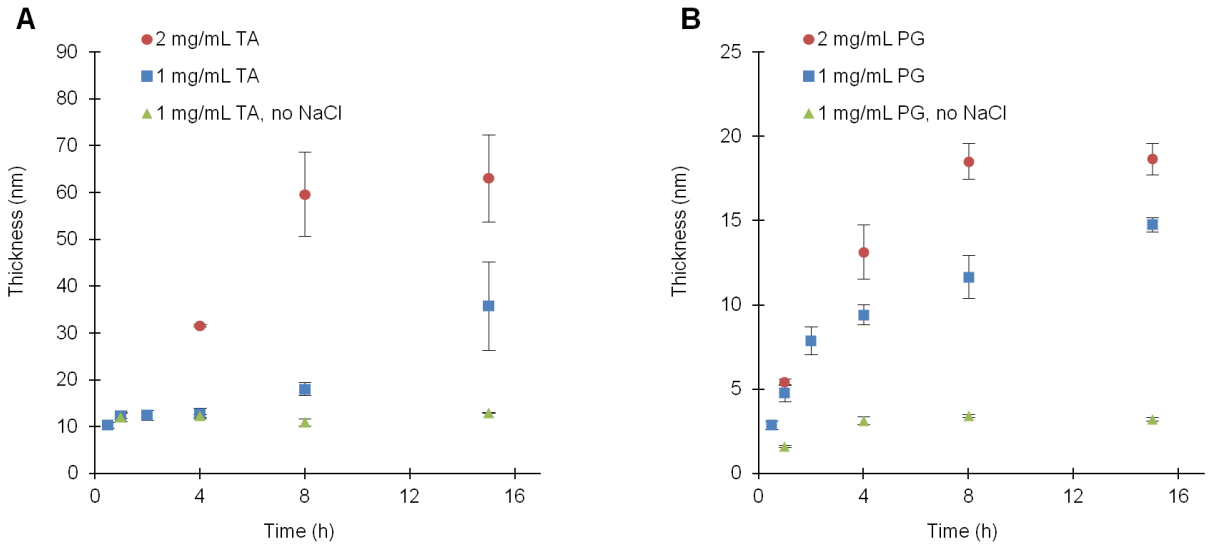


Figure S6: Kinetics of TA and PG coating deposition. Time dependence of (A) TA and (B) PG film deposition on TiO_2 and gold, respectively, as determined by ellipsometry. Coatings were deposited at the concentrations indicated either in water or in buffered saline (0.6 M NaCl, pH 7.8).

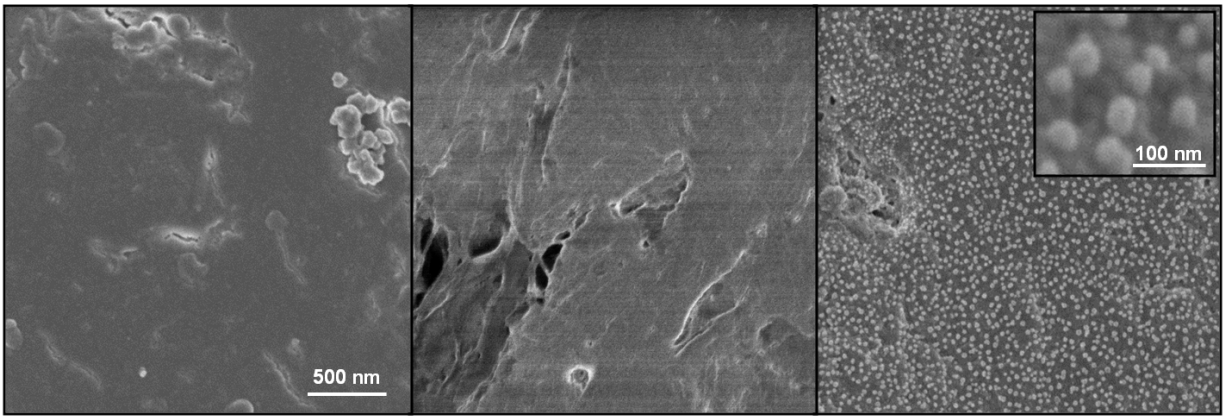


Figure S7: SEM images of PTFE modified with PG and silver. Bare PTFE (left), after modification with PG (middle), and following exposure to aqueous $AgNO_3$ (right, higher magnification in inset). Modification with PG alone does not appear to vastly alter the surface structure of PTFE; however, following incubation in $AgNO_3$, a homogeneous distribution of nanoparticles were present on the surface. These surfaces have been subjected to 10 minutes of ultrasonication in water to ensure stability of resultant coatings.

