[Supporting information to accompany Chem. Sci. manuscript SC-EDG-08-2013-052180]

Facile one-step solid-phase synthesis of multitopic organic-DNA hybrids via "Click" chemistry

Ryan V. Thaner,[†,‡] Ibrahim Eryazici,[*,†,‡] Omar K. Farha,[†] Chad A. Mirkin,[†] SonBinh T. Nguyen[*,†]

[§] These authors contributed equally

[†]*Department of Chemistry, Northwestern University, 2145 Sheridan Road, Evanston, Illinois 60208-3113*

Table of Contents

Materials and instrumentation. Unless otherwise stated, all reagents and reagent-grade solvents were purchased from Acros Organics (part of Thermo Fisher Scientific, New Jersey, NJ), Strem Chemicals (Newburyport, MA), Aldrich Chemical Company (Milwaukee, WI), or Glen Research (Sterling, VA), and used as received. Ultrapure deionized H_2O (18.2 MΩ cm resistivity) was obtained from a Milipore system (Milli-Q Biocel). *Tetrakis*(4-azidophenyl)methane[S1] (tetraazide core) and 1,6-diazidohexane[S2] (diazide core) were synthesized according to previously published procedures. Syntheses of DNA sequences were performed on an Expedite 8909 Nucleic Acid system. Unmodified DNAs and SMDHs were purified on an Agilent 1100 HPLC equipped with reverse-phase (RP) semi-preparative (Dynamax, 250×10 mm, Microsorb 300 Å/10 μm/C18, Agilent # R083213C10) and analytical columns (Dynamax, 100×4.6 mm, Microsorb 100 Å/3 μm/C18, Agilent # R0080200E3). Absorption spectra of DNA materials were recorded on a Varian Cary 300 Bio UV-vis spectrophotometer using a masked quartz cell (path length = 10 mm, catalog # 29B-Q-10-MS, Starna cells Inc., Atascadero, CA). Matrix-assisted laser desorption/ionization time-of-flight (MALDI-ToF) data was collected on a Bruker AutoFlex III MALDI-ToF mass spectrometer (Bruker Daltonics, Billerica, MA). Data from Agilent HPLC and Bruker MALDI-ToF instruments were process using MestreNova software version: 8.1.1-11591.

General synthesis procedure for single-stranded (ss) alkyne-modified DNA on CPG beads. In a typical procedure, syntheses were carried out from the 3′ direction using controlled pore glass (CPG) beads possessing 1-3 μmol of either adenine (Glen Research, dA-CPG # 20-2001-10, (1000 Å, 38 μmol/g)) or thymine attached to the surface (Glen Research, dT-CPG # 20-2030-10 (500 Å, 43 μmol/g), # 25-2030-18 (500 Å, 127 μmol/g), # 20-2031-10 (1000 Å, 26 μmol/g), and # 20-2032-10 (2000 Å, 34 μmol/g)) or universal hybridCPG™ (Glen Research # 28-5040-02 (1000 Å, 74 μmol/g)). The CPG beads were placed in a 1 μmol synthesis column and 3′-phosphoramidites (Glen Research, dA-CE phosphoramidite # 10-1000-C5, Ac-dC-CE phosphoramidite # 10-1015-C5, dmf-dG-CE phosphoramidite # 10-1029-C5, dT-CE phosphoramidite # 10-1030-C5) and 5′-hexynyl phosphoramidite (Glen Research, # 10-1908-90) were then added using the standard 1-3 μmol protocol on an Expedite 8909 synthesizer to make the CPG-3′-ss-DNA-C_4-alkyne (see Table 1 in the main text for sequences). At the end of the synthesis, the beads were dried overnight under high vacuum, removed from the column, and kept in a tightly capped vial at ambient conditions. The solid-phase coupling reactions with the organic core were performed using these dry CPG beads.

