Supporting Information

Catecholamine-Copper Redox as a Basis for Site-Specific Single-Step Functionalization of Material Surfaces

Zhaojun Jia[1], Kern Hast[1], and Enver Cagri Izgu[1,2,3]*

(1) Department of Chemistry and Chemical Biology, Rutgers University, New Brunswick, NJ 08854, USA

(2) Rutgers Center for Lipid Research, New Jersey Institute for Food, Nutrition, and Health, Rutgers University, New Brunswick, NJ 08901, USA

(3) Cancer Pharmacology Program, Rutgers Cancer Institute of New Jersey, New Brunswick, NJ 08903, USA

*Corresponding author email: ec.izgu@rutgers.edu

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Chemicals and materials

Reagents: 3,4-Dihydroxy-L-phenylalanine (L-DOPA) was purchased from Alfa Aesar. *t*-Butyldimethylsilyl chloride (TBDMSCl), 1,8-diazabicyclo-[5.4.0]-undec-7-ene (DBU), di-tert-butyl decarbonate (Boc_2O), potassium carbonate (K_2CO_3), 3-(ethylimino-methylideneamino)-*N*,*N*-dimethylpropan-1-amine (EDC), 4-(dimethylamino)-pyridine (DMAP), trifluoroacetic acid (TFA), imidazole, *p*-toluenesulfonyl chloride (*p*-TsCl), triethylamine (Et_3N), sodium azide (NaN_3), copper(II) sulfate ($CuSO_4$), sodium ascorbate, dopamine (DA) (available in hydrochloride salt form), hydrogen peroxide, paraformaldehyde, iron(III) chloride, ethylenediaminetetraacetic acid (EDTA), glutaraldehyde, α-MEM medium, bovine serum albumin (BSA), anti-vinculin monoclonal antibody, fluorescein isothiocyanate (FITC)-labeled goat anti-rabbit IgG, tetramethylrhodamine-isothiocyanate (TRITC)-phalloidin kit, 4′,6-diamidino-2-phenylindole dihydrochloride (DAPI), actin cytoskeleton/focal adhesion staining kit (FAK100), and Triton X-100 were purchased from Millipore-Sigma. Tris(3-hydroxypropyltriazolyl-methyl)amine (THPTA) was purchased from Click Chemistry Tools. 2-[2-(2-Propynyloxy)ethoxy]ethylamine was purchased from TCI America. Micro bicinchoninic acid (MicroBCA) was purchased from Thermo Scientific. LIVE/DEAD® Baclight™ Bacterial Viability Kit was purchased from Molecular Probes, Inc., Invitrogen. Calcein AM and propidium iodide (PI) were purchased from Dojindo, Japan. 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) cell proliferation assay kit was purchased from BioVision Inc. TAMRA-NH_2 was purchased from Adipogen Life Sciences.

MOI-N_3 molecules: 3-N_3-7-hydroxycoumarin, TAMRA-N_3, 5′-N_3-AGCGTGACTT-3′-Fluorescein (N_3-DNA-FAM), Polyethylene glycol-N_3 (PEG-N_3), Cyclo[Arg-Gly-Asp-D-Phe-Lys(Azide)] (c(RGDfK)-N_3), and Bovine serum albumin with an azide modification (BSA-N_3). 3-N_3-7-hydroxycoumarin was purchased from Combi-Blocks. TAMRA-N_3 was purchased from Adipogen Life Sciences. N_3-DNA-FAM was purchased from Integrated Device Technology, Inc. PEG-N_3 was synthesized from PEG methyl ether (MW~750 g/mol), which was purchased from BeanTown Chemical. c(RGDfK)-N_3 was purchased from Peptides International. BSA-N_3 was purchased from ProteinMods.

Buffers and Solvents: Phosphate-buffered saline (PBS), 4-morpholineethanesulfonic acid (MES), and tris(hydroxymethyl)aminomethane (Tris) were purchased from Millipore-Sigma. Acetonitrile (CH_3CN), dichloromethane (CH_2Cl_2), dioxane, dimethylformamide (DMF), ethanol (EtOH), and tetrahydrofuran (THF) were purchased from Millipore Sigma. Methanol (MeOH), diethyl ether (Et_2O), and ethyl acetate

(EtOAc) were purchased from Fisher Scientific. Deuterated solvents were purchased from either Cambridge Isotope Laboratories or Millipore Sigma. Deuterated solvents contained 0.05% (v/v) TMS as a secondary internal reference. Water was deionized and filtered to a resistivity of 18.2 ΩM with a Milli-Q® Plus water purification system (Millipore, Massachusetts). Buffers were prepared freshly in Milli-Q® water and their pH were adjusted using HCl or NaOH. For all experiments, the buffer concentration was 10 mM unless otherwise stated.

Materials: Poly(tetrafluoroethylene) (PTFE), poly(ether ether ketone) (PEEK), nylon, polycarbonate (PC), and silicone rubber (SiR) were purchased from McMaster-Carr. Glass was purchased from VWR International. Si/SiO_2 wafer substrate was purchased from University Wafer. All of these materials were cleaned ultrasonically in ethanol and water for 15 min before use. Commercially available pure titanium rods were cut into plates and polished up to 1200 grit using SiC paper. Ti-based materials were subjected to successive ultrasonic rinses in acetone, ethanol, and water for 15 min before use. Germanium (Ge) was purchased from Millipore-Sigma and used as received. Polypropylene (PP) membrane was purchased from Deschem Science Supply, China. Macroporous Ti-6Al-4V scaffolds were acquired from AKEC Medical Co. Ltd, China. Dental implant (ProActive Ø5.0 x 9mm) was purchased from Neoss Inc.

Biologics: Bacteria culture: *Staphylococcus aureus* (*S. aureus*, ATCC-6538) and *Escherichia coli* (*E. coli*, ATCC-25922) were purchased from American Type Culture Collection (ATCC). Mammalian cell culture: Human Umbilical Vein Endothelial Cells (HUVEC) were purchased from PromoCell, Germany. Mouse pre-osteoblast cell line MC3T3-E1 subclone 14 cells was purchased from ATCC (CRL-2594).

Material surface functionalization protocols

General single-step drop coating procedure: Solutions of $CuSO_4$ (5 mM), THPTA (10 mM), MOI-N_3 (1 μM – 0.5 mM), and p-DOPAmide (10–50 mM) were prepared using one of the following buffers: MES (pH 5.5), PBS (pH 7.4), or Tris (pH 8.5). These solutions were combined in an Eppendorf vial to provide a master coating mixture (MCM). Unless otherwise specified, all the buffers and reagent solutions were purged with N_2 gas (for ~15 min). A specified volume of the MCM was dropped onto a material in a tissue culture polystyrene plate (TCPS) or Petri dish, which was then sealed with Parafilm M (Bemis) and gently agitated on a shaker at 37 °C. After the coating is complete (typically within 30 min – 4 h unless otherwise stated), the substrate was rinsed thoroughly with Milli-Q water and dried under air and at RT.

Material substrates: Planar solid materials used in this study include Ti/TiO_2, Si/SiO_2, glass, polytetrafluoroethylene (PTFE), polyether ether ketone (PEEK), polycarbonate (PC), silicone rubber (SiR), and a dime coin. 2-dimensional porous or fibrous materials include nylon foam and polypropylene (PP) membrane. 3-dimensional objects include germanium pieces, a plastic polyhedral dice, mini dinosaur toy, cherry tomato (hydrophobic), lotus root (porous and hydrophilic), porous Ti-based tissue scaffold, and Ti-based dental implant.

Surface wettability: Static contact angles (CA) were measured at room temperature (RT) using the sessile drop method on a custom-built benchtop CA goniometer (L2004A1, Ossila) equipped with a video camera. Each time, 5-μL aliquots of Milli-Q water were added to the air side of sample surface and images were recorded after droplet spreading. Results are presented in Figure S1B.

Patterning of 2- or 3-dimensional objects: 1-2 μL droplets of the MCM (MOI-N_3 = 3-N_3-7-hydroxycoumarin, 0.5 mM) were added onto irregular surfaces of 3-dimensional objects (a plastic polyhedral dice and mini dinosaur toy), which were then incubated for 1 h.

Multiplexed patterning. 400 μL of the MCM containing two MOI-N_3 (3-N_3-7-hydroxycoumarin, 0.5 mM and N_3-DNA-FAM, 1 μM) was dropped onto a dime coin, which was then incubated for 4 h. In another example, 1-2 μL droplets of this MCM were added onto a cherry tomato and lotus root, which were both then incubated for 1 h. Other material-independent patterns were generated according to the following protocol: 1-5 μL droplets of the MCM (MOI-N_3 = 3-N_3-7-hydroxycoumarin or TAMRA-N_3, either 0.5 mM), were added onto substrates (Ti/TiO_2, glass, Si/SiO_2, and PEEK) to create micro-volume arrays or patterns.

Coating kinetics and mechanism

Effects of p-DOPAmide concentration and reaction time: Ti/TiO_2 was drop-coated for 1-12 h using 100-μL aliquots of the MCM (MOI-N_3 = 3-N_3-7-hydroxycoumarin, 0.5 mM; p-DOPAmide, 1-50 mM). Samples were taken, rinsed, and examined by X-ray photoelectron spectroscopy (XPS) and CLSM at 405 nm excitation. Concomitantly, the time-dependent evolution of various coating mixtures was monitored in small Eppendorf vials. Results are presented in Figure S3 (XPS) and Figure S4 (CLSM).

Effect of additives: Substrates were drop coated with 100-μL droplets of the MCM (MOI-N_3 = 3-N_3-7-hydroxycoumarin, 0.5 mM) that was supplemented with $FeCl_3$ (5 or 0.5 mM). Control substrates were drop coated without $FeCl_3$. After 4 h of incubation, each coated substrate was investigated by ambient light photography, XPS, and CLSM. Results are presented in Figure 2D and Figure 2E.

Effect of catechol protection: *O*-TBDMS-protected p-DOPAmide (compound ***S3***, see below) was used in this investigation. A mixture of $CuSO_4$ (5 mM), THPTA (10 mM), 3-N_3-7-hydroxycoumarin (0.5 mM), and ***S3*** (10 mM) was dropped onto Ti/TiO_2 substrates. Control substrates were drop coated with the MCM, which contained p-DOPAmide instead of ***S3***. After 4 h of incubation, each substrate was investigated by ambient light photography, XPS, and CLSM. Results are presented in Figure 2D and Figure 2E.

Grafting efficiency based on grafting density: Ti/TiO_2 and Si/SiO_2 were functionalized with TAMRA via five different methods (**i**: drop coating; **ii-v**: dip coating):

(i) *Single-step drop coating with p-DOPAmide*. An MCM (MOI-N_3 = TAMRA-N_3, 0.5 mM) was prepared using a PBS buffer (pH 7.4). A 100-μL droplet of this MCM was dropped onto the substrate, which was then incubated at 37 °C for 6 h.

(ii) *Single-step dip coating with p-DOPAmide*. 500-μL of the same MCM was added into a 24-well TCPS containing the substrate, which was then incubated at 37 °C for 6 h.

(iii) *Stepwise dip coating with p-DOPAmide*. Substrates were immersed in 10 mM of p-DOPAmide in Tris buffer (pH 8.5) at RT for 3 d. The resulting substrates were then incubated at 37 °C for 6 h with a N_2-bubbled mixture of $CuSO_4$ (5 mM), THPTA (10 mM), TAMRA-N_3 (0.5 mM), and sodium ascorbate (25 mM), which was buffered with PBS (pH 7.4).

(iv) *Single-step dip coating with DA*.[1] A mixture of DA (10 mM) and TAMRA-NH_2 (0.5 mM) was prepared using Tris buffer (pH 8.5). Substrates were immersed into this mixture and incubated at 37 °C for 24 h.

(v) *Stepwise dip coating with DA*.[2] Substrates were immersed into a solution of DA (10 mM) buffered with Tris (pH 8.5) and incubated at 37 °C for 24 h. The resulting substrates were then incubated with TAMRA-NH_2 (0.5 mM) at 37 °C for 6 h.

The resulting substrates were rinsed with Milli-Q water, dried under air at RT, and visualized by confocal laser scanning microscopy (CLSM). Images of each triplicate samples were analyzed by ImageJ to quantify the averaged fluorescence intensities. For each substrate, the lowest intensity was normalized to au of 1.0. Accordingly, the relative fluorescence intensities using methods **(i)-(v)** were found as: For Ti/TiO_2, **(i)**: 7.0, **(ii)**: 3.5, **(iii)**: 6.4, **(iv)**: 1.1, and **(v)**: 1.0. For Si/SiO_2, **(i)**: 7.9, **(ii)**: 3.4, **(iii)**: 3.7, **(iv)**: 1.2, and **(v)**: 1.0. Results are also presented in Figure 4.

Cu(II)-Lig assisted p-DOPAmide oxidative polymerization in solution: Solutions of **(i)** p-DOPAmide, **(ii)** p-DOPAmide + Lig, **(iii)** p-DOPAmide + Cu, and **(iv)** p-DOPAmide + Cu + Lig were prepared using a buffer (MES pH 5.5, PBS pH 7.4, or Tris pH 8.5) with or without N_2 bubbling. For all solutions, the

concentrations of p-DOPAmide, THPTA, and $CuSO_4$ were 10 mM, 10 mM, and 5 mM, respectively. Solutions that were not bubbled with N_2 were maintained in open Eppendorf vials at RT for 4 h. Subsequently, these vials were loosely covered with a lid to avoid evaporation. Solutions that were bubbled with N_2 (vigorously for 15 min) were transferred into Eppendorf vials, which were tightly sealed with a lid. Lig: Ligand and Cu: $CuSO_4$. Results are presented in Figure S5.

Catechol-assisted Cu(I) production from Cu(II): Cu(I) production was evaluated using a modified MicroBCA assay. This assay relies on the reduction of Cu(II) ions into Cu(I) ions by proteins (testing samples) in an alkaline buffer (kit component A), which complex with bicinchoninic acid (BCA, kit component B) to give a characteristic violet color. In fact, similar coloration could be generated by using any reagent (in addition to proteins) that has the capability to reduce Cu(II) into Cu(I).[3] In our assay, the following testing solutions were fresh prepared using PBS: **(i)** $CuSO_4$ (5 mM); **(ii)** THPTA (10 mM); **(iii)** $CuSO_4$ (5 mM) and THPTA (10 mM); **(iv)** p-DOPAmide (10 mM); **(v)** THPTA (10 mM) and p-DOPAmide (10 mM); **(vi)** $CuSO_4$ (5 mM) and p-DOPAmide (10 mM); **(vii)** $CuSO_4$ (5 mM), THPTA (10 mM), and p-DOPAmide (10 mM); **(viii)** $CuSO_4$ (5 mM), THPTA (10 mM), and ***S3*** (10 mM). These solutions were mixed with MicroBCA kit components A and B at a ratio of 1:5:5. The resulting mixtures were shaken at 37 °C for 30 min to facilitate coloration, and the absorbance was measured at 570 nm. Results are presented in Figure 2F and Figure S7.

Interfacial film formation in drop coating: *Material-independent interfacial film formation*. The MCM (MOI-N_3 = 3-N_3-7-hydroxycoumarin, 0.5 mM) was used to coat substrates of these materials: Ti/TiO_2, Si/SiO_2, Ge, glass, PTFE, PEEK, PC, nylon, SiR, and quartz cells. Film debris was collected by washing the substrates and characterized by atomic force microscopy (AFM), transmission electron microscopy (TEM), Raman spectroscopy, and CLSM. Results are presented in Figure 3A–3B and Figure S9–S14.

General methods for biological applications

Surface functionalization for antifouling: 200-μL droplets of the MCM (MOI-N_3 = PEG-N_3, 0.5 % v/v) were dropped onto PP membranes. The membrane was then incubated at 37 °C for 4 h. To investigate the antifouling properties of the PP surfaces, both uncoated and PEG-functionalized PP membranes were incubated with FITC-BSA (100 μg/mL) at 37 °C for 2 h. The resulting membranes were thoroughly rinsed with PBS buffer. The surface-retained proteins were fixed by 4 (v/v) % paraformaldehyde for 10 min. The final samples were examined by CLSM at 488 nm excitation. Results are presented in Figure 6.

Surface functionalization for antibacterial/antibiofilm property: 100-μL droplets of the same MCM mixture (above) were added onto Ti/TiO_2 substrates, which were then incubated at 37 °C for 4 h. Prior to antibacterial tests, all specimens were sterilized in 75% (v/v) EtOH for 20 min and rinsed three times with a sterile PBS buffer. Bacteria culture: *S. aureus* and *E. coli* strains were cultured using Luria–Bertani (LB) broth or LB agar plates at 37 °C. In brief, bacterial cells were shaken (180 rpm) overnight in LB broth and then sub-cultured to a concentration of ~2×10^8 CFU/mL. The resulting suspensions were diluted to desired concentrations for further tests. Antiadhesion assays: 1.0×10^5 CFU/mL of *S. aureus* and *E. coli* strains were inoculated on uncoated and PEG-functionalized Ti/TiO_2 and cultivated for 1 h at 37 °C to allow attachment. To detach surface-adhered bacteria, samples were rinsed gently with PBS and transferred to a sterile Eppendorf tube with 1 mL of fresh LB broth and then sonicated for 10 min. After ten-fold serial dilutions, the suspensions were spread onto LB agar plates and grew overnight to foster colony formation. The LIVE/DEAD® Baclight™ kit was adopted to stain and *in situ* visualize surface-adhered bacteria under CLSM. Briefly, 400 μL of SYTO (6 μM) and propidium iodide (30 μM) stain mixtures were added to each specimen and maintained for 15 min in darkness. Antibiofilm assays: *S. aureus* (1.0×10^5 CFU/mL) was inoculated and cultivated for 5 d at 37 ºC. Thereafter, samples were rinsed gently with PBS to remove loosely adherent species, followed by LIVE/DEAD staining. CLSM imaging was performed to visualize the bacteria within biofilms. Alternatively, the biomass was quantified using a crystal violet staining method. Samples were fixed in 4% PFA and stained with 0.1% (w/v) crystal violet for 15 min, and washed with PBS buffer gently to remove excess reagents. The stained sample was dissolved in 95% (v/v) EtOH, and absorbance of the solution was measured at 570 nm. Results are presented in Figure 6.

Surface functionalization for mammalian cell studies. The MCM mixture ((MOI-N_3 = BSA-N_3 or c(RGDfK)-N_3, either 20 μg/mL, or PEG-N_3, 0.1 % (v/v); $CuSO_4$ (0.5 mM), THPTA (1 mM), and p-DOPAmide (1 mM)) was dropped onto a material substrate, which was then incubated at 37 ºC for 4 h. Specimens were sterilized in 75% (v/v) EtOH for 20 min and rinsed three times with sterile PBS prior to cell studies. Results of the experiments described here are presented in Figures 7 and 8 and Figures S18–S20.