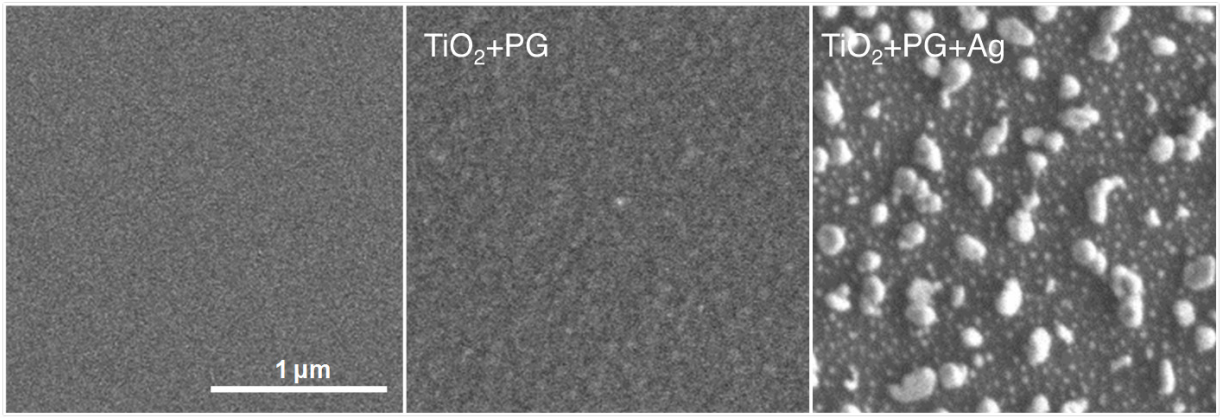
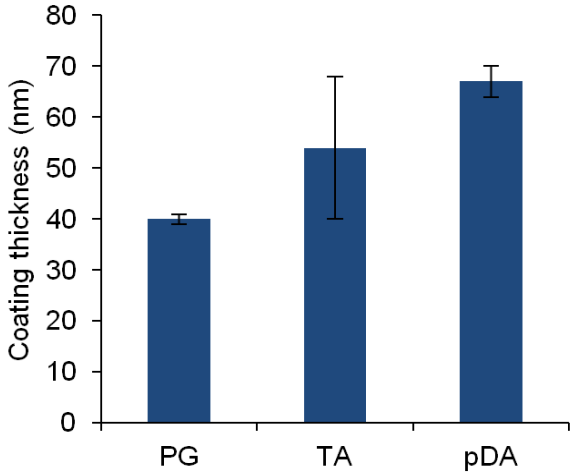
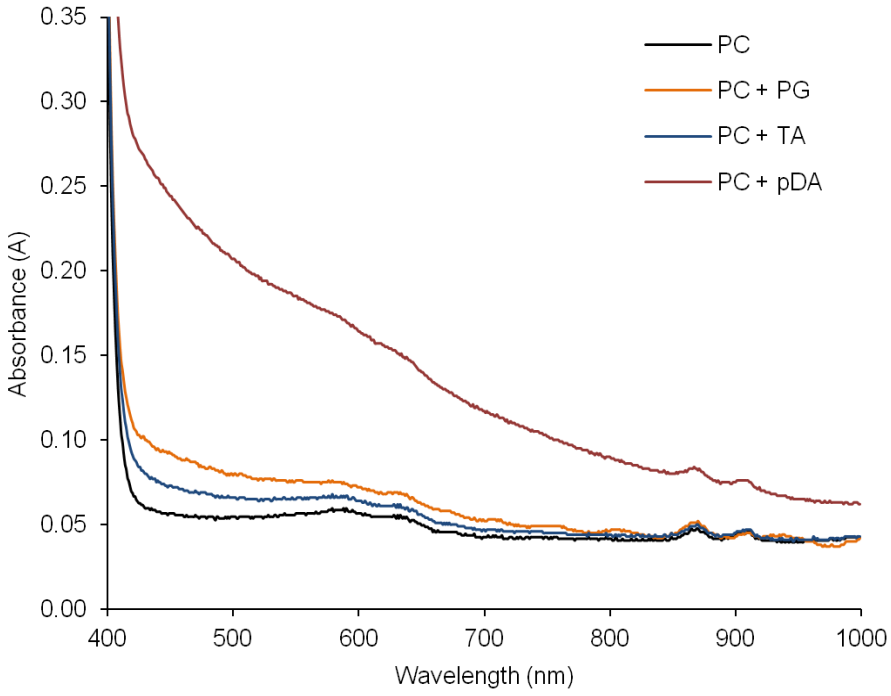


Figure S8: SEM images of TiO_2 modified with PG and silver. SEM images of TiO_2 substrate before and after modification with PG and immersion in aqueous $AgNO_3$ (100 mM). Error bars indicate s.d. (n=3).



<u>Figure S9</u>: Thicknesses of PG, TA and pDA coatings on PC were comparable, as assessed by ellipsometry.

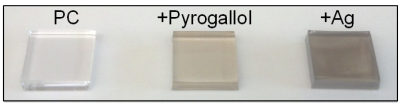


<u>Figure S10</u>: Spectroscopic properties of polycarbonate modified with thin films of PG, TA and pDA. UV-vis spectra of PC samples modified with TA and PG reveal a relatively minor alteration of the optical properties of PC, when compared to modification with pDA.

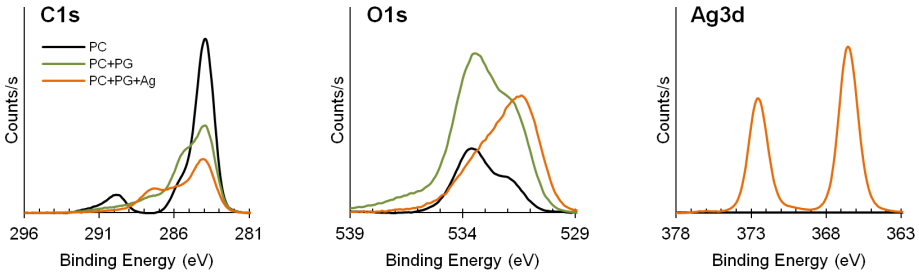
A

Surface	Elemental Composition C	O	N	Ag
PC	85.3%	14.7%	0.0%	0.0%
+ Ag	84.8%	14.9%	0.0%	0.4%
PC + pyrogallol	67.4%	32.2%	0.4%	0.0%
+ Ag	52.9%	26.7%	0.6%	19.9%