Synthesis of 3′-TTCCTT-C_4-alkyne. DNA-containing CPG beads (6.5 mg of 500 Å, 43 μmol/g) containing 3′-TTCCTT-C_4-alkyne were placed in AMA deprotecting solution (1 mL of 1:1 v/v mixture of 28-30 wt% NH_3 in water and 40 wt% methyl amine in water) at 65 °C for 15 minutes. (*CAUTION*: Only fresh AMA solutions that are not more than two weeks old and have been kept in the refrigerator below 0 °C should be used). Afterwards, ammonia and methyl amine were removed by passing a stream of N_2 over the AMA solution until the smells of ammonia and methyl amine were no longer present. The remaining solution was diluted to 0.5 mL with ultrapure deionized H_2O and filtered through 0.45 μm nylon syringe filter (Acrodisc® 13 mm syringe filter # PN 4426T) to

afford the pure product (isolated yield = 70%), which was confirmed by analytical HPLC and MALDI-ToF mass spectroscopy (See Figure 2A in the main text).

General solid-phase synthesis procedure for SMDHs. Dry CPG beads containing alkyne-modified DNA (0.1-1 µmol) were placed in an Eppendorf tube. The tetraazide core (1-15 mM in DMF, 2-30 equivalents based on the alkyne-DNA strands on CPG beads. *NOTE*: the solubility limit of the tetraazide is ~10 mM in DMF) or 1,6-diazidohexane (1-106 mM in DMF, 4-400 equivalents based on the alkyne-DNA strands on CPG) was next added to the tube. *Tris*[(1-benzyl-1H-1,2,3-triazol-4-yl)methyl]amine (TBTA, 50 mM in DMF, 30 equivalents based on the alkyne-DNA strands on CPG), $CuSO_4 \cdot 5H_2O$ (100 mM in DMF, 25 equivalents based on the alkyne-DNA strands on CPG), and L-ascorbic acid (200 mM in DMF, 50 equivalents based on the alkyne-DNA strands on CPG) were also added. The reaction mixture was then blanketed with nitrogen before capping and shook for 18 hours at 25 °C in an Eppendorf® Thermomixer® R (Eppendorf, # 022670107) at 1500 rpm (*NOTE*: efforts should be made to make sure that all the CPG beads are properly agitated in DMF solution and not just sit at the bottom of the tube during the reaction).

We note that the concentration of the copper catalyst in the solid-phase reaction must be higher than that in the solution-phase for reaction to proceed to completion. Due to the practical needs of obtaining an accurate mass of the CPG beads and for proper agitation of the beads, solid-phase experiments are best carried out at 1 µmol scale or larger.

Solid-phase synthesis procedure for mixed-rSMDH$_4$ consisting of three identical strands and one different strand. Dry CPG beads (2 mg) containing 18-bp alkyne-DNA (~20 nmol) were placed in an Eppendorf tube. The $rSMDH_3$ product containing three strands and one free azide ((3′ -TTTCCTTTTT-C_4-triazole)$_3$-core-N_3, see Figure S13 for its HPLC and MALDI-ToF data, 8.7 nmol in 25 µL in H_2O), *tris*(hydroxylpropyl)triazolylamine (THPTA, 0.52 µmol in 5.7 µL of H_2O), $CuSO_4 \cdot 5H_2O$ (0.44 µmol in 2.2 µL of H_2O), and L-ascorbic acid (3.48 µmol in 3.8 µL of H_2O) were then added. The resulting mixture was then blanketed with nitrogen and rotated for 18 h at 25 °C in a Labquake* tube rotator (Thermo Scientific, Model # 400110).

General work-up procedure for solid-phase synthesis of all SMDH strands. CPG beads were filtered using a one-side fritted 1 µmol Expedite DNA synthesis column (Glen Research, # 20-0021-01), then the beads were washed with DMF (5×1 mL) and acetone (5×1 mL) and dried by passing N_2 over the beads. CPG beads containing the products were placed in 1 mL of AMA (*CAUTION*: Only fresh AMA solutions that are not more than two weeks old and have been kept in the refrigerator below 0 °C should be used) at 65 °C for 15 minutes for standard CPGs (1 hour for hybridCPG™) to cleave the SMDHs from the solid supports. Afterwards, the ammonia and methyl amine were removed by passing a stream of N_2 over the AMA solution until the smells of ammonia and methyl amine were no longer present. The remaining SMDHs were extracted with ultrapure deionized H_2O (affording total 0.2-1 mL at the end), and the resulting solution was filtered through 0.45 µm nylon syringe filter (Acrodisc® 13 mm syringe filter # PN 4426T).