Cell culture: HUVECs were cultured in Endothelial Cell Growth Medium 2 supplemented with Supplement Mix (PromoCell). Pre-osteoblastic MC3T3-E1 cells were cultured in alpha Minimum Essential Medium (α-MEM, Sigma) supplemented with 1% penicillin-streptomycin (hereafter termed as 1% pen-strep, VWR International), and 10% (v/v) fetal bovine serum (FBS, Hyclone Laboratories Inc.)

as the growth medium. For osteogenic differentiation, α-MEM that contain the following was used: 1% pen-strep, 10% FBS, 50 μg/mL of ascorbic acid, 10 mM of β-glycerol phosphate, and 100 nM of dexamethasone as the osteogenic medium. Cultures were maintained at 37 ºC in a humidified 5% CO_2 atmosphere. Medium was refreshed every 2–3 d. Sub-confluent cells were harvested using 0.05% trypsin-EDTA, collected by centrifugation, and then resuspended to a desired density prior to seeding.

MOI-regulated adhesion of HUVECs: HUVECs (5×10^4 cells/mL) were seeded onto a drop coated Ti/TiO_2 substrate in 24-well TCPS plates and cultured at 37 ºC for 12 h. After the culturing, the cells were stained with a FAK100 kit per manufacturer instructions: Cells were fixed in 4% paraformaldehyde for 10 min, permeabilized in 0.1% Triton X-100 for 2 min, and blocked with 1% BSA/PBS for 30 min. The resulting cells were incubated with an anti-vinculin monoclonal antibody (1:500 dilution) at RT for 1 h, and stained with the following dyes: FITC-conjugated goat anti-mouse IgG (1:100 dilution; 1 h), TRITC-conjugated phalloidin (1:500 dilution; 1 h), and DAPI (1:1000 dilution; 5 min). After thorough rinses, CLSM images of the samples were recorded in a multitrack mode, wherein actin cytoskeleton (via TRITC-phalloidin), focal adhesion (via anti-vinculin), and nuclei (via DAPI) were visualized as red, green, and blue, respectively.

c(RGDfK)-mediated site-selective adhesion of HUVECs: Ti/TiO_2 surface was drop coated with 1 μL of coating mixture containing c(RGDfK)-N_3. This partially functionalized surface was seeded with HUVECs (1×10^5 cells/mL) and incubated at 37 ºC for 24 h. Thereafter, the cells were stained with 2 μM of Calcein AM for 10 min and investigated by CLSM.

c(RGDfK)-mediated adhesion of MC3T3-E1 cells. Ti/TiO_2, Si/SiO_2, PEEK, and PTFE substrates were drop coated with 100–400 μL of c(RGDfK)-N_3. The uncoated and coated materials were incubated with MC3T3-E1 cells (2×10^4 cells/mL) at 37 ºC for 4 h. Cell adhesion and survival were assessed by staining with Calcein AM (2 μM) and PI (4 μM) for 15 min. Cells were imaged by CLSM, wherein living and dead cells were colored as green and red, respectively. In addition, at 24 h, cytoskeletons for selected cultures were stained with TRITC-conjugated phalloidin and imaged by CLSM.

Cytotoxicity assay: Ti/TiO_2 was drop coated with c(RGDfK)-N_3 as described above. The uncoated and coated specimens were each immersed in serum-free α-MEM (1.25 cm^2/mL) at 37 ºC for 72 h. The leaching fluids (extracts) were collected and supplemented with 10% (v/v) FBS prior to use. Cytotoxicity was evaluated by using an MTT assay according to manufacturer's instructions. MC3T3-E1 cells (5×10^4 cells/mL) were seeded in 96-well TCPS plates and incubated for 24 h to allow attachment. Afterwards,

the medium was replaced with 100 µL of extracts. After day-1 and day-3, the medium was discarded, and 100 µL of serum-free α-MEM containing 50% (v/v) MTT solution was added to each well. The plates were incubated at 37 °C for 3 h to yield formazan crystals. The formazan was solubilized in an MTT solvent, and its absorbance was measured on a microplate reader (Molecular Devices, SpectraMax iD3) at 590 nm. Alternatively, MC3T3-E1 cells (5×10^4 cells/mL) were seeded onto a glass surface and incubated for 24 h. The cells were then treated with material extracts as described above. The cells were stained with calcein AM (2 µM) and propidium iodide (4 µM) and investigated by CLSM.

Tissue engineering. Macroporous Ti-6Al-4V scaffolds were dip coated with 1 mL of the MCM (MOI-N_3 = c(RGDfK)-N_3). MC3T3-E1 cells were seeded at a density of 2×10^5 cells/mL and incubated at 37 ºC. After 4 d, cell adhesion was evaluated by staining with Calcein AM (2 µM) and PI (4 µM) for fluorescence imaging. After 7 d, cytoskeleton development was evaluated by staining with TRITC-Phalloidin for fluorescence imaging.

In vitro osteogenesis on dental implant. A commercially available dental implant was partially coated with c(RGDfK)-N_3 as described above. MC3T3-E1 cells (2×10^5 cells/mL) were seeded and the implant was incubated at 37 ºC in a growth medium for 7 d. Afterwards, the medium was replaced by osteogenic medium and cultivation was prolonged up to 28 d. The sample was rinsed with PBS thrice and fixed in 2.5% (v/v) glutaraldehyde in PBS for 2 h, and then dehydrated in a gradient of ethanol (50%-100% v/v) for 15 min each. The sample was dried in air and investigated by scanning electron microscopy (SEM). Alternatively, bony tissues were partially detached, and AFM was used to investigate the topography of extracellular matrix.

Surface characterization techniques

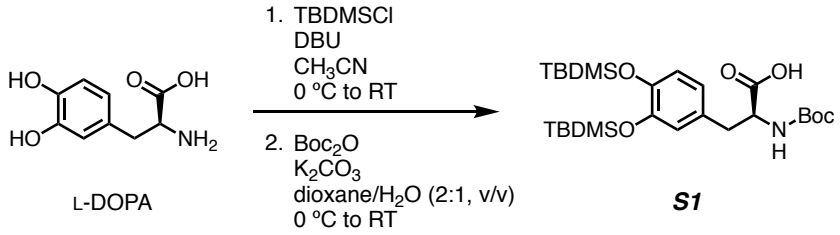
AFM was carried out in flapping mode for surface topography and roughness using a Cypher microscope (Asylum Research) and standard SiN cantilevers (AC160, Asylum). Additionally, the coating thickness was determined as height differences between the coating and a scratched area. SEM (Zeiss Sigma, German) was performed at an accelerating voltage of 2 keV under vacuum. TEM (JEOL JEM 2010F, Japan) was operated at 200 keV for microstructural images with aid of a digital camera. Fourier transform infrared spectroscopy (FTIR) was used to probe surface functional groups on a PerkinElmer Spectrum 100 Spectrometer (PerkinElmer) equipped with an attenuated total reflectance (ATR) accessory. Spectra were acquired in the range of 580–4000 cm^{-1} with 32 scans for each spectrum at a resolution of 1 cm^{-1}. Micro-Raman spectra were recorded on a Raman microscope (Renishaw inVia, Ar^+ 532 nm, UK). For

element compositions, XPS (K-Alpha™, Thermo Scientific) investigations were conducted using monochromatic Al Kα source (hν = 1486.6 eV) at an energy step of 0.05 eV (core-level spectra) or 0.5 eV (survey spectra). CLSM (Zeiss LSM780) was employed for fluorescence imaging under various scanning modes: XY, Z-stacks, and tile scans as if necessary. For Z-stacks, a series of sliced images were acquired and later reconstructed into a 3-dimentional field of view. For tile scans, a motorized stage was driven by the ZEN software (Zeiss) to capture multi-field images across the XY plane, which were merged into a full-scale image. Images were processed and analyzed using either ZEN or ImageJ.

General synthetic methods

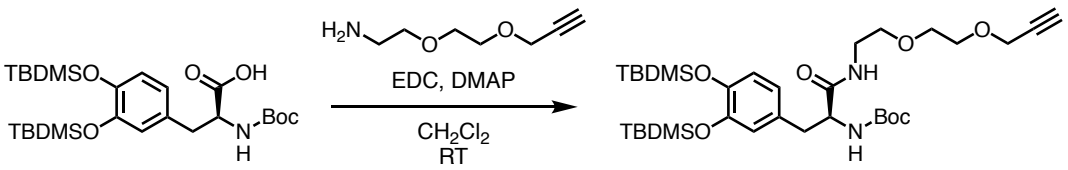
For the chemical synthesis of organic compounds, all reactions were performed under a dry nitrogen atmosphere unless otherwise stated. All glassware was oven-dried before use. Purification of the synthesized compounds was performed using a Büchi Reveleris® flash chromatography system equipped with a FlashPure EcoFlex silica (50 μm sphere) column. Observed rotation (α_{obs}) values were measured in a standard glass cell (100 mm, 1 mL) using a sodium D-line lamp at 20 ºC in a PerkinElmer Model 241 Polarimeter. Specific rotation [α] was calculated based on $[\alpha]^{20}_{D} = (\alpha_{obs})/[(g_{sample}$ in 1 mL) x 1 dm]. Nuclear Magnetic Resonance (NMR) spectroscopic analyses were carried out on either a Varian VNMRS 500 MHz or Bruker Avance Neo 500 MHz spectrometer. NMR data is provided for new compounds. ^{1}H NMR spectra were acquired at 500 MHz and ^{13}C NMR spectra were acquired at 125 MHz. Chemical shifts (δ) for ^{1}H NMR spectra were referenced to (C***H***$_3$)$_4$Si at δ = 0.00 ppm, to CD$_2$***H***CN at δ = 1.94 ppm, to C***H***D$_2$S(O)CD$_3$ at δ = 2.50 ppm, or to C***H***Cl$_3$ at δ = 7.27 ppm. ^{13}C NMR spectra were referenced to ***C***D$_3$S(O)***C***D$_3$ at δ = 39.51 ppm, to ***C***DCl$_3$ at δ = 77.23 ppm, or to CD$_3$***C***N at δ = 118.70 ppm. The following abbreviations are used to describe NMR resonances: s (singlet), d (doublet), t (triplet), m (multiplet), br (broad), and nfom (non-first order multiplet). Coupling constants (*J*) are reported in Hz. Low-resolution mass spectroscopy (LRMS) analysis was performed using a Finnigan LCQ™ DUO mass spectrometer. Liquid chromatography followed by high-resolution mass spectroscopy (LC-HRMS) analysis in the ESI mode was carried out on a Waters Acquity-Xevo G2-XS QTof.

Preparation procedures and characterization data for compounds



Synthesis of (*S*)-3-(3,4-bis((tert-butyldimethylsilyl)oxy)phenyl)-2-((tert-butoxycarbonyl)-amino)-propanoic acid, *S1*: (Protocol modified from Lee et al[4]). To a 100-mL round-bottom flask was added L-DOPA (3.0 g, 15.2 mmoles, 1 equiv), TBDMSCl (6.8 g, 45.7 mmoles, 3 equiv), and anhydrous CH_3CN (30 mL). The heterogenous mixture was cooled at 0 °C while being stirred vigorously and purged with N_2 gas continuously. To the cooled mixture, DBU (6.9 mL, 45.7 mmoles, 3 equiv) was added dropwise and N_2 purging was stopped. The resulting mixture was stirred at RT for 16 h and then filtered. The filtrate was cooled at 0-4 ºC for 15 min, after which bis-*O*-TBDMS-protected DOPA crushed out of the liquid as an amorphous white solid. This product was collected by vacuum filtration, washed with cold EtOH (-20 ºC). Subsequent crops of the product were collected from the mother liquor via the same process. The product batches were combined and dried to give pure bis-*O*-TBDMS-protected DOPA (5 g, 77% isolated yield), which was used in the next step.

A 100-mL round-bottom flask was charged with K_2CO_3 (4.2 g, 30.4 mmoles, 3 equiv), dioxane (20 mL) and water (10 mL). To the mixture cooled at 0 °C, was added bis-*O*-TBDMS-protected DOPA (4.3 g, 10.0 mmoles, 1 equiv) and Boc_2O (2.4 g, 11.1 mmoles, 1.1 equiv). After being stirred at RT for 1 d, the reaction mixture was diluted with water (10 mL) and treated with acetic acid until the pH reached to ca. 4.7. The resulting mixture was extracted twice with CH_2Cl_2. The organic layers were washed with water and brine, dried over Na_2SO_4, and concentrated under reduced pressure to afford the known carboxylic acid[4] ***S1*** (4.9 g, 75% isolated yield over 2 steps). The product was used in the next step without further purification. The stereochemistry of *N*-Boc-DOPA has been reported to be maintained under saponification conditions that use K_2CO_3.[5] This, combined with the fact that we did not obtain a racemic product in subsequent chemical steps, suggests that epimerization is unlikely.



S1 ***S2***

Synthesis of tert-butyl (*S*)-(3-(3,4-bis((tert-butyldimethylsilyl)oxy)phenyl)-1-oxo-1-((2-(2-(prop-2-yn-1-yloxy)ethoxy)-ethyl)amino)propan-2-yl)carbamate, *S2*: To a stirred mixture of ***S1*** (3.4 g, 6.5 mmoles, 1.0 equiv) and EDC (1.5 g, 7.8 mmoles, 1.2 equiv) in CH_2Cl_2 (60 mL) at RT, was added a mixture of 2-[2-(2-propynyloxy)ethoxy]ethylamine (1.2 g, 8.4 mmoles, 1.3 equiv) and DMAP (0.2 g, 1.8 mmoles, 0.3 equiv) in CH_2Cl_2 (5 mL). After being stirred at RT for 12 h, the reaction mixture was quenched with water (20 mL). The resulting heterogenous mixture was extracted twice with CH_2Cl_2. The organic layers were washed with water and brine, dried over Na_2SO_4, and concentrated under reduced pressure. The resulting crude residue was purified by silica gel flash chromatography (CH_2Cl_2/MeOH step gradient) to afford ***S2*** (2.3 g, 55% isolated yield).

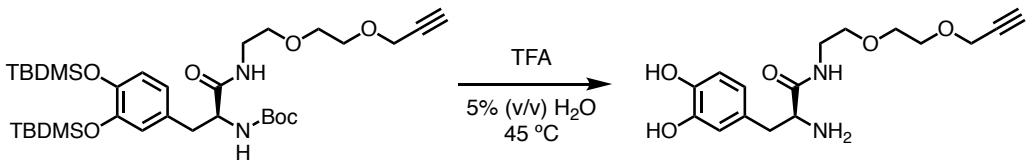
R_f = 0.35 (CH_2Cl_2/MeOH 95:5).

^{1}H NMR (500 MHz, $CDCl_3$) δ 6.74 (d, *J* = 8.0 Hz, 1H), 6.66 (d, *J* = 2.0 Hz, 1H), 6.63 (dd, *J* = 8.0, 2.0 Hz, 1H), 6.26 (br t, *J* = 5.0 Hz, 1H), 4.95 (br s, 1H), 4.26 (br s, 1H), 4.19 (t, *J* = 2.5 Hz, 2H), 3.65 (ddd, *J* = 6.0, 4.0, 2.0 Hz, 2H), 3.59 (ddd, *J* = 6.0, 4.0 Hz, 2H), 3.52–3.35 (overlapping m, 4H), 2.94 (apparent d, *J* = 6.0, 2H), 2.44 (t, *J* = 2.5, 1H), 1.41 (s, 9H), 0.99 (s, 9H), 0.98 (s, 9H), 0.19 (overlapping s, 6H), and 0.18 (overlapping s, 6H).

^{13}C NMR (125 MHz, $CDCl_3$) δ 171.4 (*C*=O), 155.5 (C=O), 146.9 (Ar-C), 146.0 (Ar-C), 129.8 (Ar-C), 122.3 (Ar-C), 122.4 (Ar-C), 121.2 (Ar-C), 80.2 (C), 79.7 (C≡C), 75.0 (C≡C), 70.3, 70.0, 69.2, 58.6, 55.9, 39.4, 38.0, 28.5 (3 *C*H_3), 26.1 (overlapped 6 *C*H_3), 18.62 (C), 18.60 (C), -3.82 (*C*H_3), -3.85 (*C*H_3), -3.87 (*C*H_3), and -3.90 (*C*H_3).

HRMS (ESI): Calculated for [$C_{33}H_{58}N_2O_7Si_2 + Na^+$] 673.3675, found 673.3740.

$[\alpha]^{20}_D$ = +7.5° (c = 4 g/100mL, 0.04 g/mL, $CHCl_3$).



S2 ***p-DOPAmide***

Synthesis of (*S*)-2-amino-3-(3,4-dihydroxyphenyl)-*N*-(2-(2-(prop-2-yn-1-yloxy)ethoxy)ethyl)-propanamide, Propargyl-2EG-DOPAmide (p-DOPAmide): A 10-mL pressure flask was charged with ***S2*** (1.5 g, 2.3 mmoles, 1 equiv), TFA (4 mL, 52.6 mmoles, 23 equiv), and Milli-Q® water (0.2 mL, 11.1 mmoles, 5 equiv). The flask was sealed with a PTFE cap and placed in an oil bath heated at 45 ºC. The reaction solution was stirred for 4 h, then allowed to cool to RT, and was slowly added into cooled (-30 ºC) and stirred Et_2O (60 mL). The resulting suspension was vortexed and the supernatant was removed. The remaining precipitate was washed twice with Et_2O and kept under high vacuum for 1 h. The resulting white solid essentially contained **p-DOPAmide** as an ammonium trifluoroacetate salt (0.8 g, 80% isolated yield) in a powder form with an opaque white color, which turns into a wax at RT upon handling.

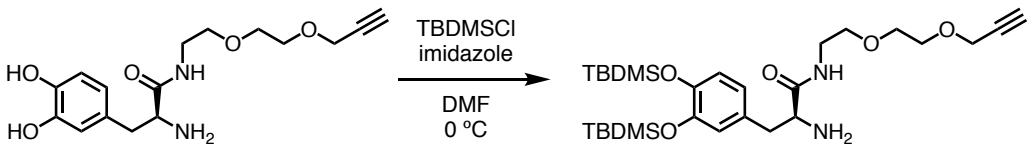
^{1}H NMR (500 MHz, d_6-DMSO) δ 8.88 (s, 1H), 8.84 (s, 1H), 8.43 (br t, *J* = 5.0 Hz, 1H), 8.08 (br s, 3H), 6.66 (d, *J* = 8.0 Hz, 1H), 6.61 (d, *J* = 2.0 Hz, 1H), 6.47 (dd, *J* = 8.0, 2.0 Hz, 1H), 4.13 (d, *J* = 2.5 Hz, 2H), 3.83 (br t, *J* = 6.5 Hz, 1H), 3.55 (m, 2H), 3.51 (m, 2H), 3.42 (m, *J* = 2.5, 1H), 3.39 (m, 1H), 3.31 (m, 2H), 3.18 (m, 1H), 2.84 (dd, *J* = 14.0, 6.5, 1H), and 2.75 (dd, *J* = 14.0, 7.5, 1H).