B



C



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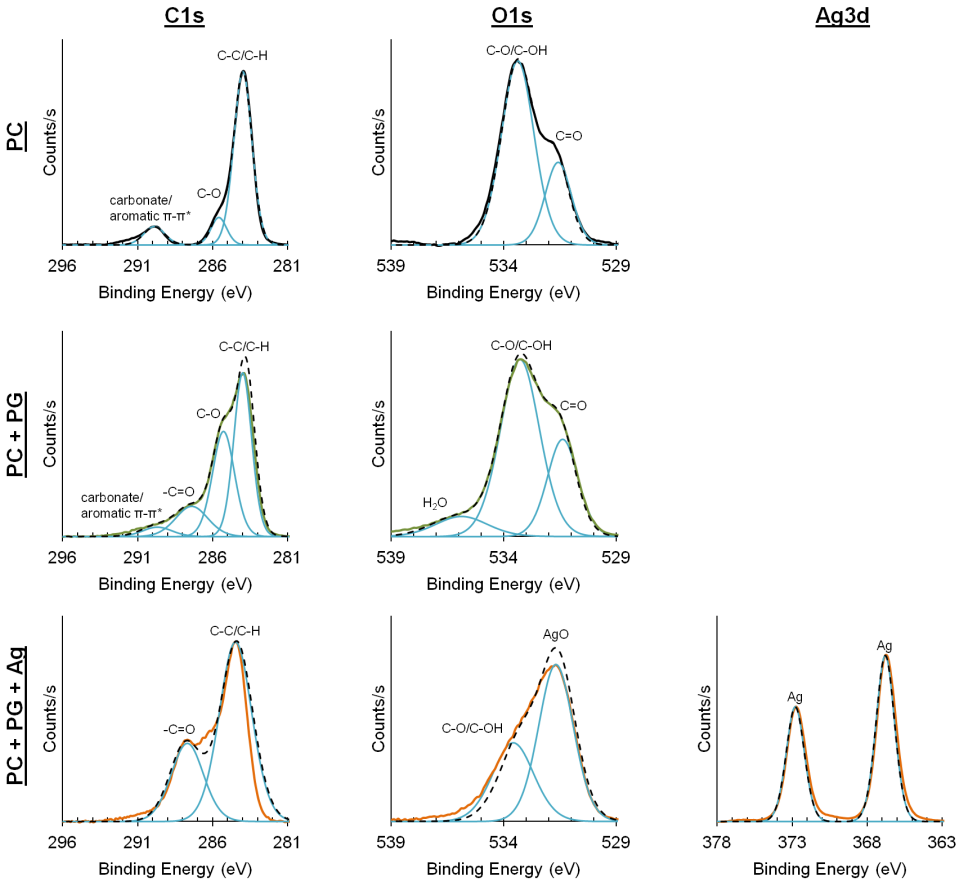
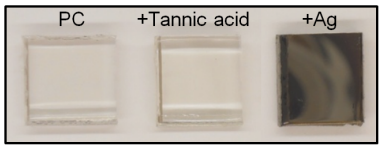


Figure S11: XPS analysis of PC surfaces modified with PG and crude extracts of tea and chocolate. (A) Elemental compositions of polycarbonate (PC) substrates before and after modification with PG and selected crude extracts, and after exposure to $AgNO_3$. (B) Photographs of PC surfaces modified with pyrogallol and silver. (C) XPS spectra of PC, and PC modified with pyrogallol, before and after $AgNO_3$ exposure. (D) Detailed XPS spectra with fitted peaks (blue) and the corresponding envelope (dashed black).

A

Surface	Elemental Composition C	O	N	Ag
PC	85.3%	14.7%	0.0%	0.0%
+ Ag	84.8%	14.9%	0.0%	0.4%
PC + tannic acid	59.8%	39.3%	0.9%	0.0%
+ Ag	52.2%	34.6%	0.0%	13.2%

B



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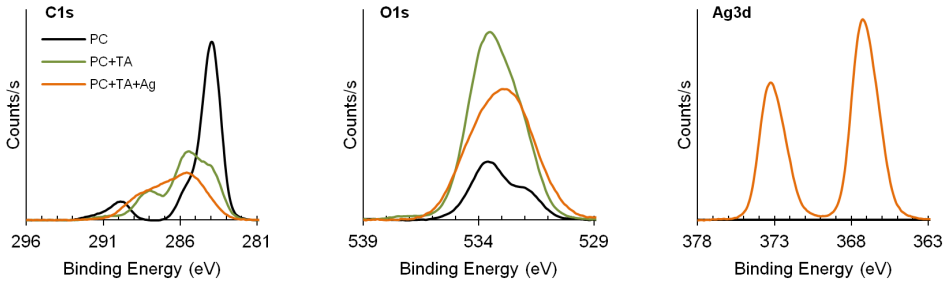

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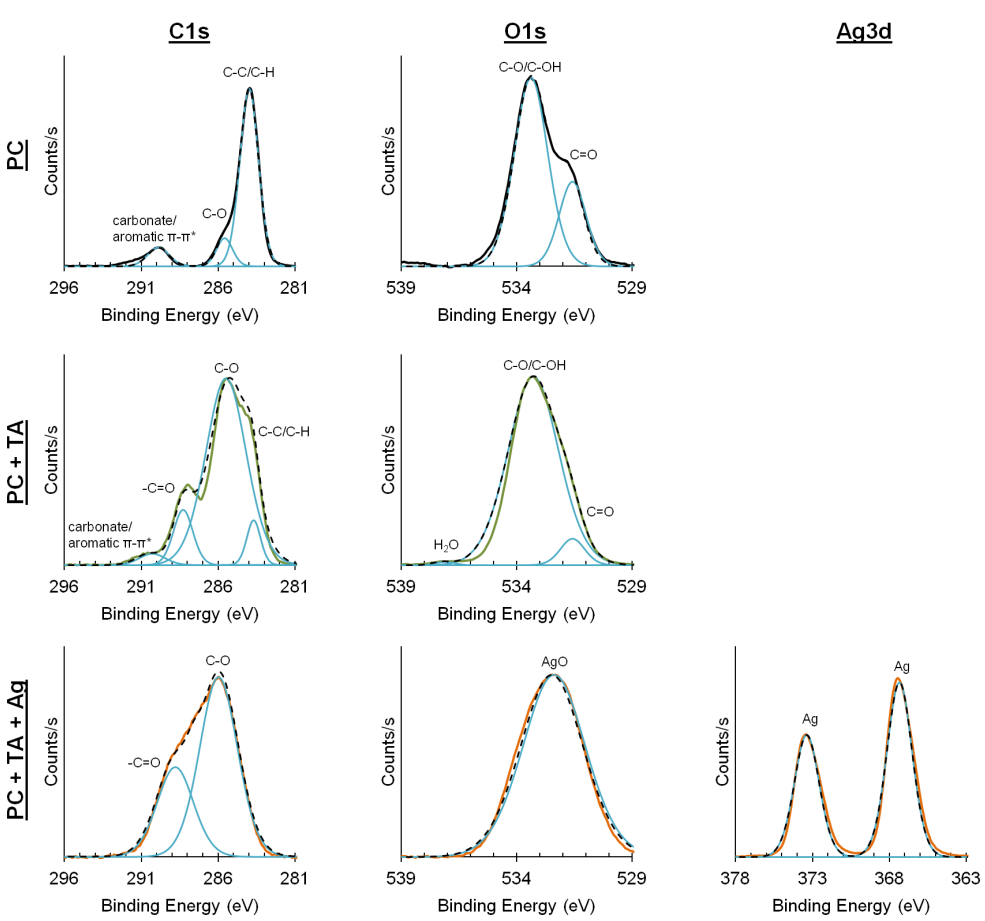