Solution-phase synthesis procedure for SMDH strands. 3′-TTCCTT-C_4-alkyne-DNA (3.82 nmol in 10 µL H_2O) was placed in an Eppendorf tube. The tetraazide core (114.6 nmol in 79.3 µL of DMF, 30 equiv based on the alkyne-DNA), TBTA (114.6 nmol in 6.1 µL of DMF, 30 equiv based on the alkyne-DNA strands on CPG), $CuSO_4 \cdot 5H_2O$ (76.4 nmol in 9.5 µL of DMF, 20 equiv based on the alkyne-DNA), and L-ascorbic acid (152.8 nmol in 5.4 µL of DMF, 40 equiv based on the alkyne-DNA) were then added into the tube. The reaction mixture was blanketed with nitrogen and kept at 25 °C for 30 minutes. Then, all of the reaction mixture (110 µL) was injected into an analytical HPLC column and run with +1 vol% MeCN/minute gradient method (See purification section below).

We note that the concentration of the copper catalyst in the solution-phase reaction was greatly reduced to avoid copper-induced damages to the DNA components. In addition, a smaller scale of the solution-phase experiment was used to avoid possible damage to the stationary phase of the HPLC column (due to the presence of DMF) during purification.

Purification of rSMDH strands. The reaction mixtures were first analyzed using an analytical RP-HPLC column (Dynamax, 100 × 4.6 mm, Microsorb 100 Å/3 µm/C18, Agilent # R0080200E3) and a gradient method beginning with 95:5 v/v 0.1 M TEAA (aq):MeCN (TEAA (aq) = triethylammonium acetate, aqueous solution), and increasing to 60:40 v/v 0.1 M TEAA(aq):MeCN over 35 minutes (at a ramp of +1 vol% MeCN/minute), with a flow rate of 1 mL/min. Then, SMDHs were purified using a semi-preparative RP-HPLC column (Dynamax, 250 × 10 mm,

Microsorb 300 Å/10 µm/C18, Agilent # R083213C10) and a gradient method beginning with 95:5 v/v 0.1 M TEAA (aq):MeCN and increasing to 60:40 v/v 0.1 M TEAA(aq):MeCN over 70 minutes (at a ramp of +0.5 vol% MeCN/minute, a slower gradient rate was used for the semi-preparative RP-HPLC to ensure adequate separation of the peaks), with a flow rate of 3 mL/min. All fractions that show UV-vis absorptions from the DNA bases (260 nm) and have elution times that correspond to the analytical RP-HPLC chromatograms were collected, lypholized, and dissolved in H_2O (1 mL). Upon MALDI-ToF analysis, fractions were assigned as rSMDH, $rSMDH_2$, $rSMDH_3$, and $rSMDH_4$ (r = rigid) products or fSMDH and $fSMDH_2$ (f = flexible) products. The dissolved pure products eluted as a single peak through the analytical RP-HPLC column using the aforementioned analytical RP-HPLC solvent program and at a ramp of +1 vol% MeCN/minute gradient method, with a flow rate of 1 mL/min (See Figures S1-21 for details).

MALDI-ToF Experiments of SMDHs. The matrix was prepared by addition of aqueous ammonium hydrogencitrate (0.6 µL of a solution of 15 mg in 30 µL of H_2O) to a solution of 2-hydoxypicolinic acid (Fluka # 56297, 2 mg in H_2O:MeCN (30 µL, 1:1 v/v). An aliquot (1 µL, 10-100 pmol) of the isolated pure DNA-hybrid sample was then mixed with a portion of this matrix (5 µL). A small amount of the resulting mixture (0.3-3 µL) was dropped on a steel MALDI-ToF plate and dried at 25 °C before being analyzed on a Bruker (Billerica, MA) Daltonics AutoFlex III MALDI-ToF mass spectrometer as negative or positive ions using the linear mode. The instrument was equipped with Smartbeam™ laser technology operated at 30-90% power with a sampling speed of 10 Hz. Five hundred spectra were averaged for a mass spectrum. The instrument was operated using the following parameters: ion source voltage 1 = 20 kV, ion source voltage 2 = 18.5 kV, lens voltage = 8.5 kV, linear detector voltage = 0.6 kV, deflection mass = 3000 Da. The MALDI-ToF spectra of SMDHs were shown in the inset of the pure analytical traces of the rSMDHs.