^{13}C NMR (125 MHz, d_6-DMSO) δ 168.1 (*C*=O), 145.2 (Ar-C), 144.2 (Ar-C), 125.4 (Ar-C), 120.1 (Ar-C), 116.8 (Ar-C), 115.5 (Ar-C), 80.3 (C≡C), 77.2 (C≡C), 69.3, 68.8, 68.5, 57.5, 53.8, 38.7, and 36.6.

HRMS (ESI): Calculated for trifluoroacetate-free product [$C_{16}H_{22}N_2O_5 + H^+$] 323.1601, found 323.1594.

LRMS (ESI): Calculated for ammonium trifluoroacetate salt product [$C_{18}H_{23}F_3N_2O_7 + H^+$] 437.2, found 437.3.

$[\alpha]^{20}_D$ = +16.0º (c = 3.0 g/100mL, 0.030 g/mL, 1 M HCl).



p-DOPAmide ***S3***

Synthesis of (*S*)-2-amino-3-(3,4-bis((tert-butyldimethylsilyl)oxy)phenyl)-*N*-(2-(2-(prop-2-yn-1-yloxy)ethoxy)ethyl)-propanamide, *S3*: To a stirred solution of the trifluoroacetate salt of ***p-DOPAmide*** (116 mg, 0.27 mmoles, 1.0 equiv) in anhydrous DMF (0.7 mL) at 0 ºC, was added TBDMSCl (140 mg, 0.93 mmoles, 3.5 equiv) and imidazole (275 mg, 4.1 mmoles, 15.0 equiv). After being stirred for 12 h, the reaction mixture was diluted with CH_2Cl_2 (10 mL) and stirred with 1 M $NaHCO_3$ solution (2 mL) and 1 M NaCl solution (3 mL). The resulting emulsion was vortexed and the aqueous layer was discarded. The isolated organic layer was washed with 1 M NaCl solution (3 mL) twice and concentrated under reduced pressure. The resulting crude residue was purified by silica gel flash chromatography (CH_2Cl_2/MeOH step gradient) to afford ***S3*** (138 mg, 93% isolated yield) in a colorless oil form.

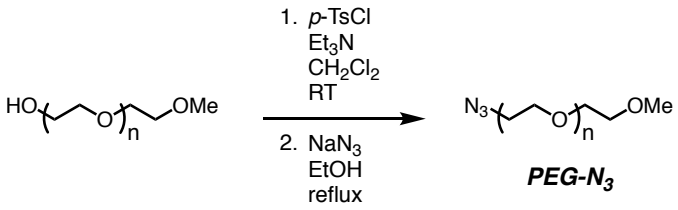
R_f = 0.25 (CH_2Cl_2/MeOH 97:3).

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.56 (br t, *J* = 5.5 Hz, 1H), 6.76 (d, *J* = 8.0 Hz, 1H), 6.70 (d, *J* = 2.0 Hz, 1H), 6.65 (dd, *J* = 8.0, 2.0 Hz, 1H), 4.21 (d, *J* = 2.5 Hz, 2H), 3.69 (ddd, *J* = 10.0, 4.0, 1.0 Hz, 2H), 3.65 (ddd, *J* = 10.0, 4.0, 1.0 Hz, 2H), 3.57 (t, *J* = 5.0 Hz, 2H), 3.52 (dd, *J* = 4.0, 1.0 Hz, 1H), 3.47 (m, 2H), 3.16 (dd, *J* = 14.0, 4.0, 1H), 2.51 (dd, *J* = 14.0, 4.0, 1H), 2.43 (t, *J* = 2.5, 1H), 1.36 (br s, 2H), 0.984 (s, 9H), 0.982 (s, 9H), 0.191 (overlapping s, 6H), and 0.189 (overlapping s, 6H).

^{13}C NMR (125 MHz, $CDCl_3$) δ 174.7 (*C*=O), 147.1 (Ar-C), 145.9 (Ar-C), 131.2 (Ar-C), 122.5 (Ar-C), 122.2 (Ar-C), 121.2 (Ar-C), 79.8 (C≡C), 74.8 (C≡C), 70.3, 70.1, 69.2, 58.7, 56.9, 40.6, 39.0, 26.14 (3 *C*H_3), 26.13 (3 *C*H_3), 18.6 (overlapping 2 C), -3.85 (overlapping 2 CH_3), -3.87 (*C*H_3), and -3.88 (*C*H_3).

HRMS (ESI): Calculated for [$C_{28}H_{50}N_2O_5Si_2 + Na^+$] 573.3150, found 573.3161.

$[\alpha]^{20}_D$ = -21.1º (c = 1.8 g/100mL, 0.018 g/mL, $CHCl_3$).



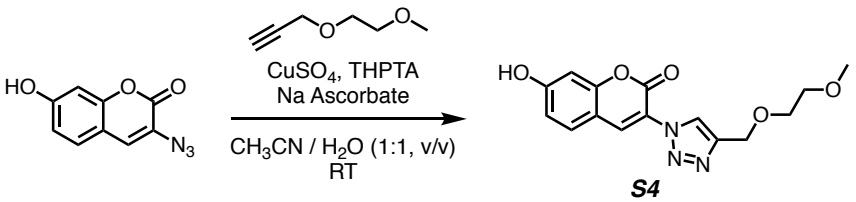
Synthesis of PEG-N_3: To a stirred solution of the PEG methyl ether (MW~750 g/mol, 2.00 g, 2.7 mmoles, 1.0 equiv) in CH_2Cl_2 (26 mL) at RT, was added Et_3N (0.54 mL, 5.3 mmoles, 2.0 equiv) and *p*-TsCl (0.76 g, 4.0 mmoles, 1.5 equiv). After being stirred for 12 h, the reaction mixture was diluted with CH_2Cl_2 (50 mL) and washed with 1 N HCl (20 mL), saturated $NaHCO_3$ (20 mL), water (20 mL), and brine (20 mL). The organic layer was dried over Na_2SO_4 and concentrated under reduced pressure. The resulting crude residue was then added to a suspension of NaN_3 in EtOH (45 mL). The reaction mixture was refluxed for 12 h, and then diluted with water (50 mL). Bulk EtOH was removed from the mixture using rotary evaporation and the aqueous phase was extracted with CH_2Cl_2 (100 mL). The organic layer was dried over Na_2SO_4 and concentrated under reduced pressure. The resulting crude residue was purified by silica gel flash chromatography (CH_2Cl_2/MeOH step gradient) to afford ***PEG-N_3*** (1.4 g, 73% isolated yield over 2 steps) in a colorless oil form.

R_f = 0.45 (CH_2Cl_2/MeOH 98:2).

^{1}H NMR (500 MHz, $CDCl_3$) δ 3.69 (br t, *J* = 5.0, 2H), 3.63–3.68 (overlapping 58 H), 3.54–3.56 (m, 2 H, C*H*$_2$CH$_2$N$_3$), 3.40 (br t, *J* = 5.0, 2H, C*H*$_2$N$_3$), and 3.38 (s, 3 H, OC*H*$_3$).

^{13}C NMR (125 MHz, $CDCl_3$) δ 72.0, 70.71, 70.68, 70.65, 70.62, 70.58 (overlapping –*C*H$_2$-*C*H$_2$-O–), 70.5, 70.0, 59.1 (*C*-OMe), and 50.7 (*C*-N$_3$).

LRMS (ESI): Calculated for [$C_{33}H_{67}N_3O_{16}$ + Na^+] 784.9 (Avg MW), found 784.7.



S4

Synthesis of 7-hydroxy-3-(4-((2-methoxyethoxy)methyl)-1H-1,2,3-triazol-1-yl)-2H-chromen-2-one, *S4*: To a stirred solution of 3-Azido-7-hydroxycoumarin (77 mg, 0.38 mmoles, 1.0 equiv) in acetonitrile (6 mL) and Milli-Q® water (6 mL) in a 20-mL amber vial, was added 3-(2-methoxyethoxy)prop-1-yne (86 mg, 0.76 mmoles, 2.0 equiv), $CuSO_4$ (30 mg, 0.19 mmoles, 0.5 equiv), and THPTA (83 mg, 0.19 mmoles, 0.5 equiv). The resulting homogenous liquid was purged with N_2 gas for 5 min. Sodium ascorbate (75 mg, 0.38 mmoles, 1.0 equiv) was added and the liquid was purged with N_2 gas for another 5 min, immediately after which the vial was sealed. The reaction mixture (~12 mL) was stirred at RT for 12 h and concentrated down to 1 mL under reduced pressure and by successive additions of acetonitrile to help remove bulk water. The resulting liquid was diluted with CH_2Cl_2 (2 mL) and purified by silica gel flash chromatography (CH_2Cl_2/MeOH step gradient) to afford ***S4*** (107 mg, 88% isolated yield) as a liquid with a lime green color that glows.

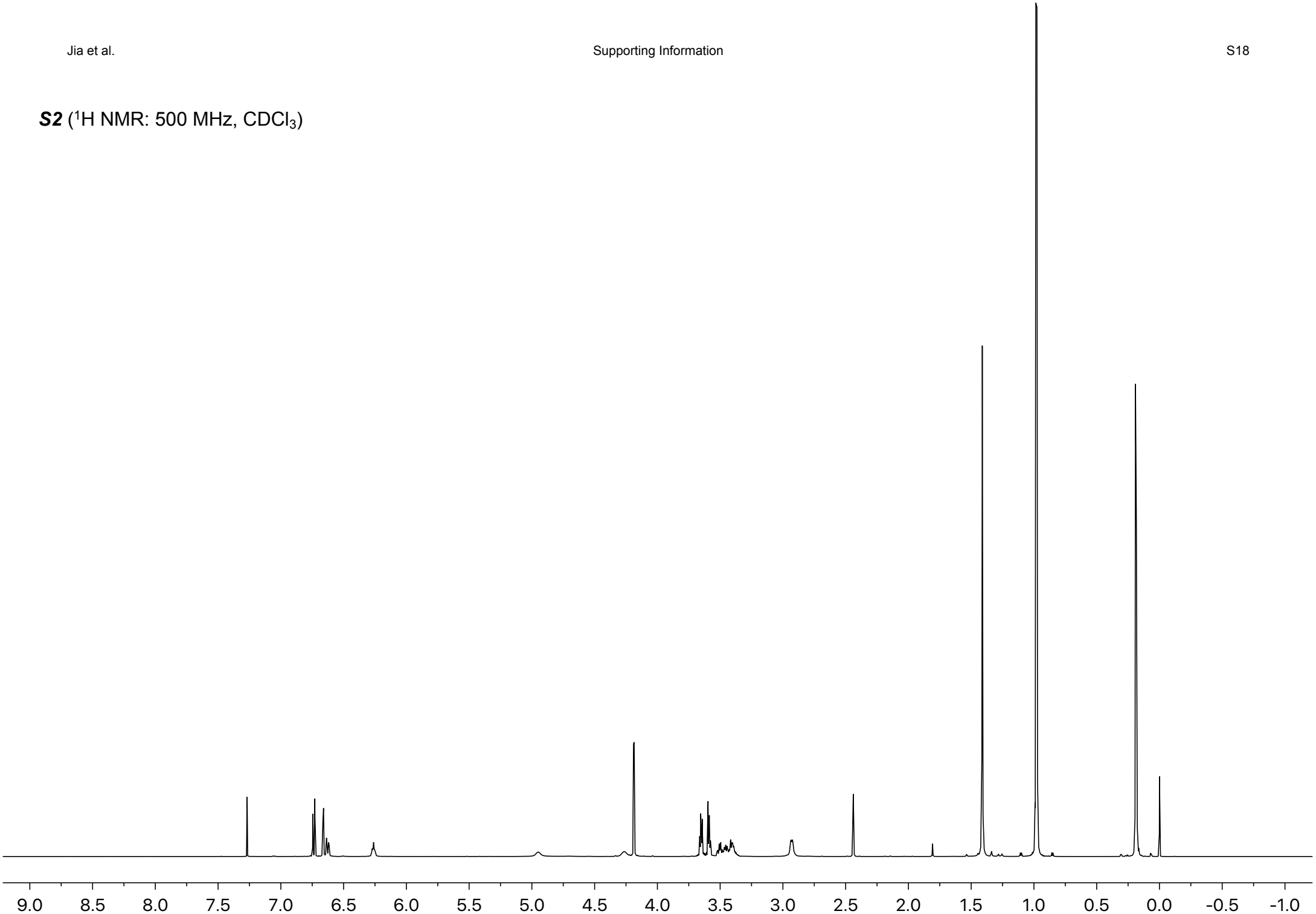
R_f = 0.35 (CH_2Cl_2/MeOH 97:3).

^{1}H NMR (500 MHz, d_6-DMSO) δ 8.59 (s, 1 H), 8.52 (s, 1H), 7.74 (d, *J* = 8.5, 1H), 6.91 (dd, *J* = 8.5, 2.0, 1H), 6.85 (d, *J* = 2.0, 1H), 4.62 (s, 1 H), 3.61 (m, 1 H), 3.48 (m, 1 H), and 3.25 (m, 3 H).

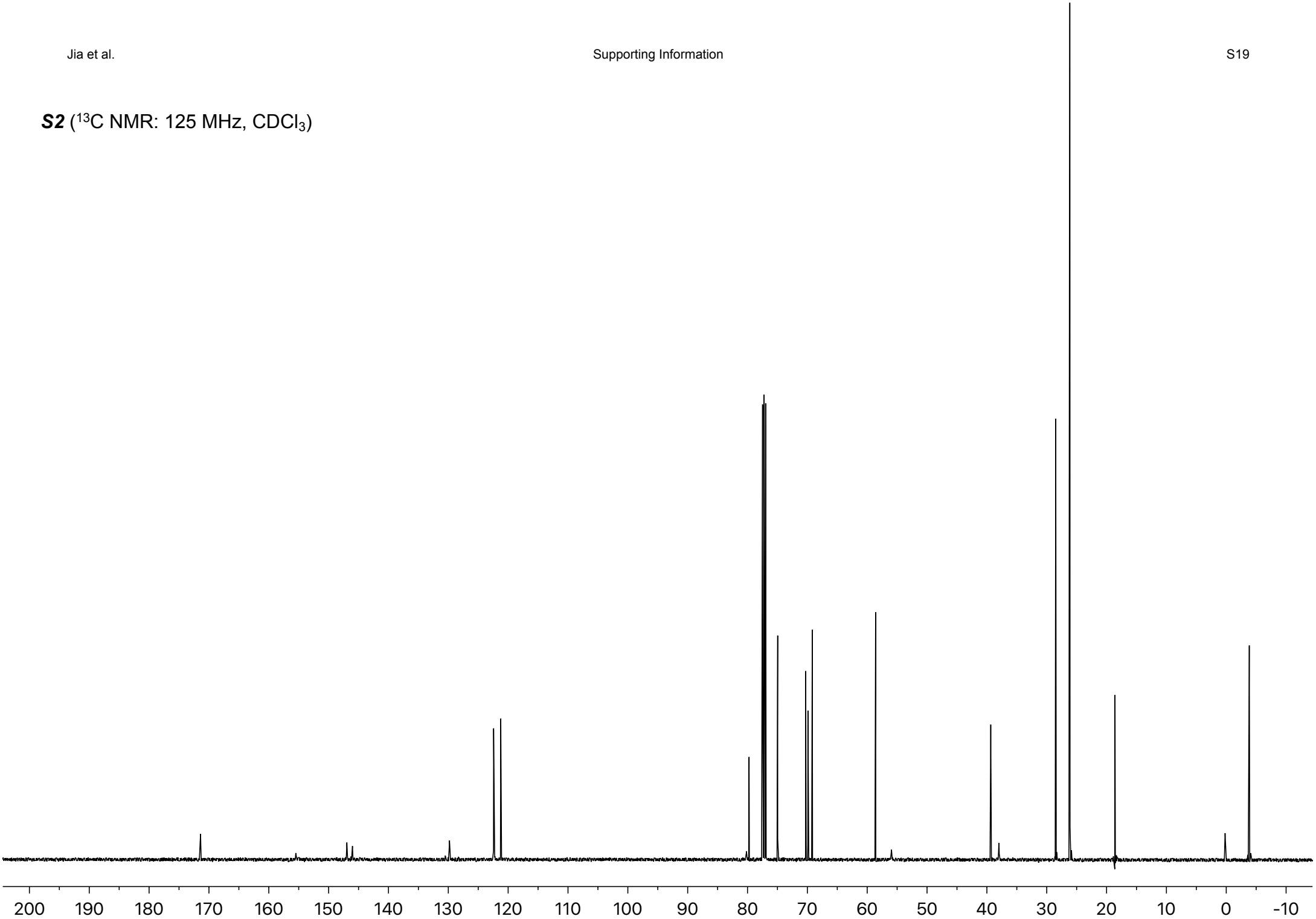
^{13}C NMR (125 MHz, d_6-DMSO) δ 162.5, 156.3, 154.6, 144.1, 136.4, 124.9, 119.3, 114.3, 110.3, 102.2, 71.1, 68.9, 63.2, and 58.1.

HRMS (ESI): Calculated for [$C_{15}H_{15}N_3O_5$ – (H^+)] 316.0939, found 316.0938.