Figure S12: XPS analysis of PC surfaces modified with TA. (A) Elemental compositions of polycarbonate (PC) substrates before and after modification with TA, and after exposure to $AgNO_3$. (B) Photographs of PC surfaces modified with tannic acid and silver. (C) XPS spectra of PC, and PC modified with tannic acid, before and after $AgNO_3$ exposure. (D) Detailed XPS spectra with fitted peaks (blue) and the corresponding envelope (dashed black).

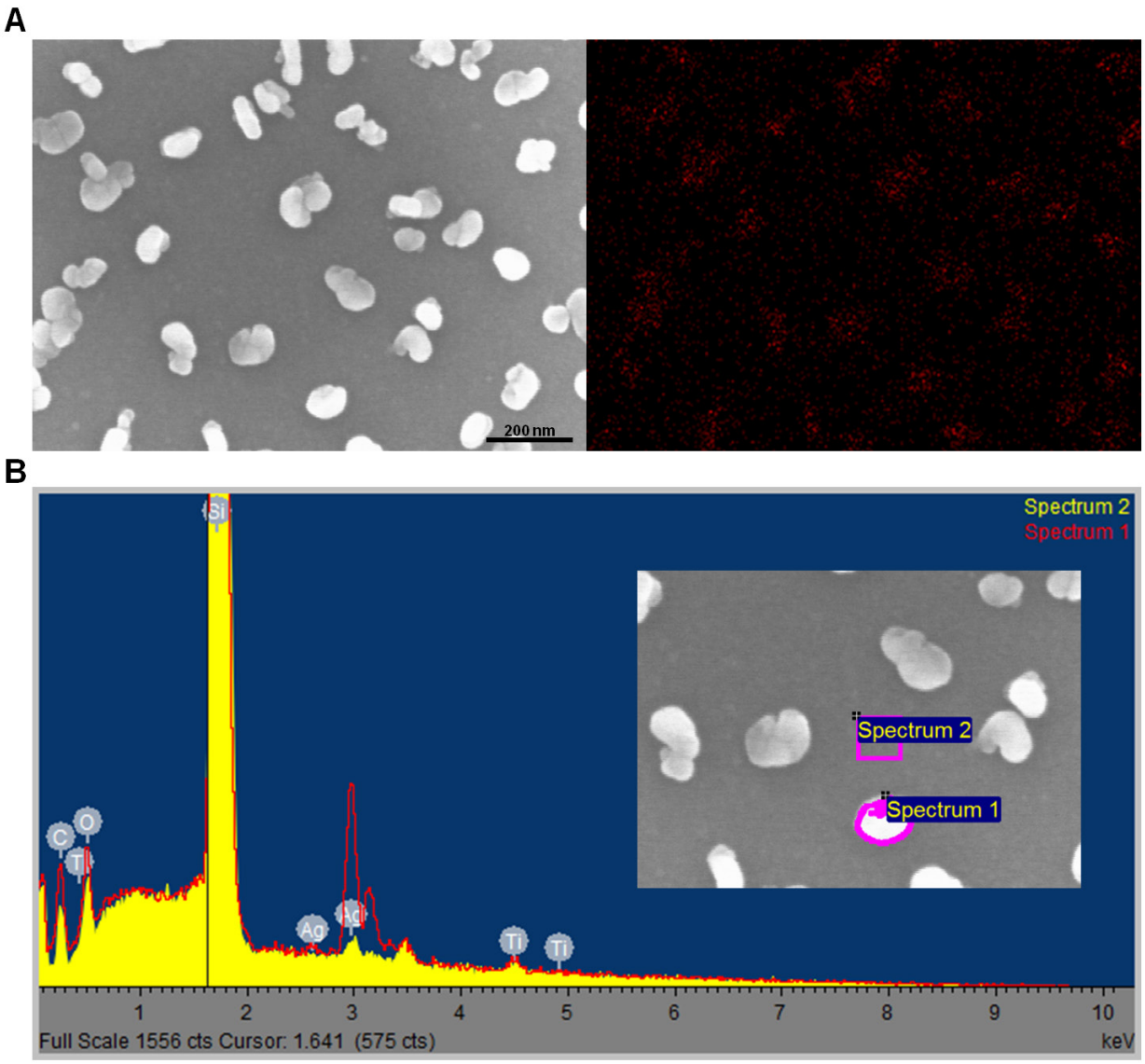


Figure S13: EDS analysis of TiO_2 modified with PG and silver. (A) EDS elemental composition mapping, with secondary electron mode (left) and silver map (right) images shown. (B) Isolated EDS spectra of a silver nanoparticle (Spectrum 1) and background material (Spectrum 2).