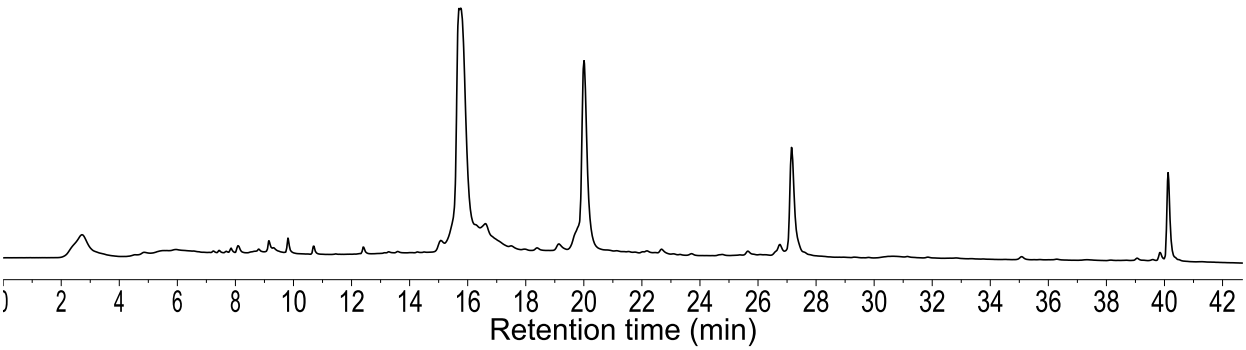

Figure S1. Analytical RP-HPLC trace of the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 6-bp alkyne-DNA (3′-TTCCTT) on the CPGs. The trace is the signal from the diode detector set at 260 nm. The fractions between 15.5-16.5 min ($rSMDH_4$ product, see Figure 2A in the main text for the analytical RP-HPLC and MALDI-ToF data of the pure product), 19.9-20.4 min ($rSMDH_3$ product, see Figure S2 for the analytical RP-HPLC and MALDI-ToF data of the pure product), 27-27.5 min ($rSMDH_2$ product, see Figure S3 for the analytical RP-HPLC and MALDI-ToF data of the pure product), and 40-40.3 min (rSMDH product, see Figure S4 for the analytical RP-HPLC and MALDI-ToF data of the pure product) were collected.

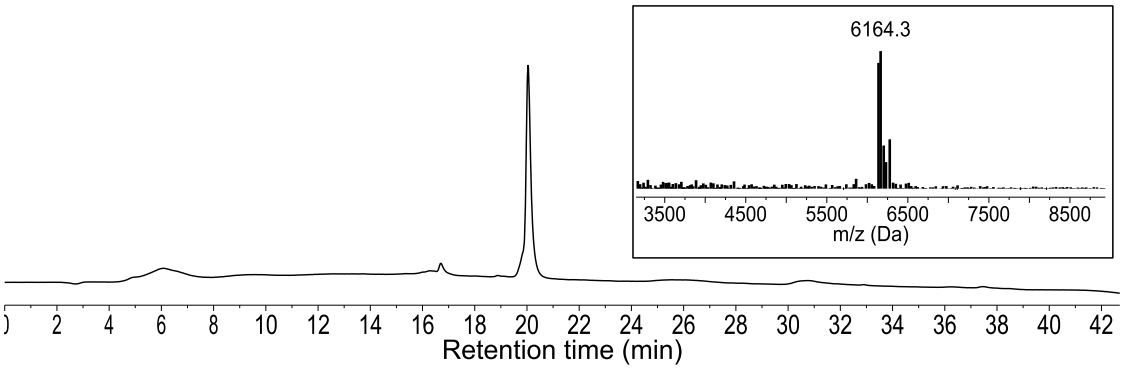

Figure S2. Analytical RP-HPLC trace of the pure $rSMDH_3$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 6-bp alkyne-DNA (3′-TTCCTT) on the CPGs. This material was from the 19.9-20.4 min fraction collected during an initial purification of the reaction mixture using an analytical RP-HPLC column (Figure S1). The trace is the signal from the diode detector set at 260 nm. The inset shows the MALDI-ToF spectra of the pure product: m/z = 6164.3 (6164.4 theoretical).