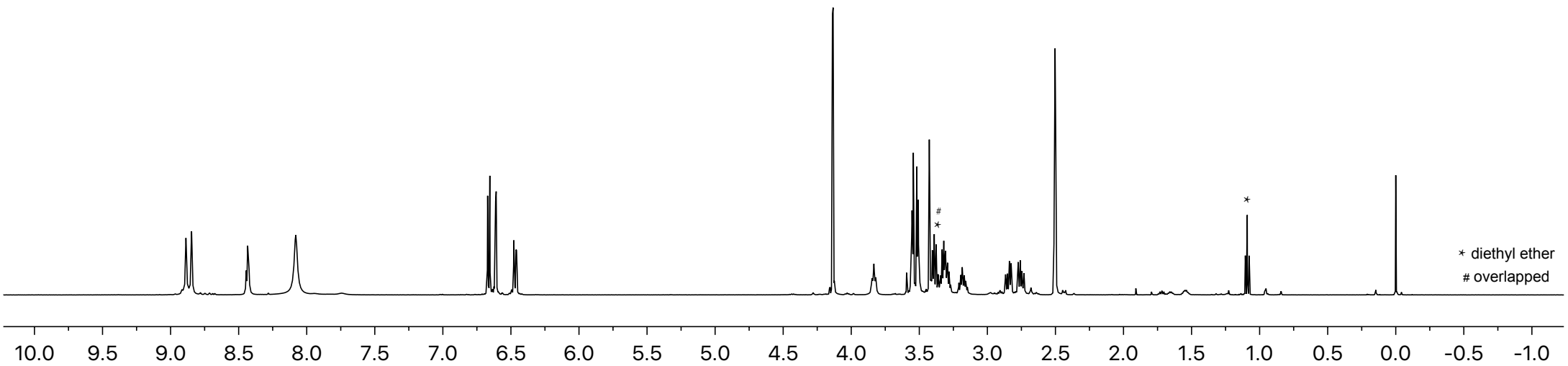
S2 (^{1}H NMR: 500 MHz, $CDCl_3$)



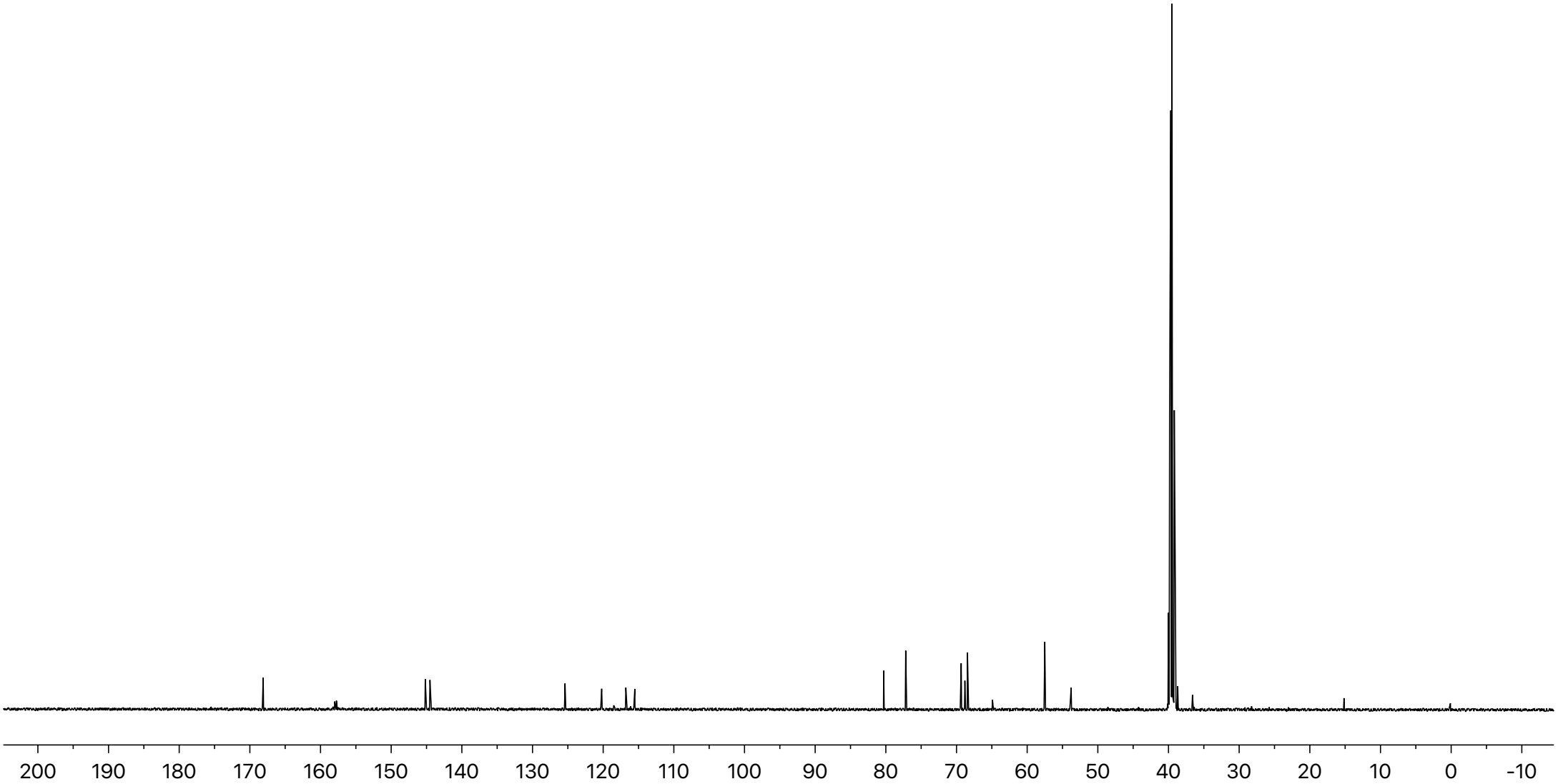
S2 (^{13}C NMR: 125 MHz, $CDCl_3$)



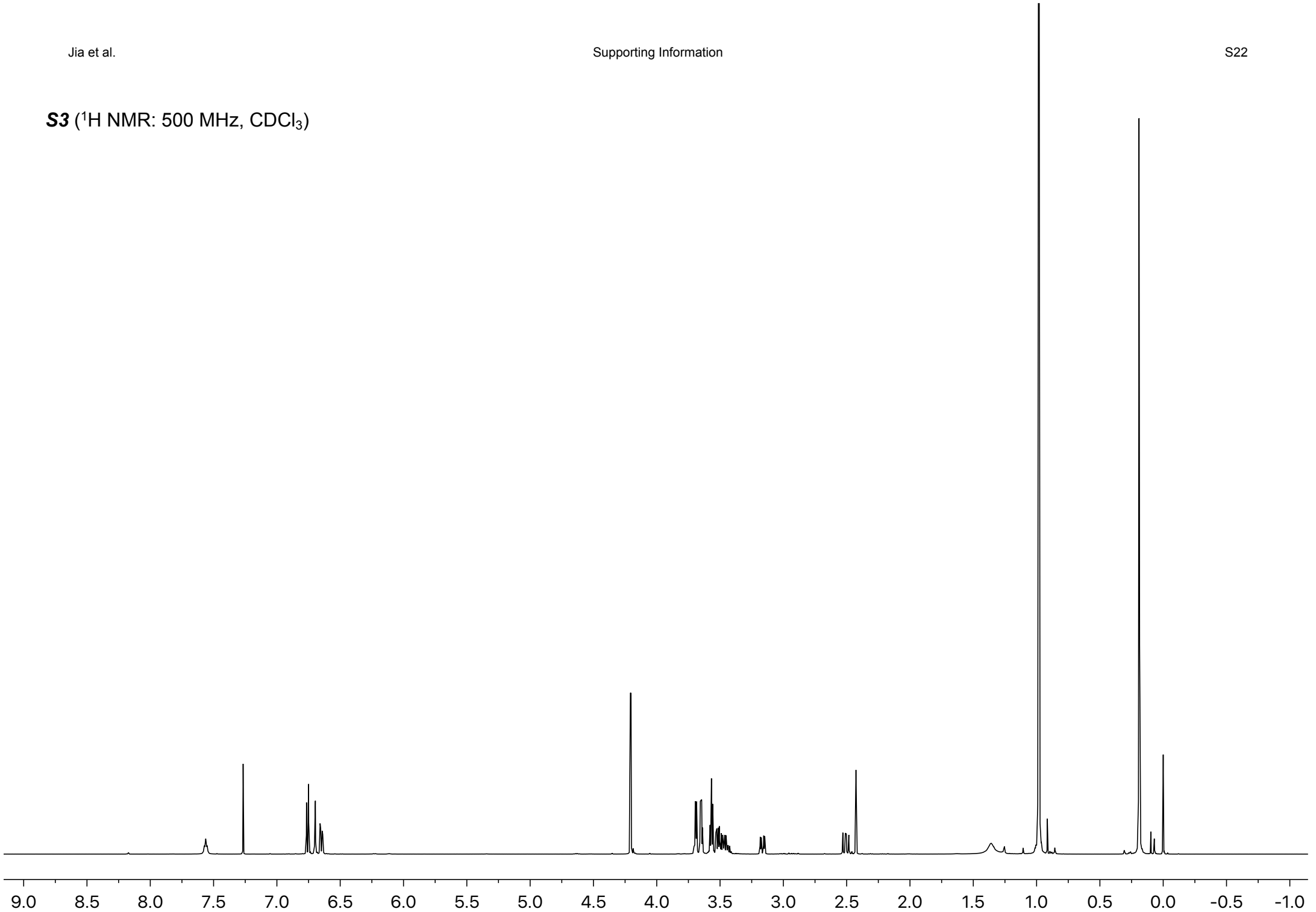
Propargyl-2EG-DOPAmide (p-DOPAmide) (^{1}H NMR: 500 MHz, d_6-DMSO)



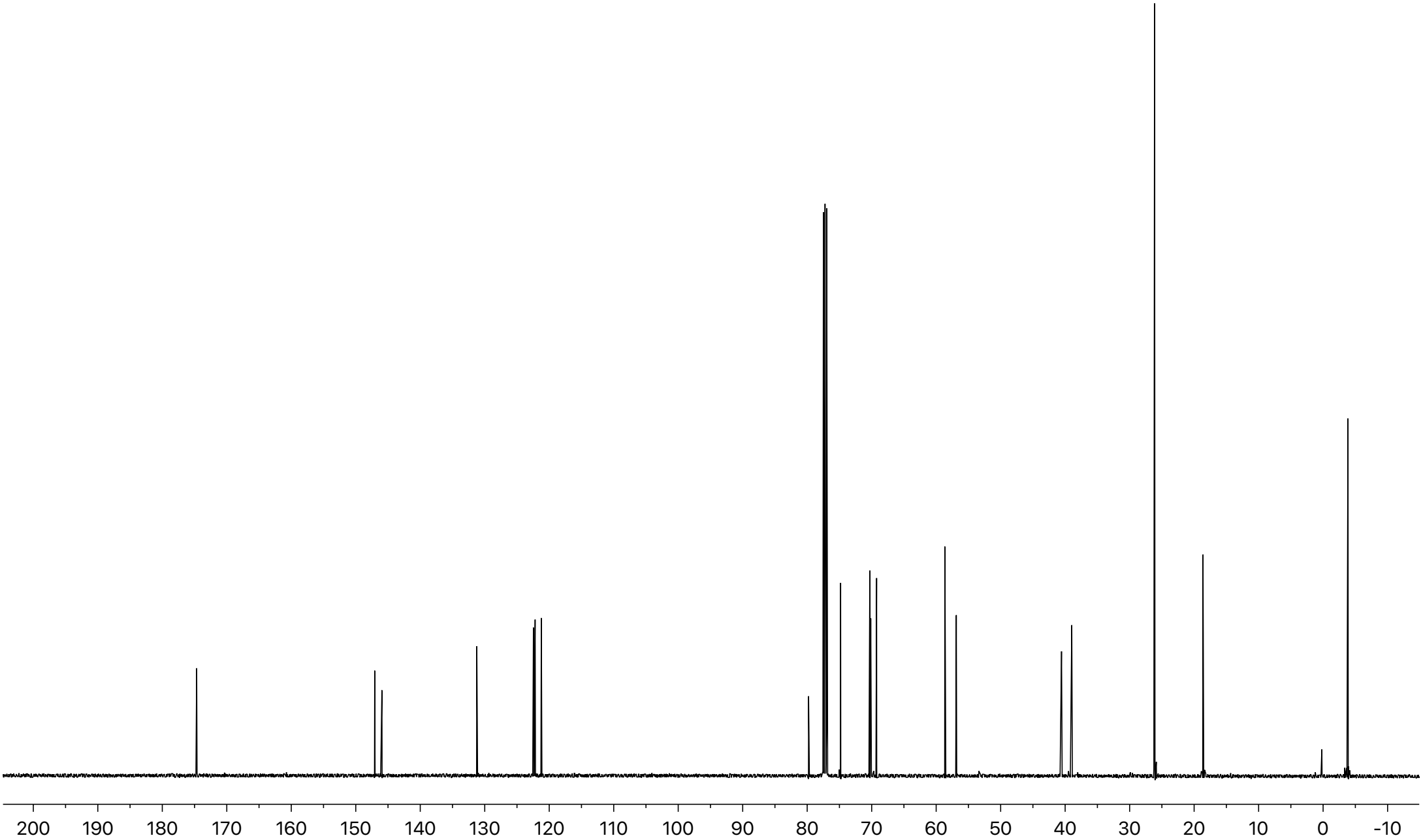
Propargyl-2EG-DOPAmide (p-DOPAmide) (^{13}C NMR: 125 MHz, d_6-DMSO)



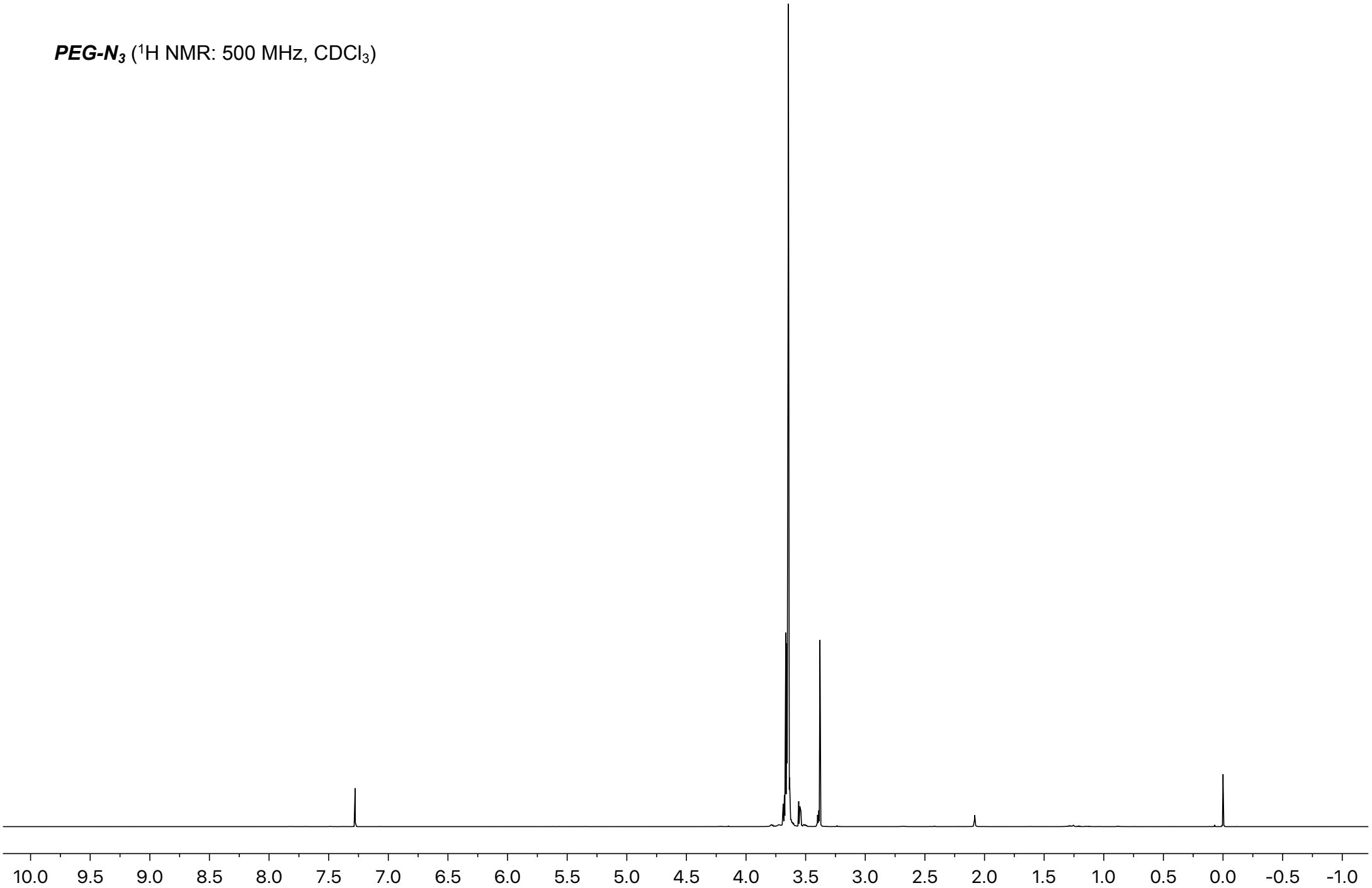
S3 (^{1}H NMR: 500 MHz, $CDCl_3$)



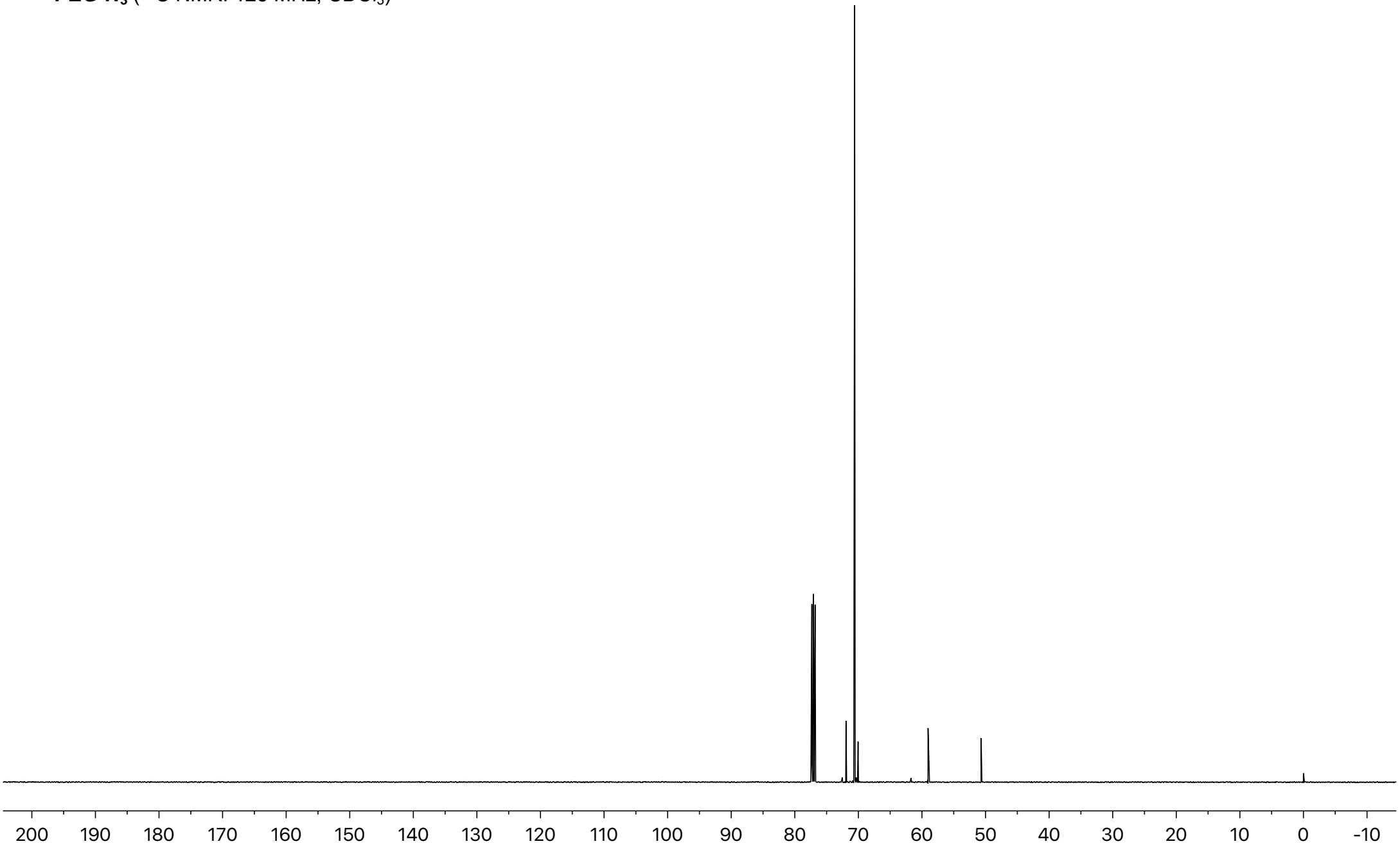
S3 (^{13}C NMR: 125 MHz, $CDCl_3$)