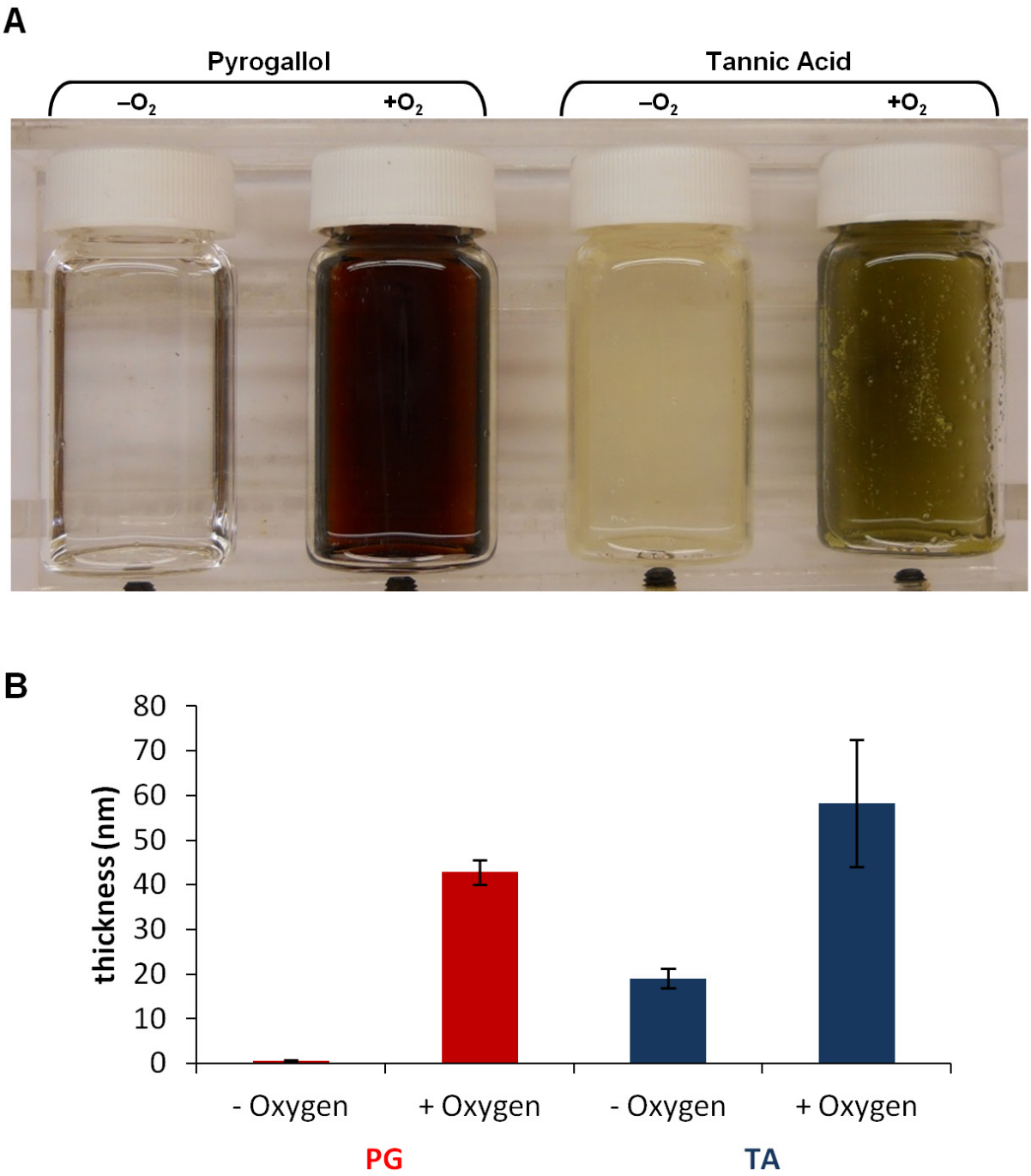
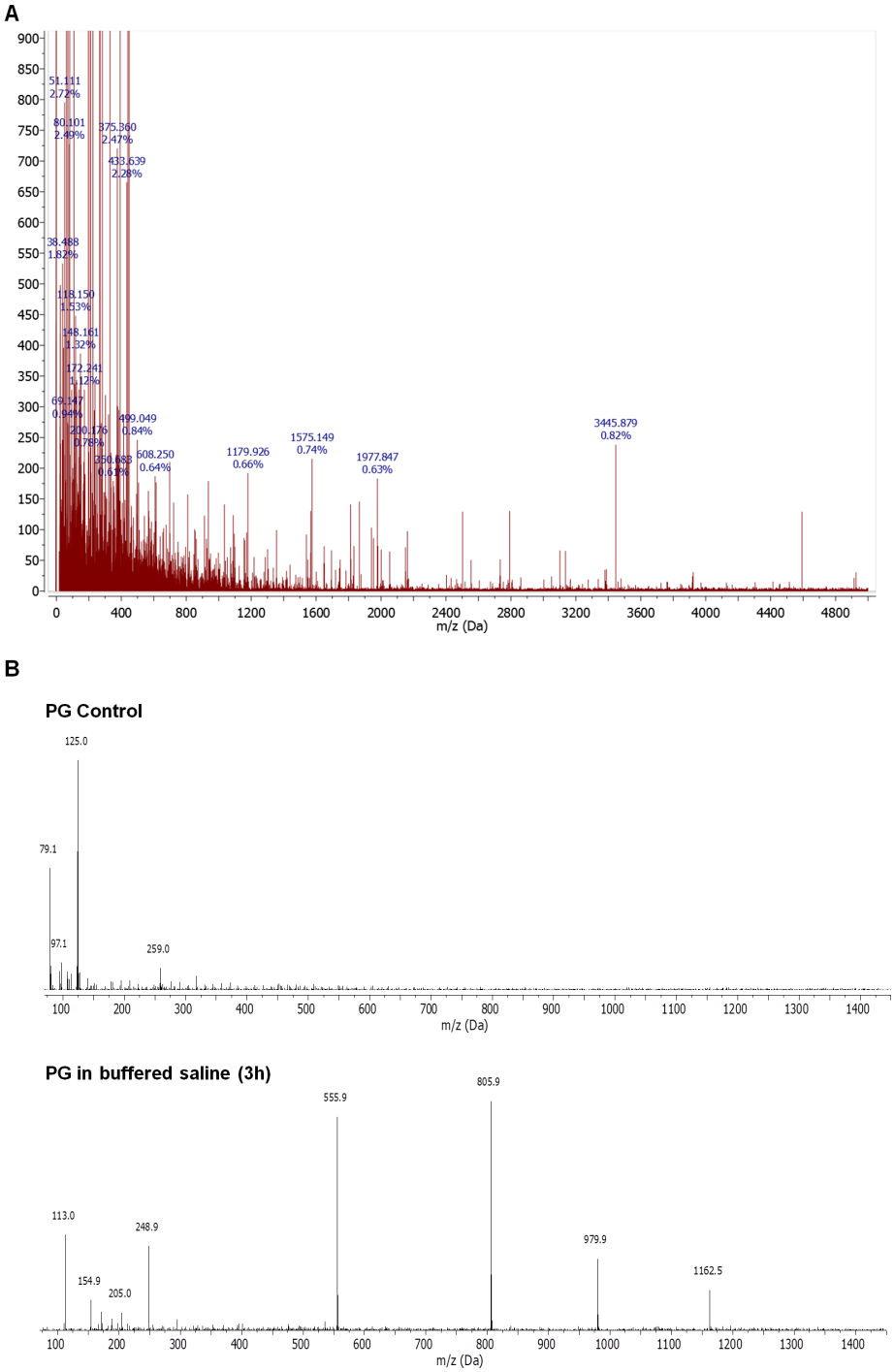