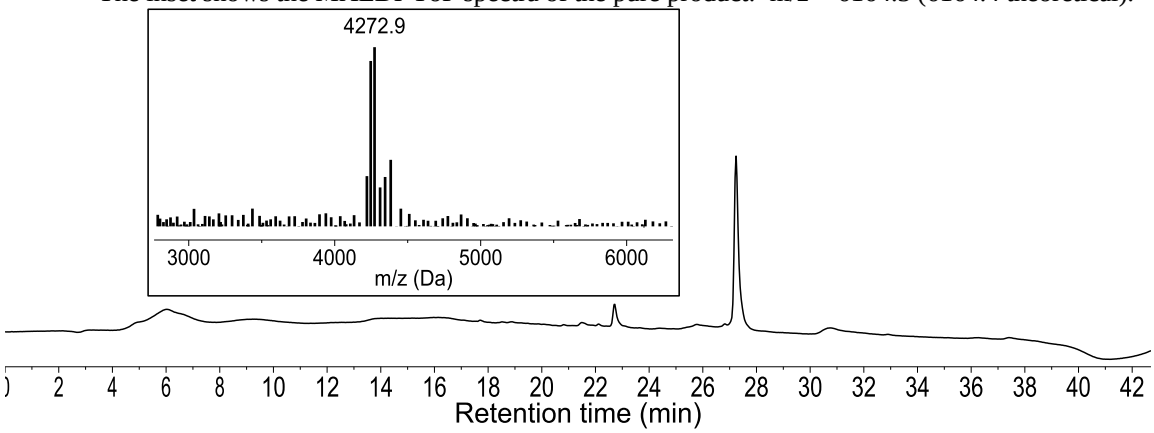

Figure S3. Analytical RP-HPLC trace of the pure $rSMDH_2$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 6-bp alkyne-DNA (3′-TTCCTT) on the CPGs. This material was from the 27-27.5 min fraction collected during an initial purification of the reaction mixture using an analytical RP-HPLC column (Figure S1). The trace is the signal from the diode detector set at 260 nm. The inset shows the MALDI-ToF spectra of the pure product: m/z = 4272.9 (4271.1 theoretical).

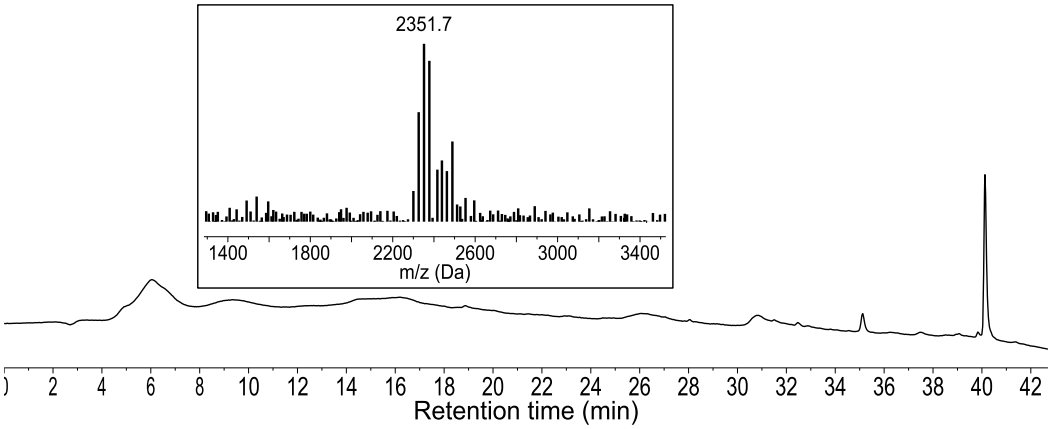

Figure S4. Analytical RP-HPLC trace of the pure rSMDH product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 6-bp alkyne-DNA (3′-TTCCTT) on the CPGs. This material was from the 40-40.3 min fraction collected during an initial purification of the reaction mixture using an analytical RP-HPLC column (Figure S1). The trace is the signal from the diode detector set at 260 nm. The inset shows the MALDI-ToF spectra of the pure product: m/z = 2351.7 (2377.8 theoretical).