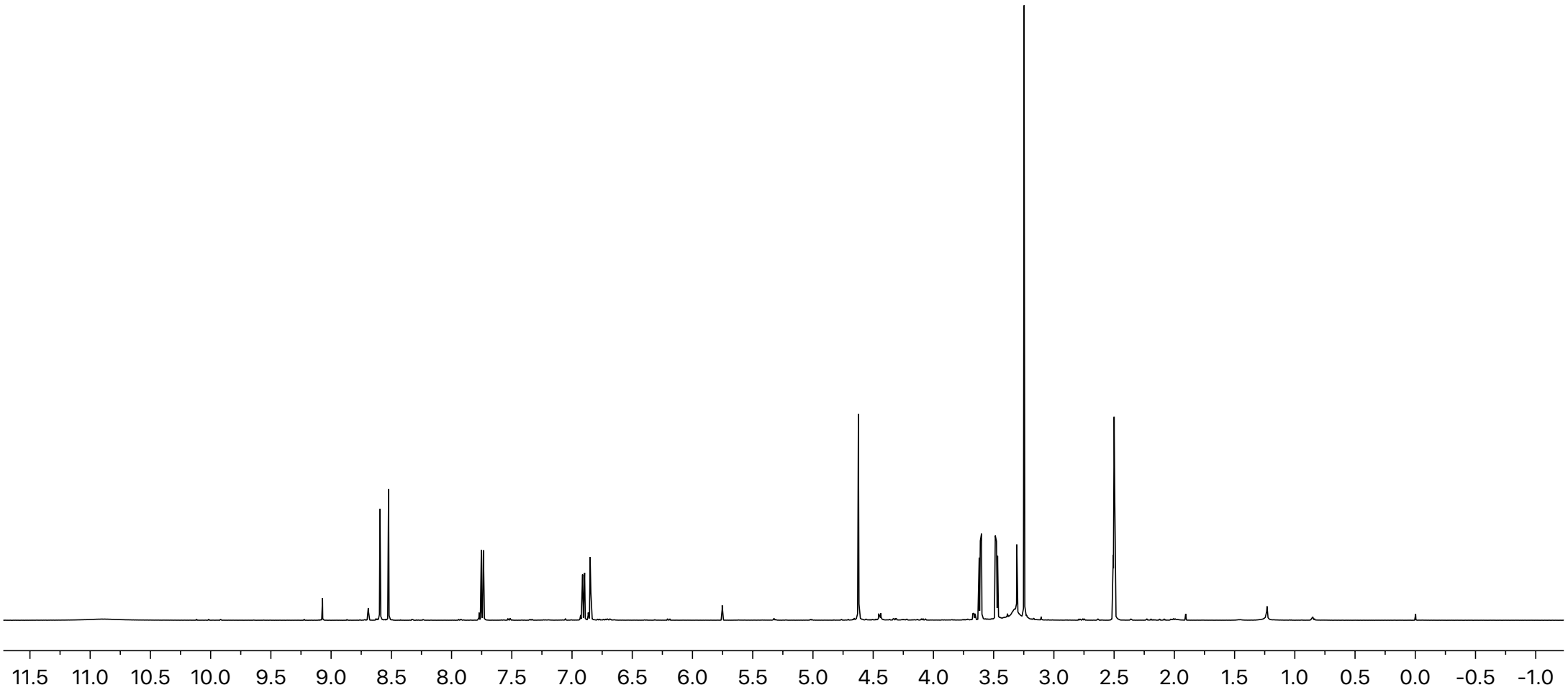
PEG-N$_3$ (^{1}H NMR: 500 MHz, $CDCl_3$)



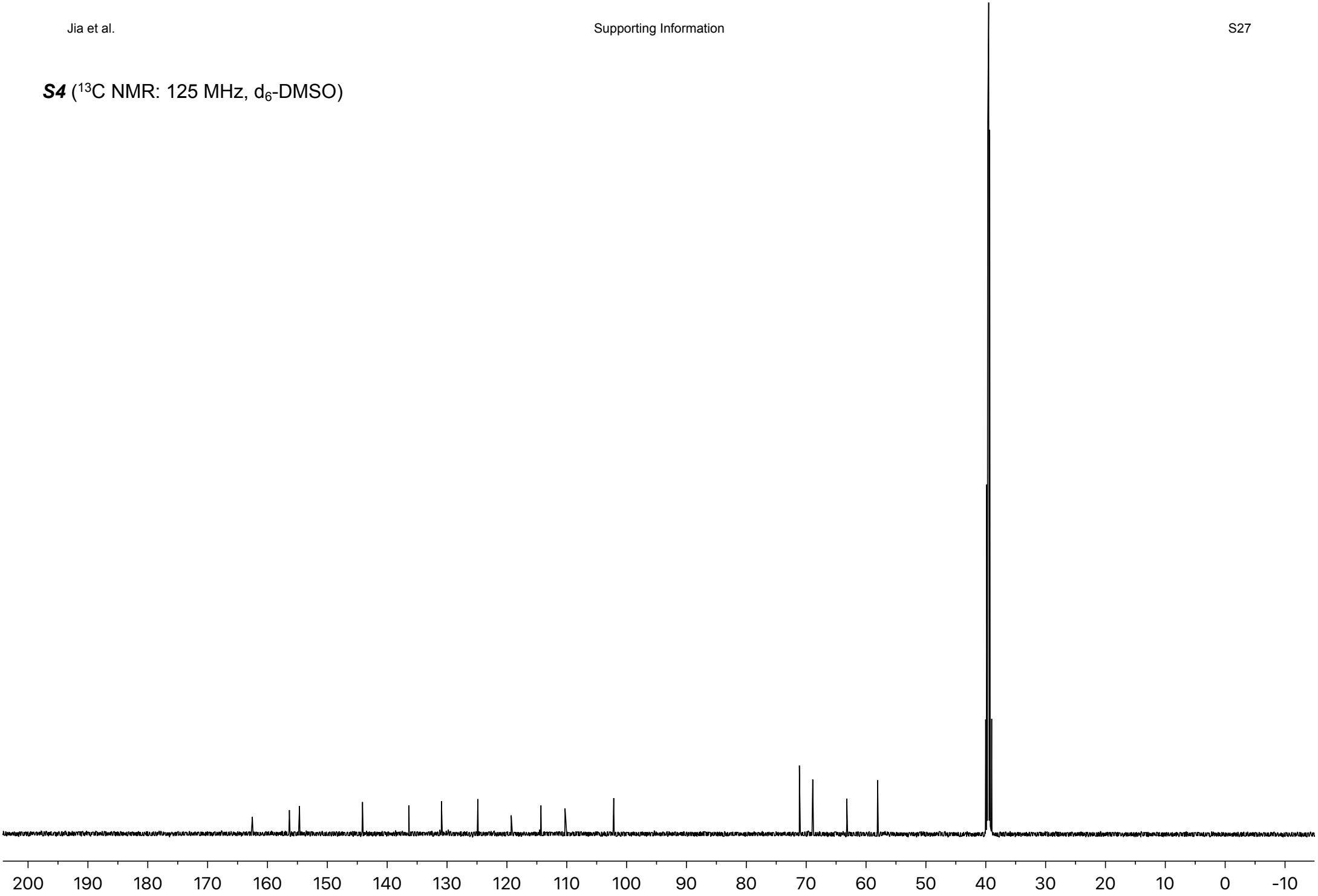
PEG-N$_3$ (^{13}C NMR: 125 MHz, $CDCl_3$)



S4 (^{1}H NMR: 500 MHz, d_6-DMSO)



S4 (^{13}C NMR: 125 MHz, d_6-DMSO)



Material-independent nature of coating

Fluorescence imaging and Contact angle analyses:

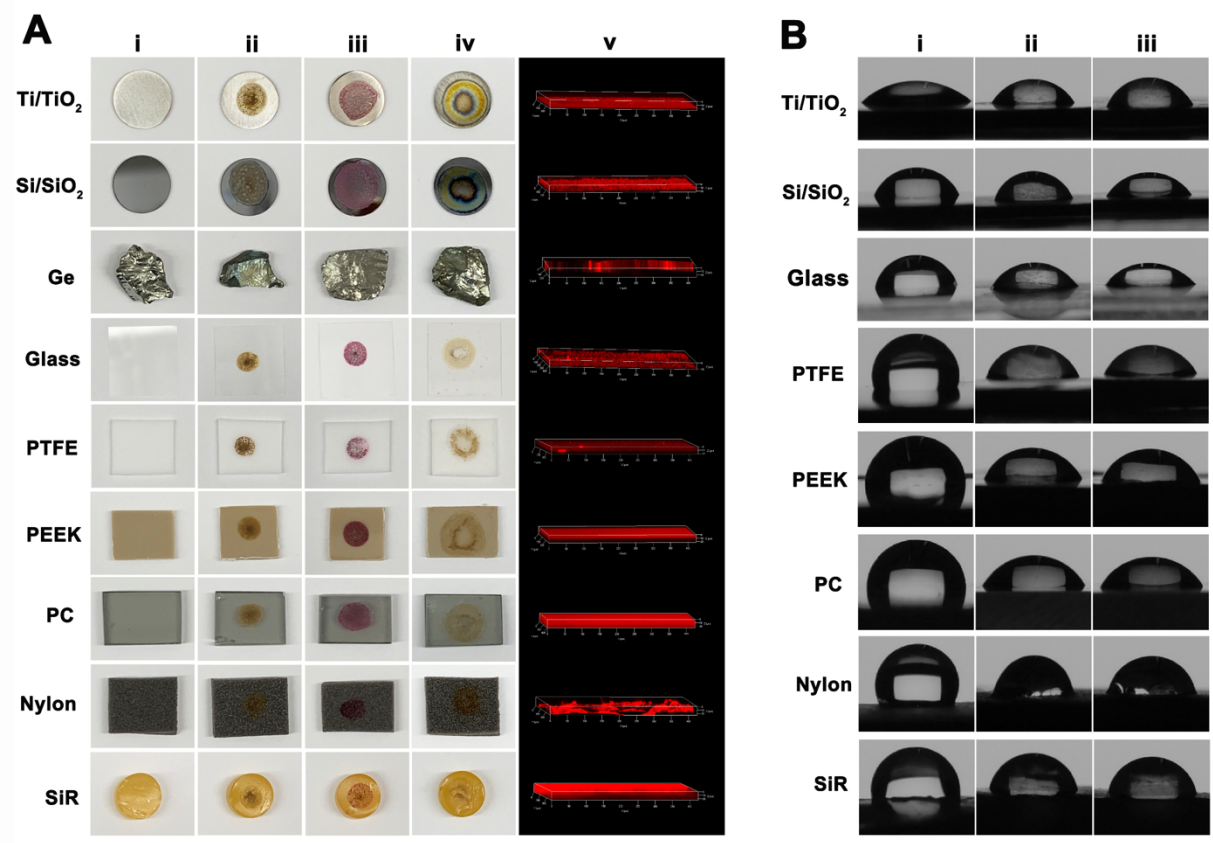
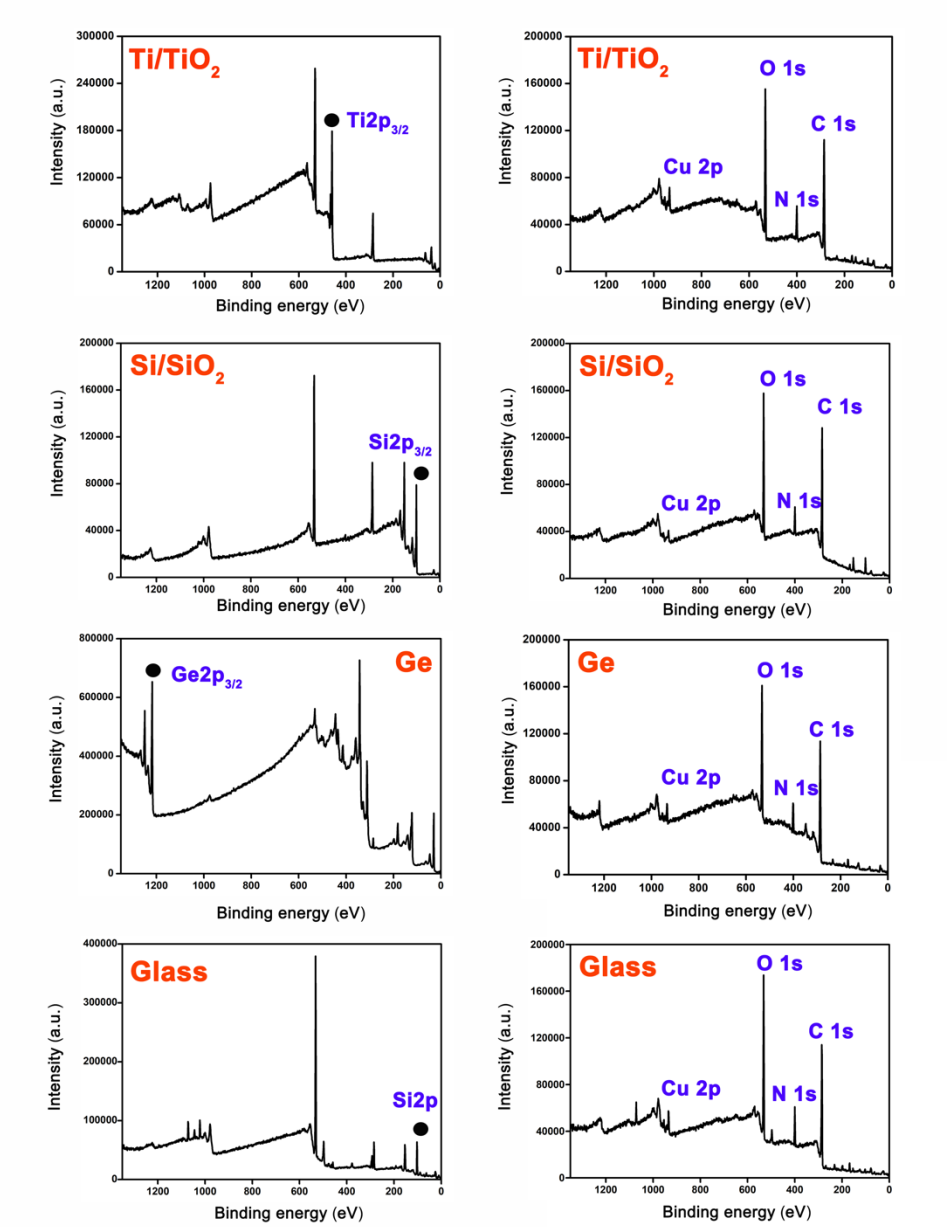


Figure S1. A. (i) Uncoated materials. Materials drop-coated using the MCM, **(ii)** without MOI-N_3, **(iii)** with TAMRA-N_3, and **(iv)** with 3-N_3-7-hydroxycoumarin. **(v)** Fluorescence images of coated materials in **(iv)**. Materials shown in **(ii)**, **(iii)**, and **(iv)** were incubated at 37 ºC for 4 h. Coated materials were washed, in some cases also sonicated, before the surface characterization. The sizes of substrates: Ti/TiO_2 and Si/SiO_2: 10 mm in diameter; glass: 18 mm × 18 mm; PTFE, PEEK, PC, and Nylon: 15–20 mm, and SiR: 5 mm in diameter. **B**. Contact angle measurements for regular surfaces: **(i)** uncoated surfaces; **(ii)** surfaces coated without a MOI-N_3; and **(iii)** surfaces coated with the MCM (MOI-N_3 = 3-N_3-7-hydroxycoumarin).

XPS analyses:

The substrate signals disappeared while C 1s, N 1s, O 1s, and Cu 2p peaks either newly emerged or altered. The coating compositions were majorly C, N, and O, suggesting the polymeric nature of the resultant coatings.