Figure S14: Participation of dissolved oxygen in polyphenol thin film deposition. (A) Photograph of vials containing 2.0 mg/mL solutions of TA and PG following 8 h reaction at R.T. under aerobic and anaerobic conditions. (B) Resultant coating thicknesses of PG and TA films following reactions under aerobic and anaerobic conditions.



<u>Figure S15</u>: MALDI and ESI-MS spectra of a PG coating and polymerization solution. (A) MALDI spectrum acquired directly from a PG-modified MALDI chip. High molecular weight species are evident, some with m/z ratios above 3kDa. The experiment was performed in positive reflection mode at 30% nominal power of the laser source, with data acquired for 20 seconds. (B) ESI-MS spectra indicate the presence of higher molecular weight species in polymerization solutions. Samples were prepared by size-exclusion chromatography using ~700 Da MW cut-off Sephadex media (G10).

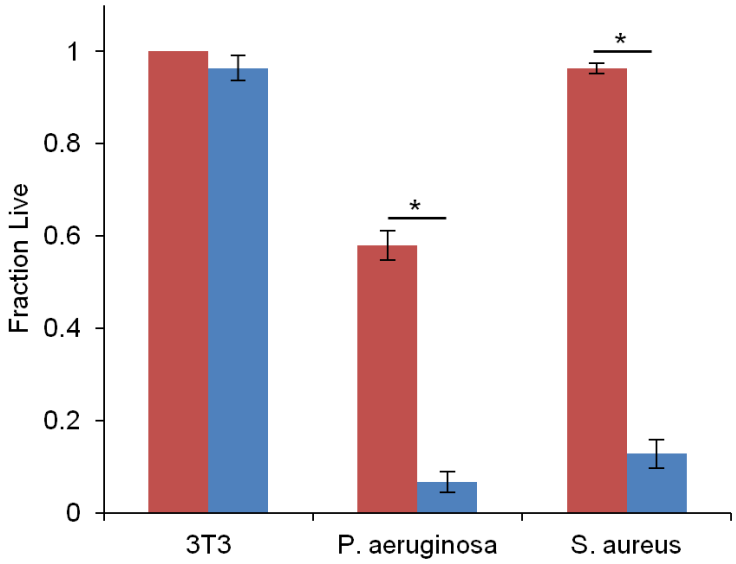
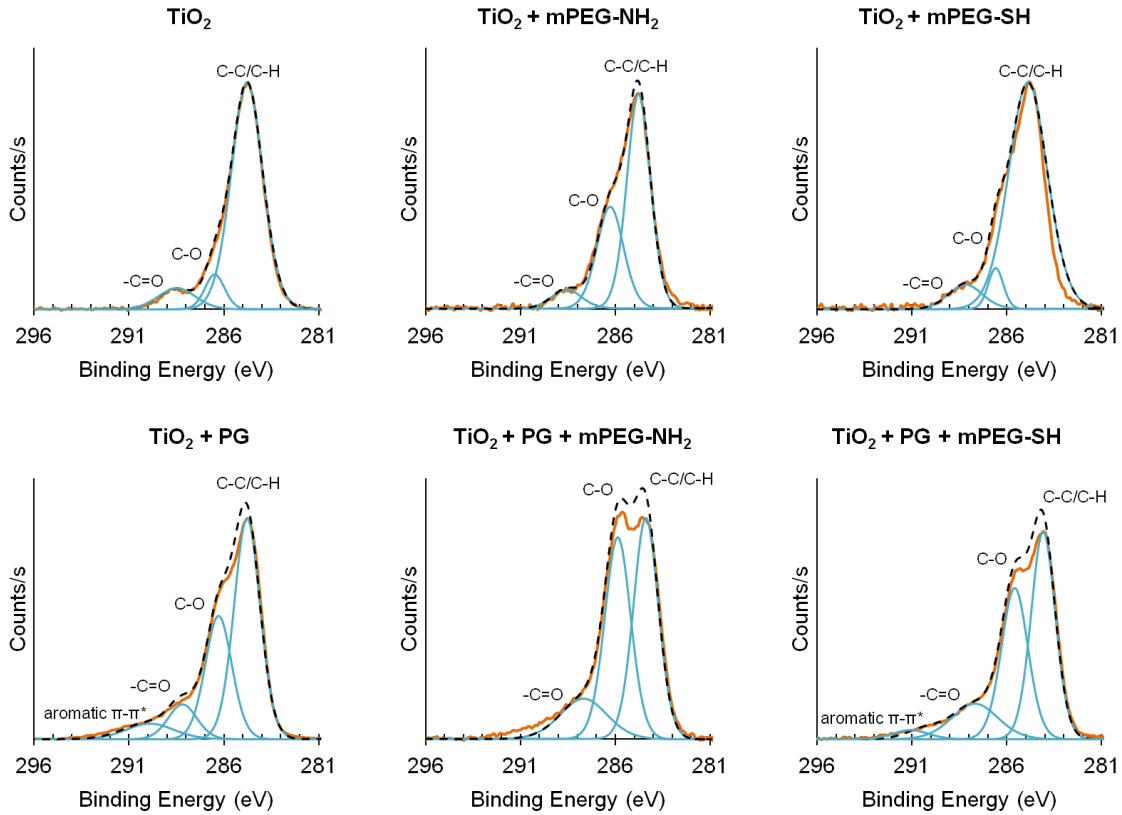