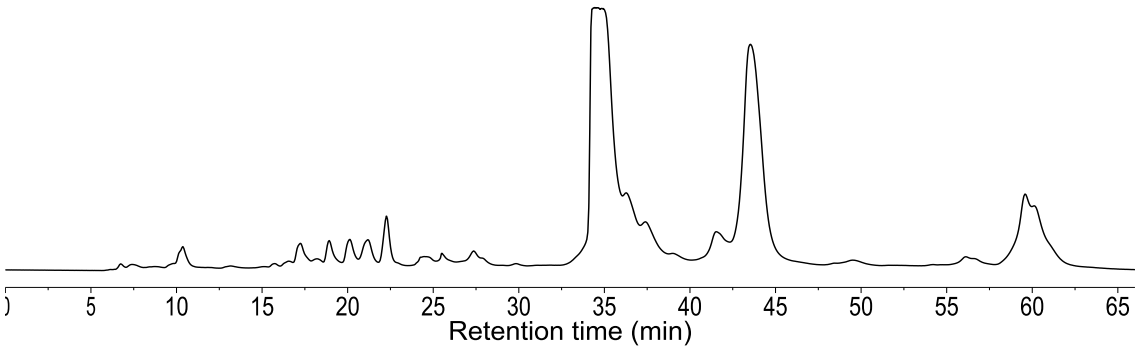

Figure S5. Semi-preparative RP-HPLC trace of the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 7-bp alkyne-DNA (3′-TTTCCTT) on the CPGs. The trace is the signal from the diode detector set at 260 nm. The fractions between 34-35.5 min ($rSMDH_4$ product, see Figure S6 for analytical HPLC and MALDI-ToF data of the pure product) and 43-44.5 min ($rSMDH_3$ product, see Figure S7 for analytical HPLC and MALDI-ToF data of the pure product) were collected.

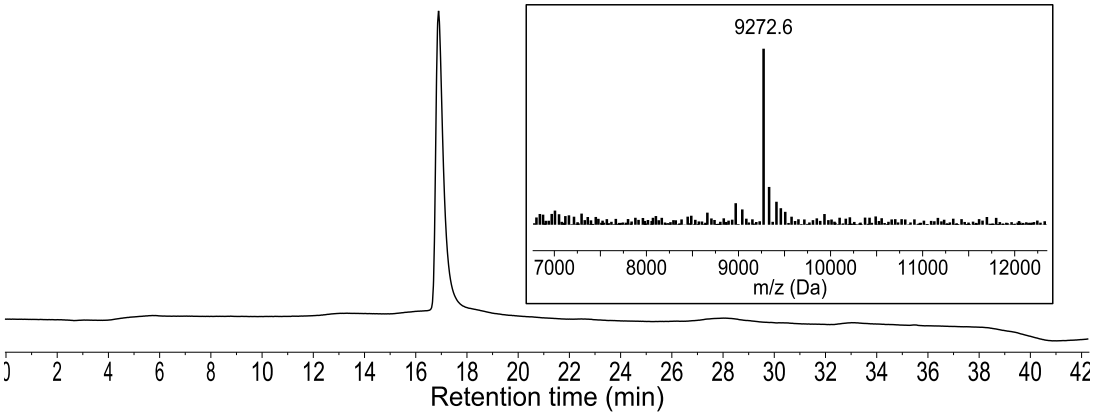

Figure S6. Analytical RP-HPLC trace of the pure $rSMDH_4$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 7-bp alkyne-DNA (3′-TTTCCTT) on the CPGs. This material was from the 34-35.5 min fraction collected during an initial purification of the reaction mixture using a semi-preparative RP-HPLC column (Figure S5). The trace is the signal from the diode detector set at 260 nm. Inset shows the MALDI-ToF spectra of the pure product: m/z = 9272.6 (9274.5 theoretical).