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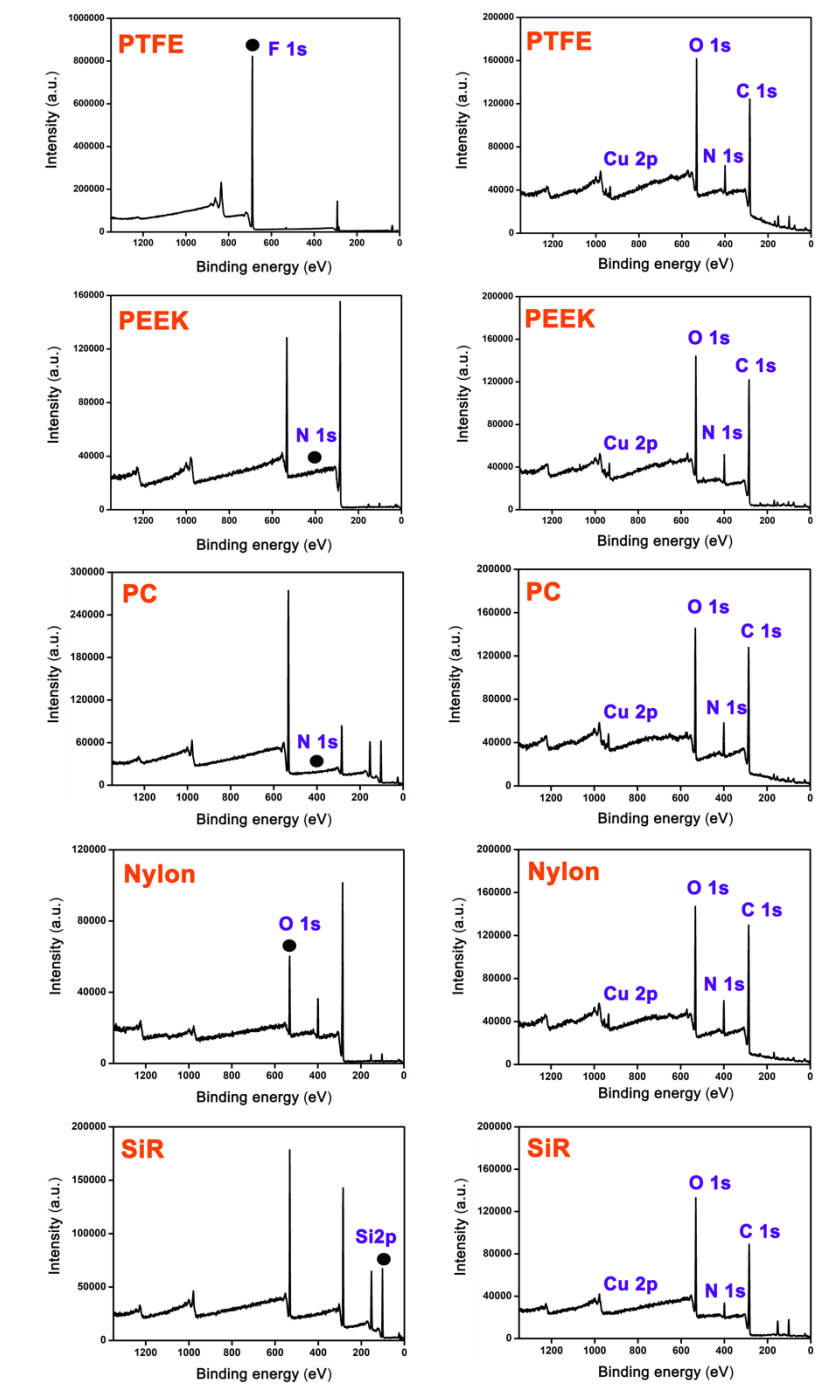


Figure S2. XPS survey spectra of various materials before (left) and after (right) 3-N_3-7-hydroxycoumarin coating. The black dots denote substrate peaks (for PEEK, PC, and Nylon, no special substrate peaks were identified; instead, N 1s or O 1s served as substrate peaks for ease of comparison with the coated materials).

Kinetics of drop coating by XPS

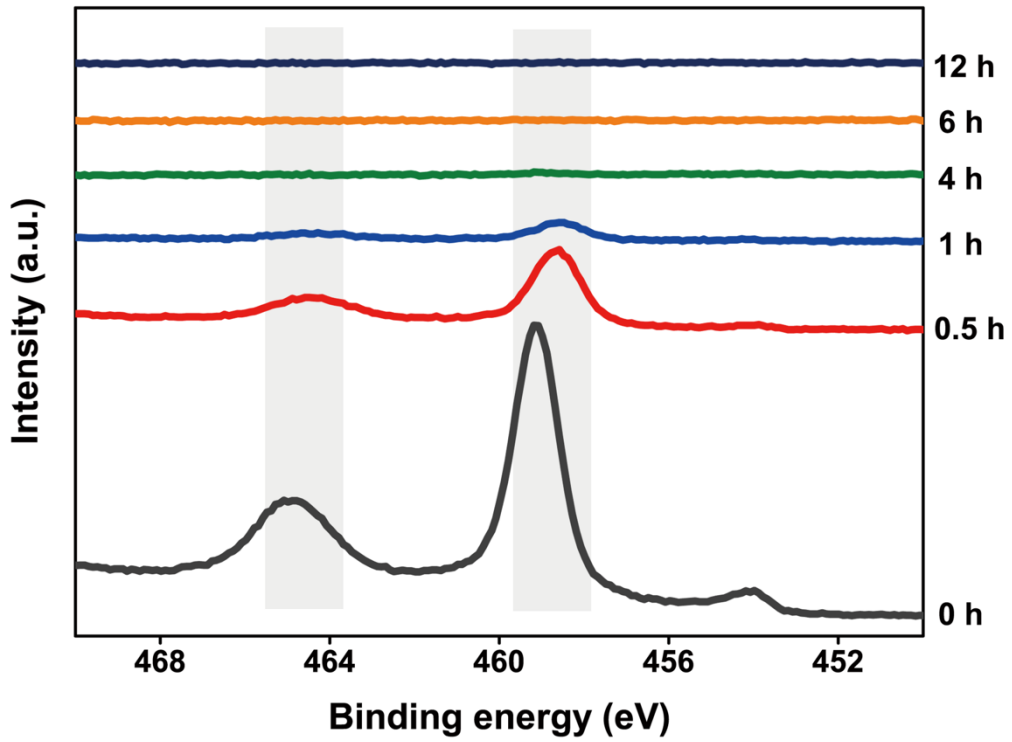


Figure S3. Change in Ti 2p XPS signals over time when the Ti/TiO_2 surface was drop coated with the MCM (MOI-N_3 = 3-N_3-7-hydroxycoumarin).

The original substrate signals (468-456 eV range) decreased significantly within the first 30 min, suggesting the coating process was successful and the coating thickness increased rapidly.

Effects of p-DOPAmide concentration and reaction time on p-DOPAmide self-assembly and grafting

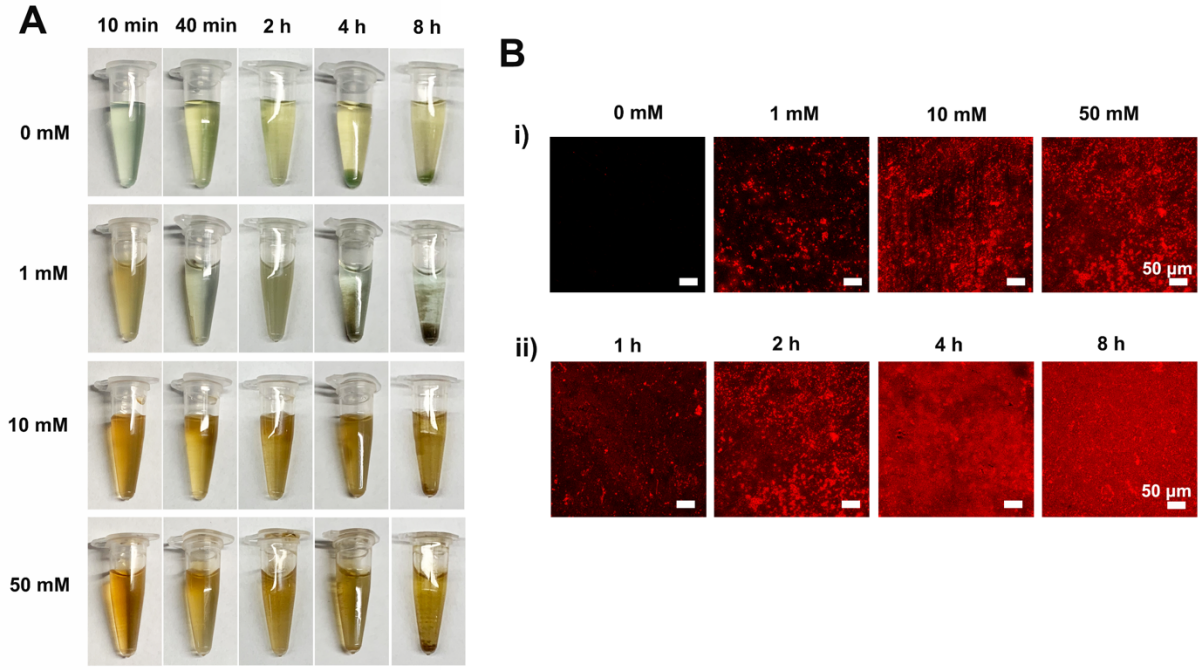


Figure S4. (A) Polymerization of p-DOPAmide in MCMs (MOI-N_3 = 3-N_3-7-hydroxycoumarin) with various p-DOPAmide concentrations and reaction time. **(B)** Effects of **(i)** p-DOPAmide concentration and **(ii)** incubation time on the coumarin density on Ti/TiO_2 surfaces. In **(i)**, the reaction time was 2 h. In **(ii)**, the sample with 50 mM initial p-DOPAmide concentration was imaged at different time points.

In **(A)**, the characteristic brown color for p-DOPAmide-containing mixtures darkened with increasing p-DOPAmide concentration and time. In **(B)**, compared to the p-DOPAmide-free samples with random weak florescence spots, p-DOPAmide groups had much greater fluorescence on the surface, suggesting successful surface functionalization. The density of the clicked coumarin increased over time.

Free 3-N_3-coumarins typically exhibit a low fluorescence emission intensity at 405 nm wavelength.[6] However, once they undergo click reaction with alkynes, the resulting triazole adducts display a substantial increase in their fluorescence quantum yield.6

Cu(II)-Lig assisted p-DOPAmide oxidative polymerization in solution

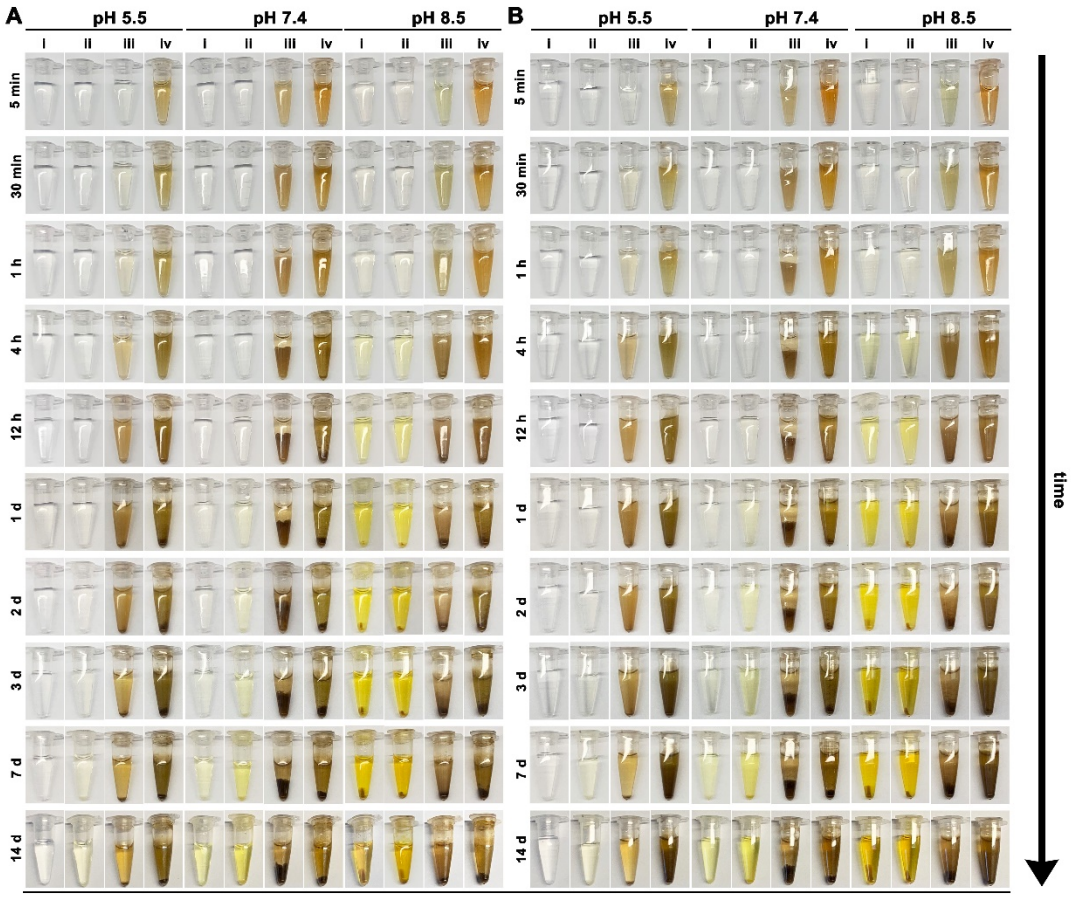


Figure S5. Oxidative polymerization of p-DOPAmide **(i)** in the absence of any additive, **(ii)** in the presence of THPTA, **(iii)** in the presence of $CuSO_4$, and **(iv)** in the presence of both $CuSO_4$ and THPTA. Buffers used: MES for pH 5.5, PBS for pH 7.4, and Tris for pH 8.5. **(B)** Solutions of p-DOPAmide and additives as well as buffers were bubbled with N_2 for 15 min prior to mixing.

Investigating the effect of reagents in drop coating by Raman spectroscopy

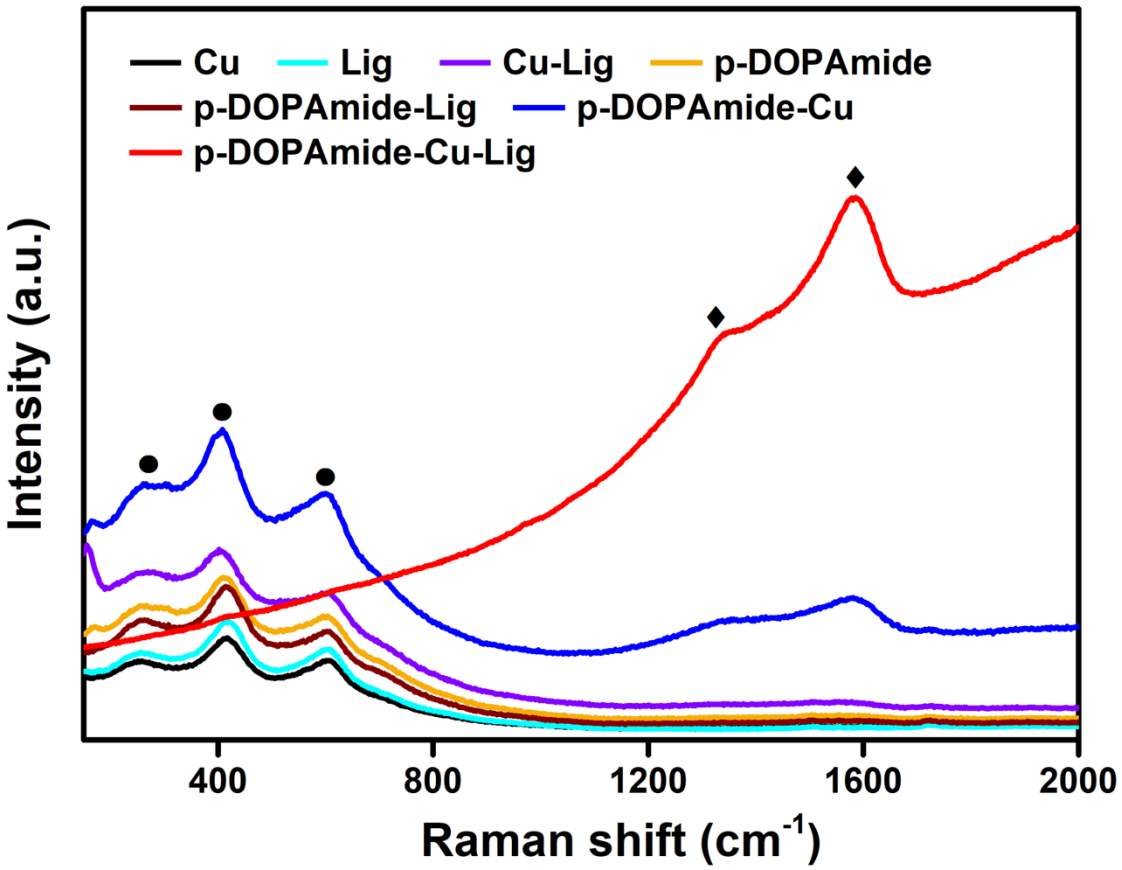
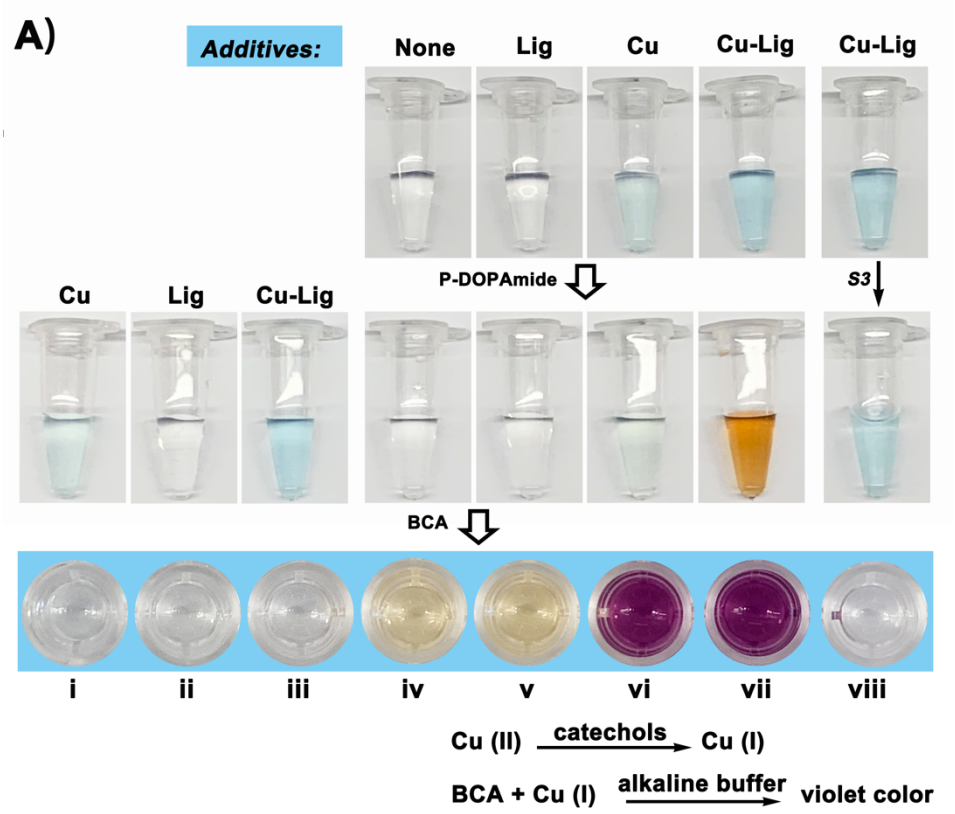


Figure S6. Raman spectra of Ti/TiO_2 treated with coating mixtures with different combinations of Cu (5 mM), Lig (10 mM), and p-DOPAmide (10 mM). Cu = $CuSO_4$, Lig = THPTA. Black dots indicate substrate signals belonging to TiO_2,[7] while the prismatic indicates bands from p-DOPAmide and its oxidative products: catecholic C−OH (1335 cm^{-1}) and aromatic C−C stretching (1580 cm^{-1}). [8] To fit the spectral line into the graph, the intensity of p-DOPAmide-Cu-Lig spectrum (red) has been kept at 20% (for a similar processing, see Holten et al.[9]). In the Raman spectra, substrate bands were still visible for Cu, Lig, Cu-Lig, p-DOPAmide, and p-DOPAmide-Lig groups. In contrast, those bands disappeared while new catechol bands emerged for p-DOPAmide-Cu-Lig. New bands were also observed for p-DOPAmide-Cu samples at similar positions, but they showed a much weaker intensity. These indicated that neither Cu, Lig, or their combination, nor p-DOPAmide or the p-DOPAmide-Lig combination could result in a detectable coating. In summary, the efficient coating hinged on the co-existence of p-DOPAmide, Cu, and Lig.