Figure S16: Viability of P. aeruginosa, S. aureus and 3T3 fibroblast cells upon contact with unmodified PC (red) and PG-modified PC (blue).

A



B

Surface	Carbon Species Composition			
	C-C / C-H	C-O	-C=O	aromatic π-π*
TiO_2	83.1%	8.0%	9.0%	0.0%
TiO_2 + PG	53.3%	30.4%	9.2%	7.0%
TiO_2 + mPEG-NH_2	60.5%	32.5%	7.1%	0.0%
TiO_2 + PG + mPEG-NH_2	43.5%	41.5%	15.0%	0.0%
TiO_2 + mPEG-SH	86.0%	6.1%	8.0%	0.0%
TiO_2 + PG + mPEG-SH	45.6%	36.6%	14.8%	3.0%

Figure S17: Grafting of poly(ethylene glycol) onto PG-modified surfaces. (A) Following modification with pyrogallol, mPEG-SH and mPEG-NH_2 were successfully grafted under buffered saline conditions as described in the methods section. Experimental data (orange) was fitted (blue), with the envelope (dashed black) reflecting the quality of the respective fit. Evidence of mPEG grafting is apparent through the emergence of a C1s shoulder region at 286.5 eV, corresponding to ether bonds (upshifted from the C–C / C–H peak at 284.7 eV). (B) Carbon species composition assessed by peak fitting of experimental data. While a small amount of mPEG adsorbs to the bare TiO_2 surface, use of the interfacial pyrogallol layer allows for substantially greater mPEG grafting.