In situ Cu(I) generation observed by MicroBCA assay



B)

Sample #	Final mixture
i	PBS + Cu + BCA
ii	PBS + Lig + BCA
iii	PBS + Cu + Lig + BCA
iv	PBS + p-DOPAmide + BCA
v	PBS + Lig + p-DOPAmide + BCA
vi	PBS + Cu + p-DOPAmide + BCA
vii	PBS + Cu + Lig + p-DOPAmide + BCA
viii	PBS + Cu + Lig + *S3* + BCA

Figure S7. A. Design and observations of the MicroBCA assay using different media. Cu = $CuSO_4$, Lig = THPTA. **B.** Summary of compositions of the final mixtures.

Only (vi) and (vii) generated a violet color, indicative of Cu(I)-BCA complex. Protection of the hydroxyl groups of p-DOPAmide (***S3***) inhibited the colorization. Together, these results showed that Cu(I) was produced in the drop coating mixtures and this process is dependent on the redox reaction between p-DOPAmide and Cu(II) species.

Investigating the effect of reagents in drop coating by CLSM

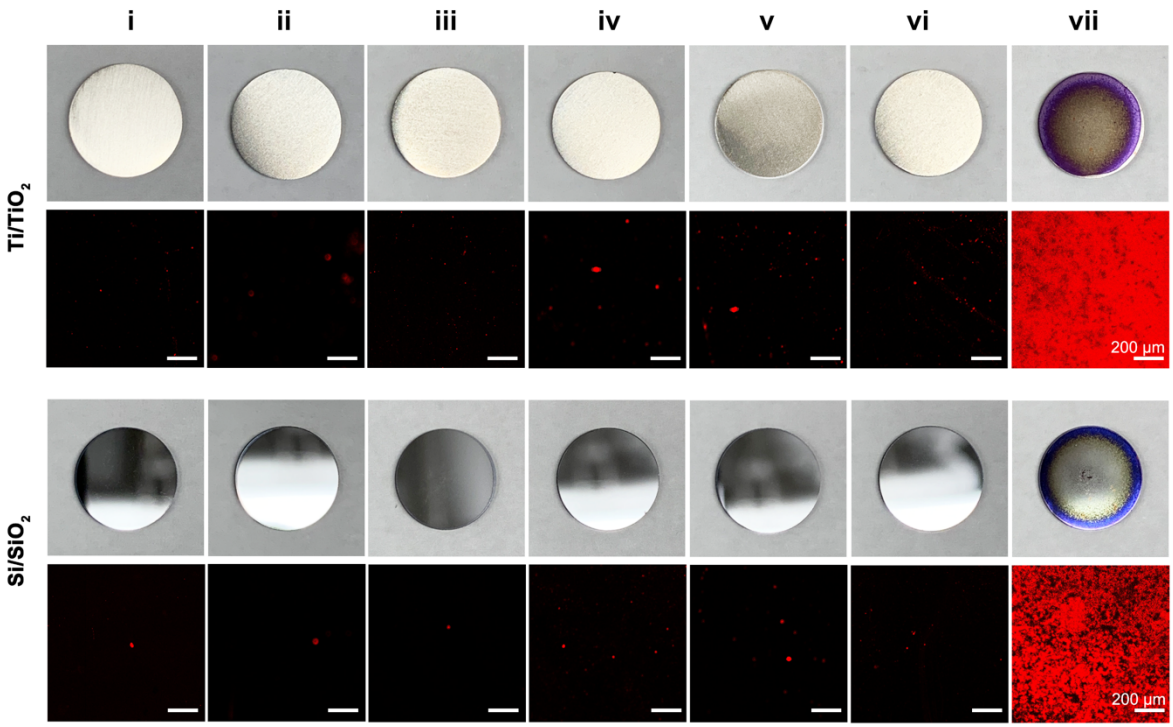


Figure S8. Ambient light images (upper) and fluorescence images (lower; inset with tile scans) of Ti/TiO_2 and Si/SiO_2 substrates treated with **(i)** $CuSO_4$ and TAMRA-N_3, **(ii)** THPTA and TAMRA-N_3, **(iii)** $CuSO_4$, THPTA, and TAMRA-N_3, **(iv)** p-DOPAmide and TAMRA-N_3, **(v)** p-DOPAmide, THPTA, and TAMRA-N_3, **(vi)** p-DOPAmide, $CuSO_4$, and TAMRA-N_3, and **(vii)** p-DOPAmide, $CuSO_4$, THPTA, and TAMRA-N_3.

Based on the ambient light and fluorescence images, only the mixture that containing p-DOPAmide, $CuSO_4$, THPTA, and TAMRA-N_3 all together **(vii)** underwent an efficient click reaction.

Material-independent formation of films

Polymeric films were formed in a generally material-independent manner. Films were alike for coating mixtures with and without 3-N_3-7-hydroxycoumarin, suggesting that film formation was a result of copper, ligand, and p-DOPAmide and/or their interactions.

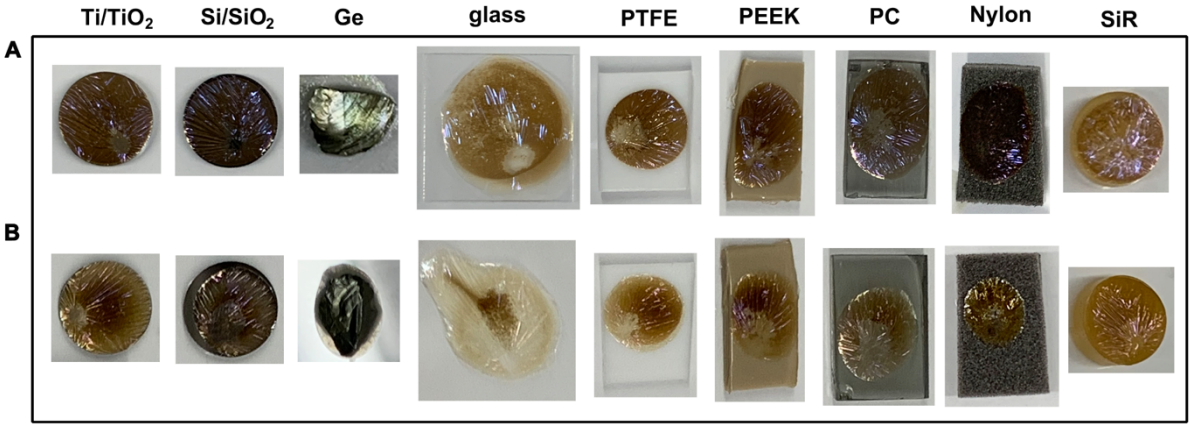


Figure S9. Film formation on different materials during drop-coating using coating mixtures **(A)** without or **(B)** with 3-N_3-7-hydroxycoumarin.

Interfacial behaviors of mixtures with or without p-DOPAmide

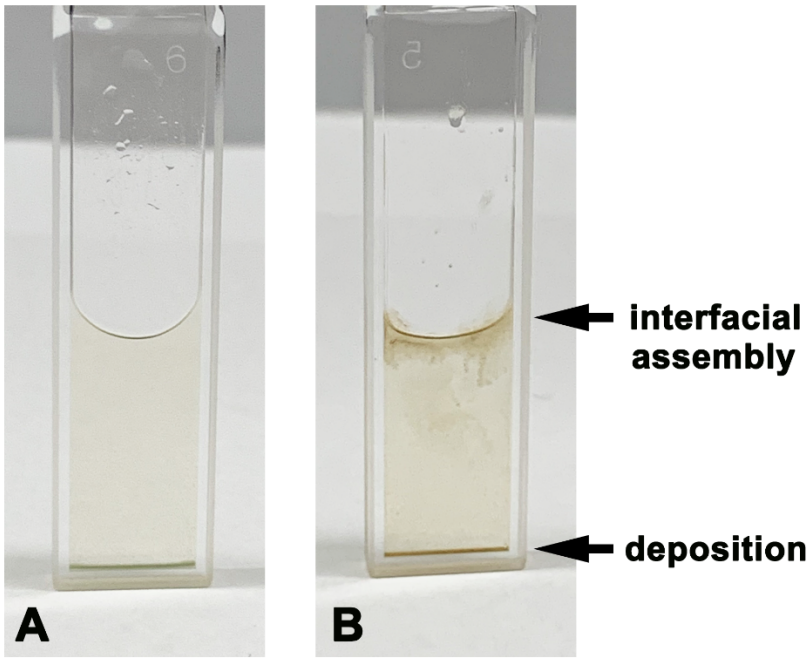


Figure S10. Interfacial oxidative p-DOPAmide self-assembly in a quartz cuvette. Interfacial behaviors of 3-N_3-7-hydroxycoumarin-containing mixtures **(A)** without and **(B)** with p-DOPAmide (10 mM, 4 h).

Over time, deposition occurred at the bottom of both cuvettes, with a green color in **(A)** and a brown color in **(B)**. We associate this green color to copper-containing insoluble species. A darker brown region appeared at the liquid-air interface, likely due to an increased rate of catechol oxidization as p-DOPAmide was directly exposed to air.

Film formation on Ti/TiO_2 via drop coating

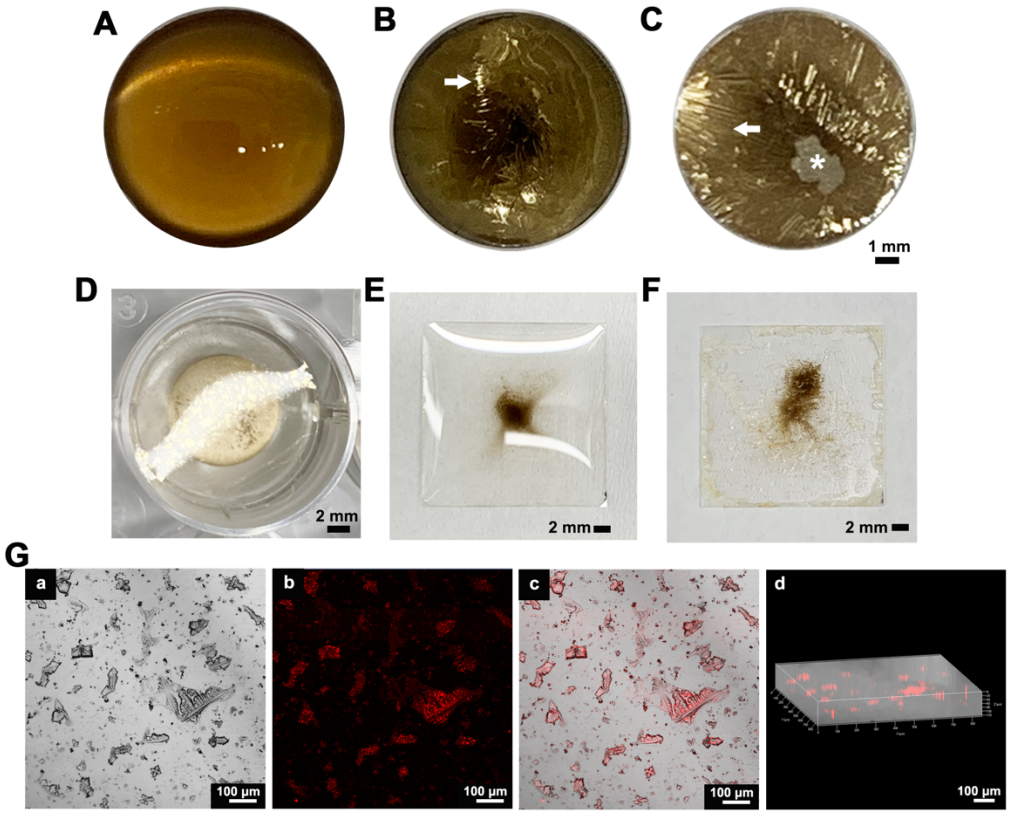


Figure S11. (A) Ti/TiO_2 substrate was drop coated with 100-µL of the MCM (MOI-N_3 = 3-N_3-7-hydroxycoumarin). **(B)** The droplet became covered with a light-reflecting polymeric thin film (arrow) after incubation at 37 °C for 1 h. The film spontaneously formed along the meniscus (contact angles) of the droplet. **(C)** The film showed a wrinkled morphology with a light-reflecting property (arrow) after aspirating the liquid with a pipette (disrupted site is indicated by the asterisk (*)). **(D)** Small debris from the film floated on water that was used to rinse the Ti/TiO_2 substrate. **(E)** The washing solution dropped on a glass coverslip, which contained the film debris and some brown-colored deposits. **(F)** Air-dried coverslip. **(G)** Examination of the film debris by CLSM: **(a)** brightfield image, **(b)** fluorescence image, **(c)** merged, **(d)** z-stack image. The overlap of the film debris and red fluorescence suggested that the film contained clicked coumarin.

Characterization of film debris on Ti/TiO_2 by AFM

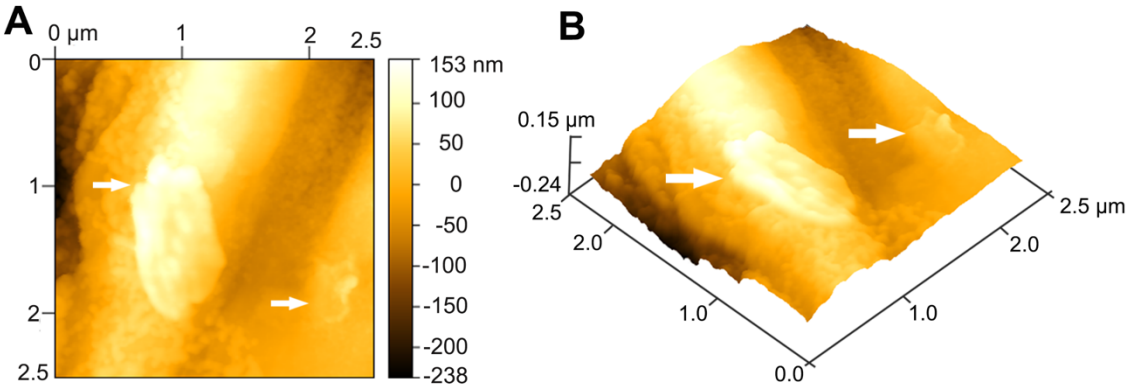


Figure S12. Examination of surface-adhered film debris (white arrows) on drop coated Ti/TiO_2 substrate by AFM. **(B)** is 3D visualization of **(A)**.

Characterization of film debris on Si/SiO_2 by TEM

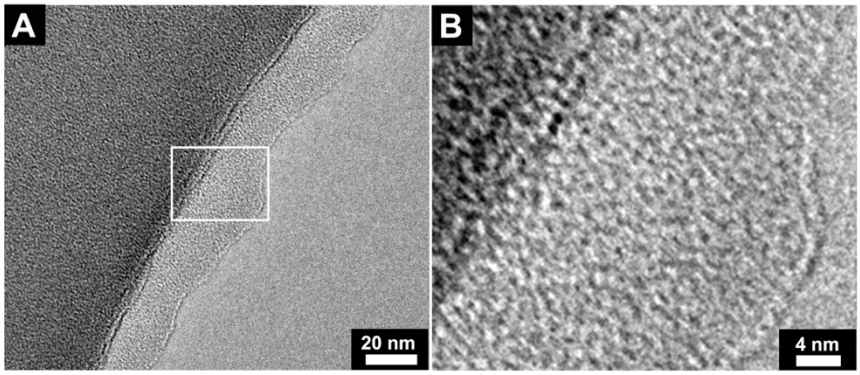


Figure S13. TEM images of a film formed during drop coating of 3-N_3-7-hydroxycoumarin onto a Si/SiO_2 substrate. **(B)** Magnified image of the highlighted region in **(A)**.

Characterization of film debris on Si/SiO_2 by Raman spectroscopy

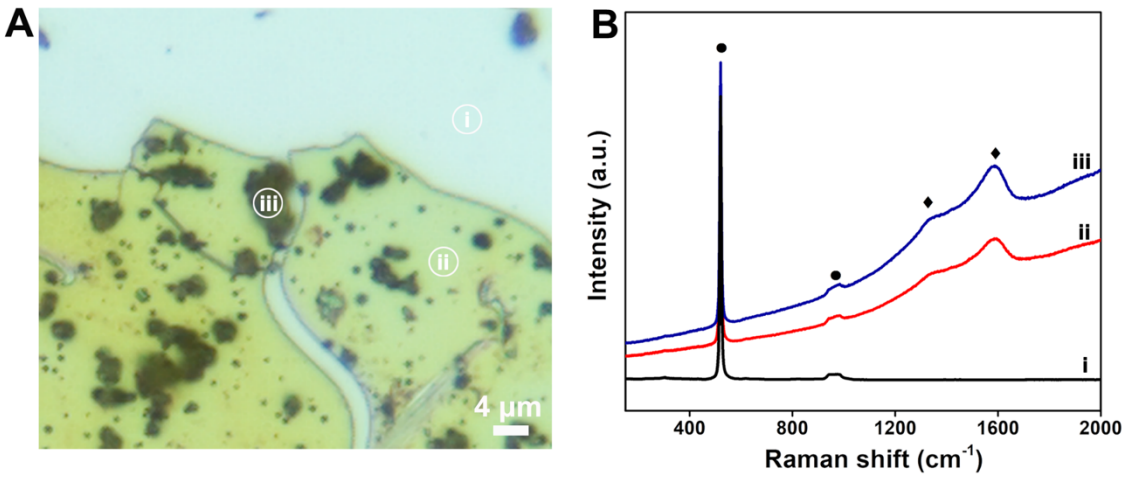


Figure S14. Examination of film debris deposited on a Si/SiO_2 substrate by Raman spectroscopy. **(A)** Optical image. **(B)** Raman spectra of spots (i)-(iii) in **(A)**. Black dots indicate substrate signals belonging to Si/SiO_2, while the prismatic indicates bands from p-DOPAmide and its oxidative products: catecholic C−OH (1335 cm^{-1}) and aromatic C−C stretching (1580 cm^{-1}). Compared to the supporting substrate (i), both the film (ii) and film-adhered particles (iii) showed catecholic characteristics.