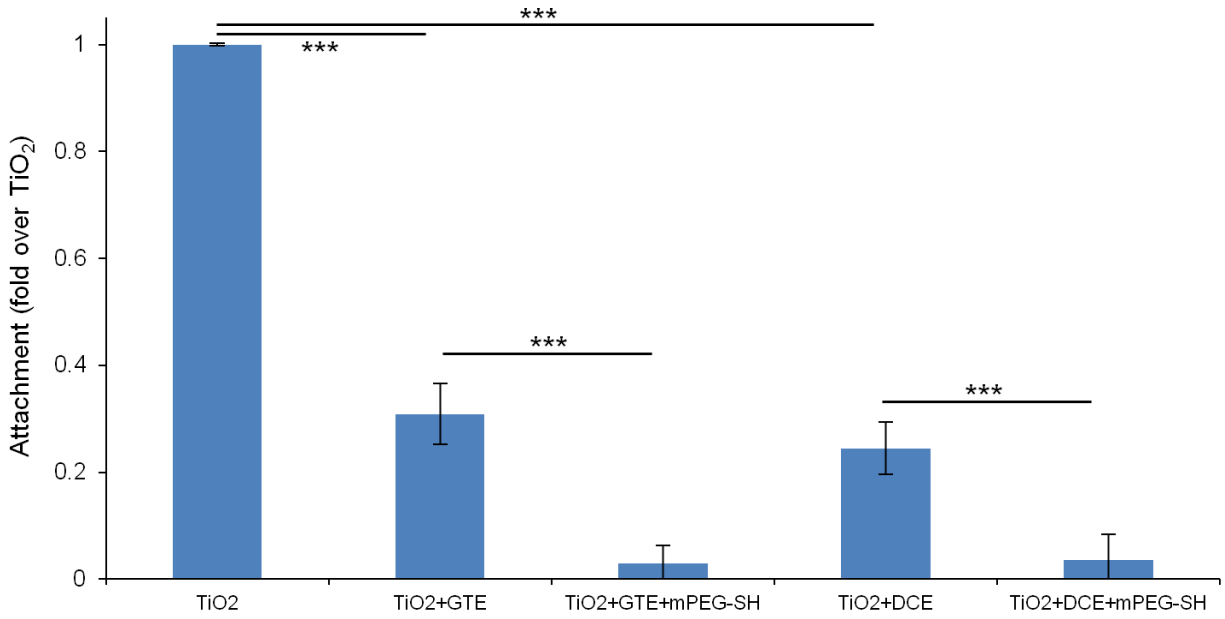
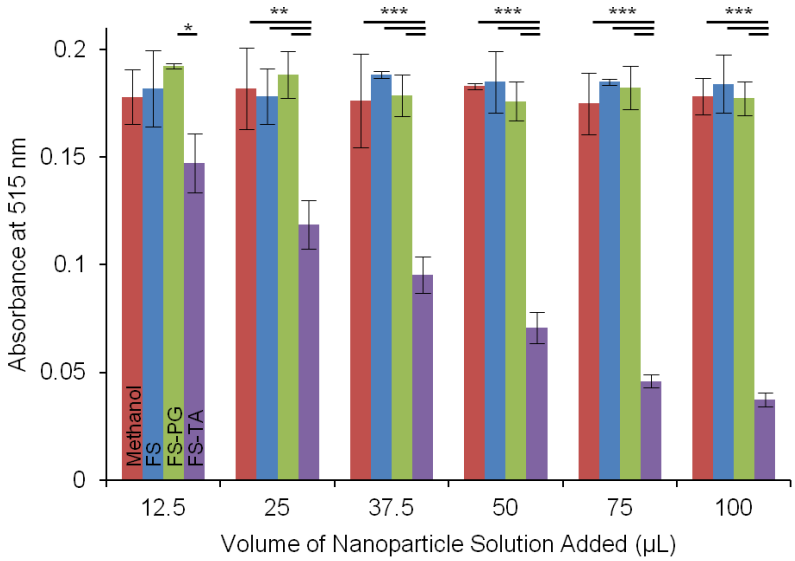
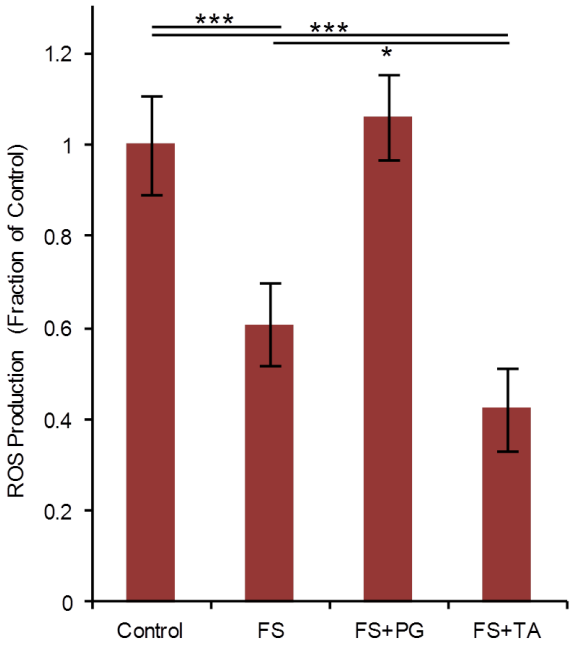


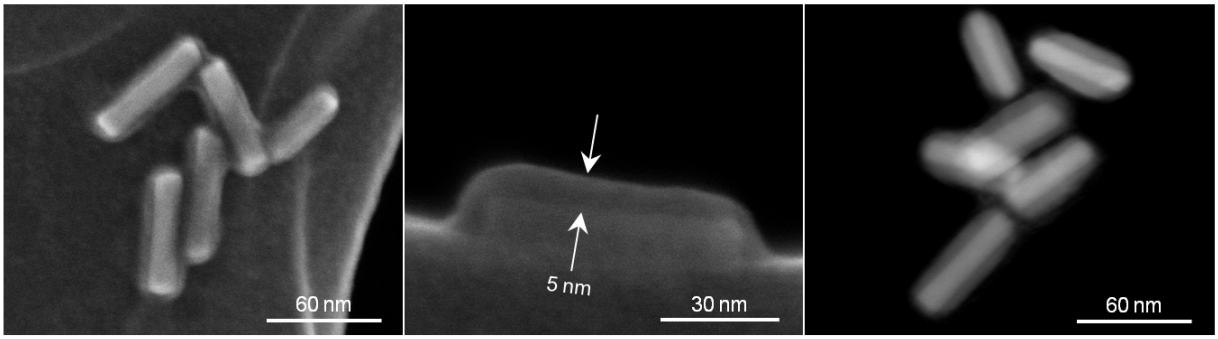
Figure S18: Cell fouling resistance of surfaces modified with food-based polyphenol extracts. Green tea extract (GTE) and dark chocolate extract (DCE) were utilized to modify TiO_2 substrates, followed by grafting of mPEG-SH. Error bars indicate s.e.m. (n=9). Statistical analysis using student's t-test. *** indicates $p < 0.001$.



<u>Figure S19</u>: Radical scavenging properties of polyphenol-modified fumed silica (FS) nanoparticles. Radical scavenging activity of FS, FS-PG, and FS-TA was determined using a stable radical, DPPH. Decreased absorbance corresponds to increasing radical scavenging behavior. It is evident that FS-TA nanoparticles were effective in functioning as radical scavenging materials when compared to methanol-only control, bare FS, and FS-PG. Error bars indicate s.d. (n=3). Statistical analysis using one-way ANOVA followed by a Bonferroni post-hoc analysis. *, **, and *** indicate $p < 0.05$, $p < 0.01$, and $p < 0.001$, respectively.



<u>Figure S20</u>: Reactive oxygen species generation mediated by PG and TA materials. Intracellular ROS production following exposure of 3T3 fibroblasts to FS and polyphenol-modified FS. Error bars indicate s.e.m. (n=3 wells, 9 scans/well).



<u>Figure S21</u>: Gold nanorod modification with PG and silver. STEM image, in secondary electron mode, of gold nanorods modified with PG (left). At higher magnification, the less electron-dense adlayer, attributed to PG, is visible against the gold nanorod core (middle). Following electroless metallic silver deposition, gold-core silver-shell structures are formed, as seen in z-contrast mode (right).