Effect of interfacial film formation on solvent evaporation

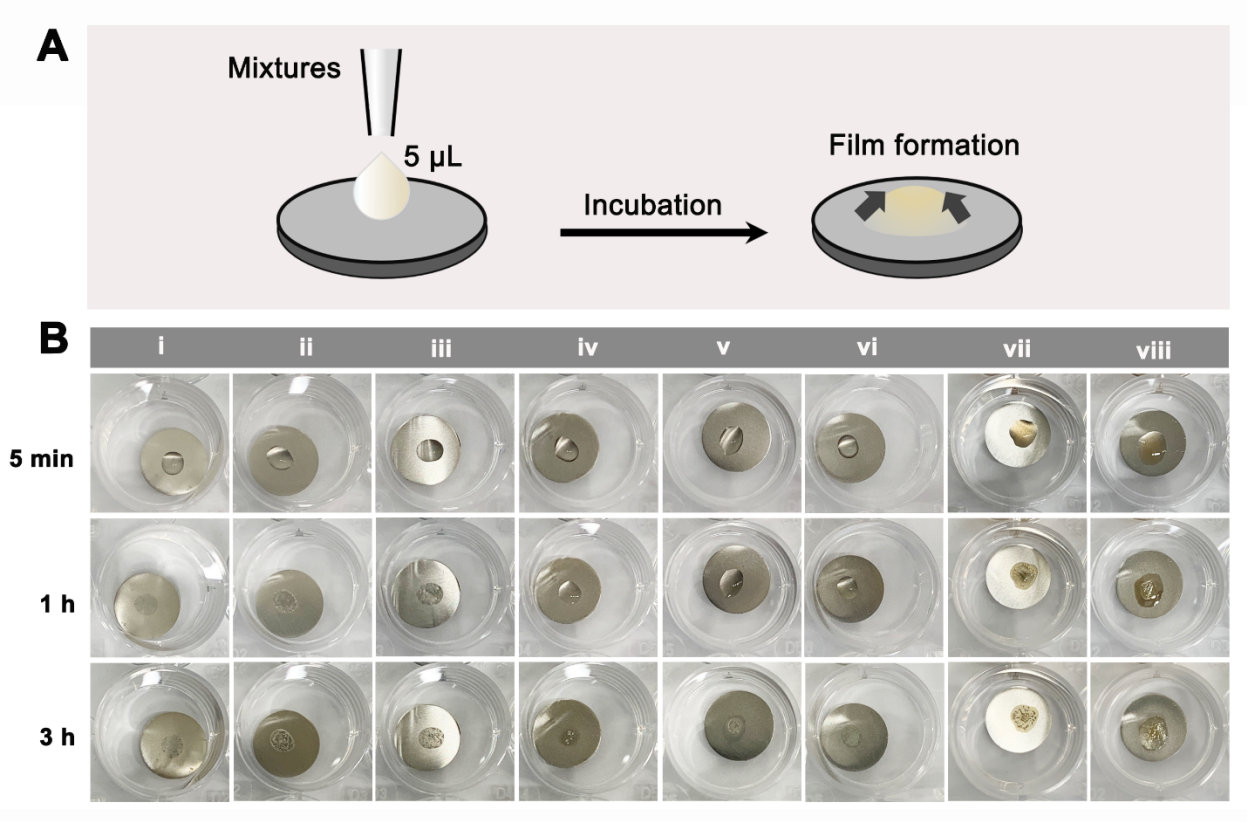


Figure S15. Investigating interfacial film formation on substrate with regard to coating mixture compositions (i-viii) and incubation time: **(A)** experimental design; **(B)** observations (bird view). i: Cu, ii: Lig, iii: Cu-Lig, iv: p-DOPAmide, v: p-DOPAmide-Lig, vi: p-DOPAmide-Cu, vii: p-DOPAmide-Cu-Lig, viii- p-DOPAmide-Cu-Lig-Coumarin, where Cu = $CuSO_4$, Lig = THPTA, Coumarin = 3-N_3-7-hydroxycoumarin. Arrows in the schematics indicate the liquid/air interface.

Single-step multiplexed functionalization

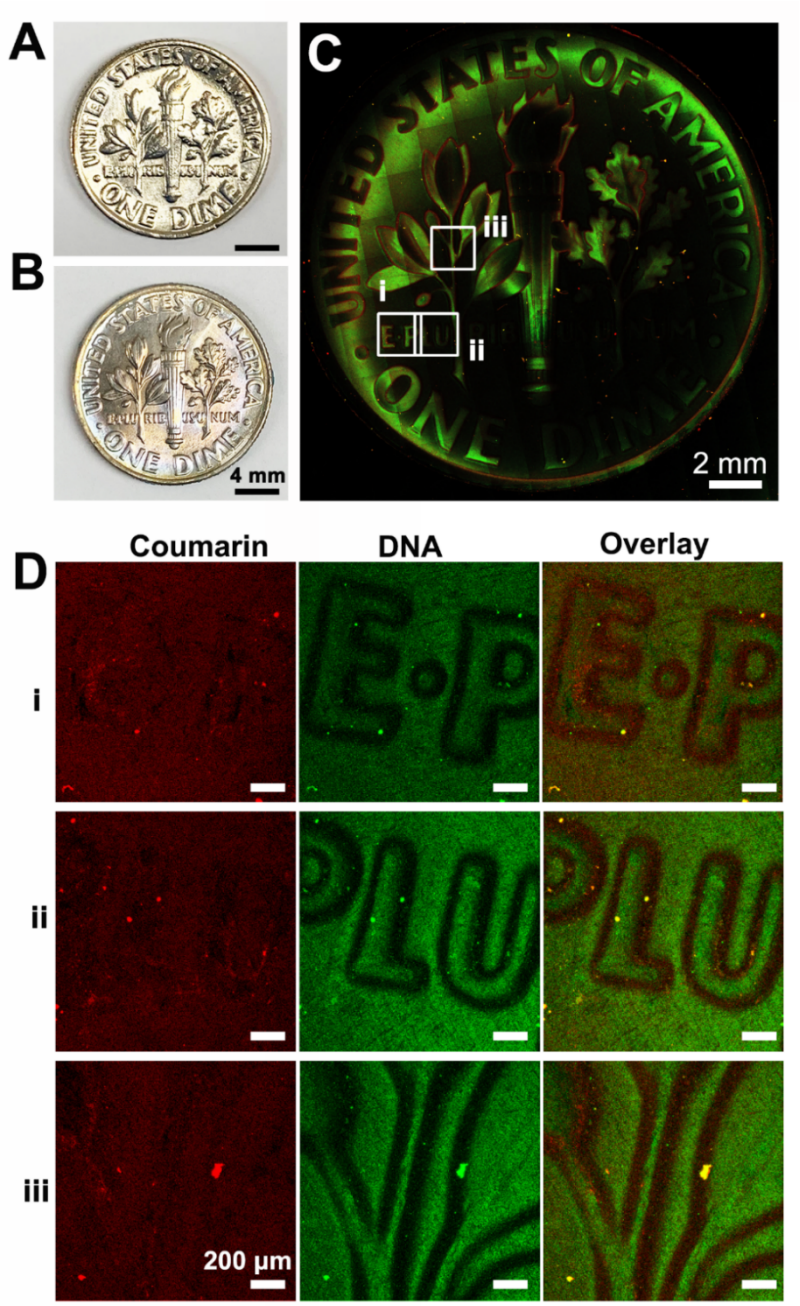


Figure S16. Ambient light appearance of a dime **(A)** before and **(B)** after drop coating with the MCM containing two MOI-N_3, 3-N_3-7-hydroxycoumarin (0.5 mM) and N_3-DNA-FAM (1 µM). **(C)** Fluorescence image of the drop coated dime. **(D)** High-magnification images highlighting the selected patterns on the coin **(i)**–**(iii)**.

Material-independent and template-free patterning

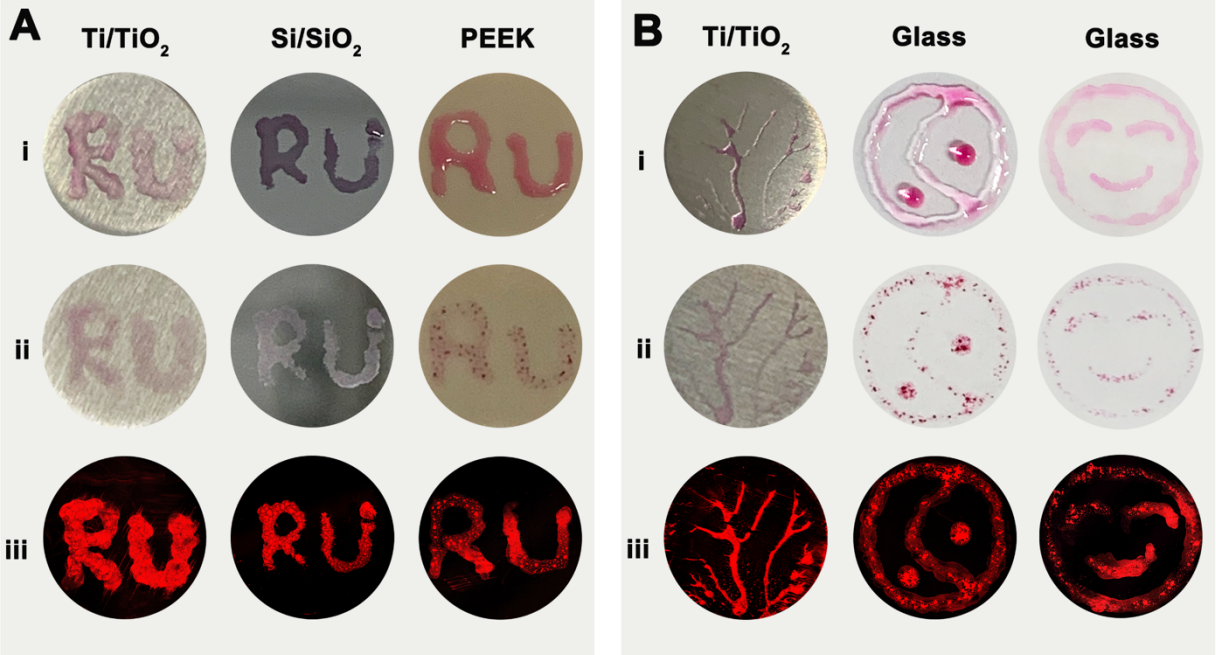


Figure S17. Template-free writing **(A)** and drawing **(B)** on material surfaces using the MCM (MOI-N_3 = TAMRA-N_3, 0.5 mM). **(i)** Ambient light images of the mixtures just added on the surfaces. **(ii)** Ambient light images and **(iii)** fluorescence images of the resulting surface patterns after 1 h of incubation and subsequent washing.

MC3T3-E1 cell adhesion on different substrate materials

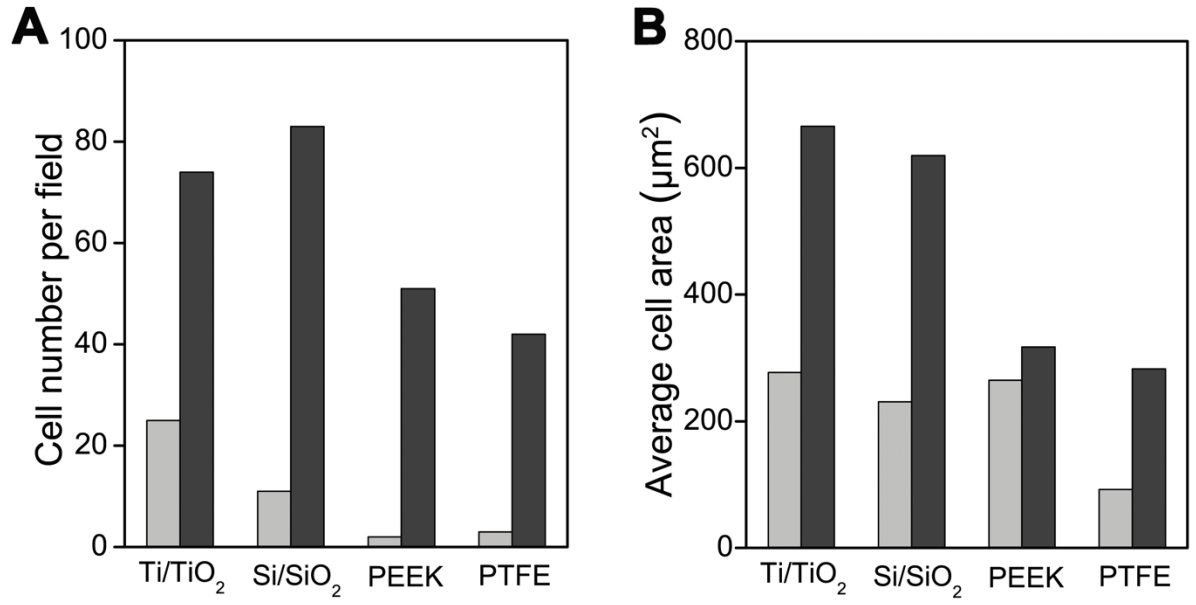


Figure S18. Quantification of **(A)** adhered cells and **(B)** average cell spreading area on various materials without (light gray bar) and with (dark gray bar) c(RGDfK) functionalization.

Cytocompatibility assay

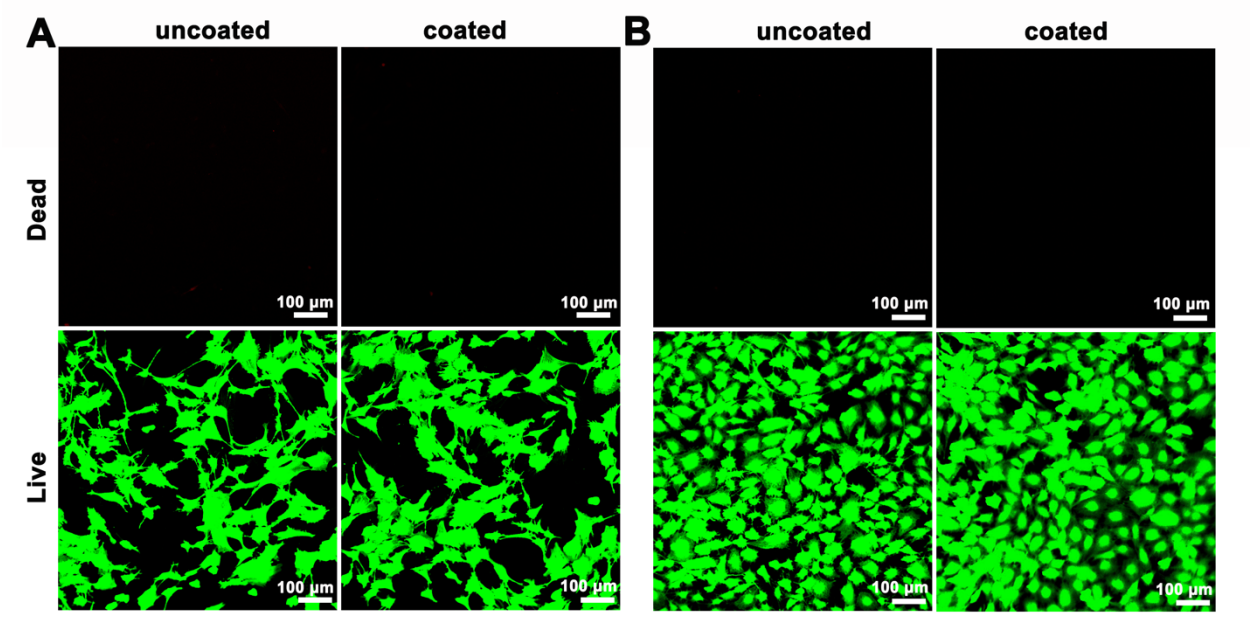


Figure S19. Cytocompatibility of MC3T3-E1 cells that were treated with extracts of uncoated and c(RGDfK)-coated Ti/TiO_2 at **(A)** 1 d and **(B)** 3 d. The cells were stained with both calcein AM and PI. Calcein AM stains live cells to give green color, while PI stains dead cells to give red color.

Cell adhesion on 3D scaffolds

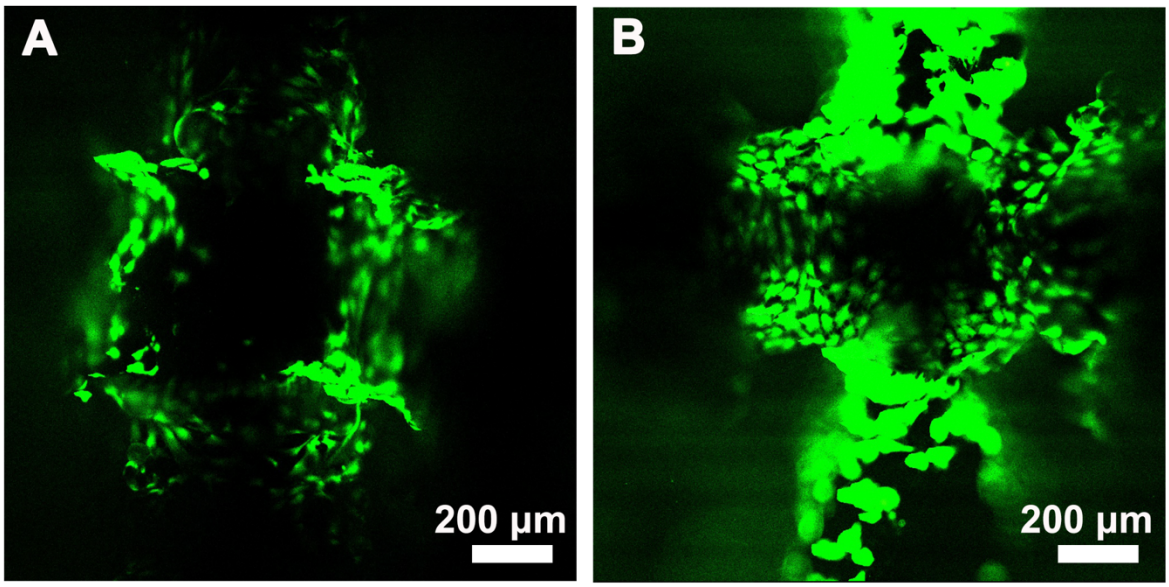


Figure S20. Adhesion of MC3T3-E1 cells on the struts of porous, 3-dimensional Ti alloy scaffolds **(A)** without and **(B)** with c(RGDfK) coating for tissue engineering. The cells were stained with both calcein AM and PI. Calcein AM stains live cells to give green color, while PI stains dead cells to give red color.

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