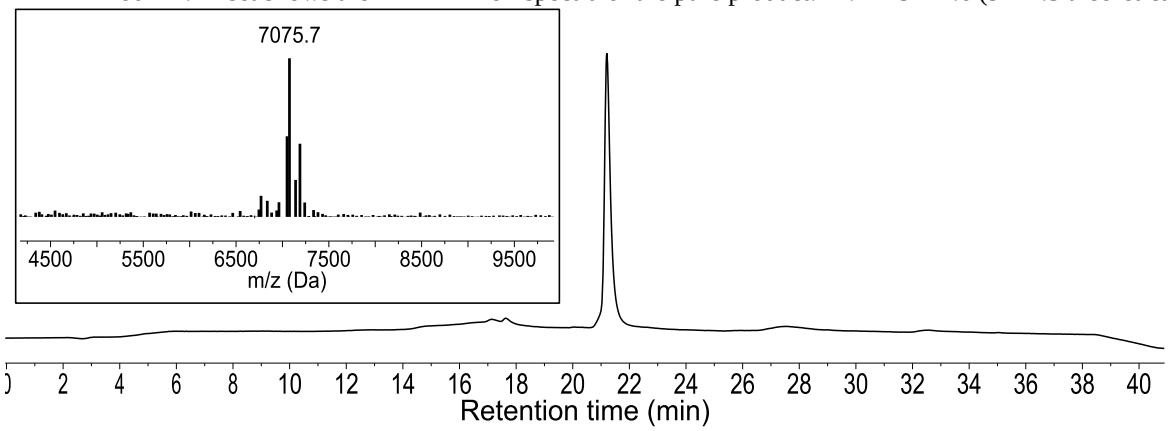

Figure S7. Analytical RP-HPLC trace of the pure $rSMDH_3$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 7-bp alkyne-DNA (3′-TTTCCTT) on the CPGs. This material was from the 43-44.5 min fraction collected during an initial purification of the reaction mixture using a semi-preparative RP-HPLC column (Figure S5). The trace is the signal from the diode detector set at 260 nm. Inset shows the MALDI-ToF spectra of the pure product: m/z = 7075.7 (7077.0 theoretical).

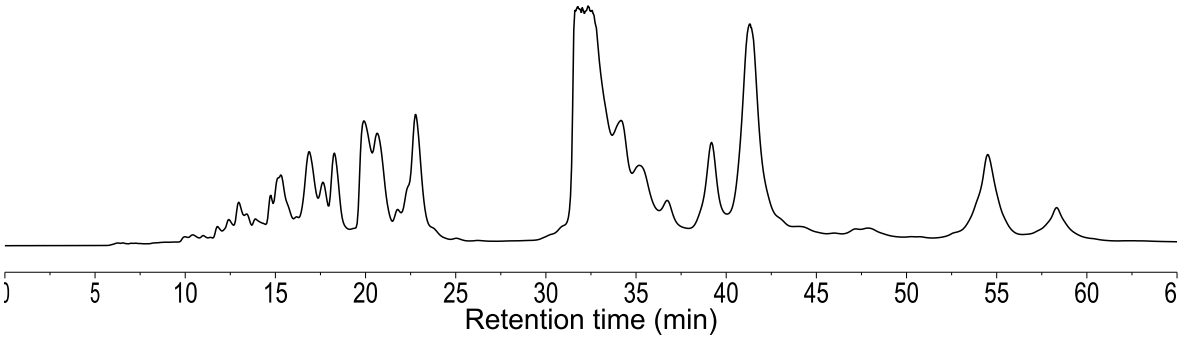

Figure S8. Semi-preparative RP-HPLC trace of the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 7-bp alkyne-DNA (3′-AAGGAAA) on the CPGs. The trace is the signal from the diode detector set at 260 nm. The fractions between 31.4-33.2 min ($rSMDH_4$ product, see Figure S9 for analytical HPLC and MALDI-ToF data of the pure product) and 40.5-42.2 min ($rSMDH_3$ product, see Figure S10 for analytical HPLC and MALDI-ToF data of the pure product) were collected.

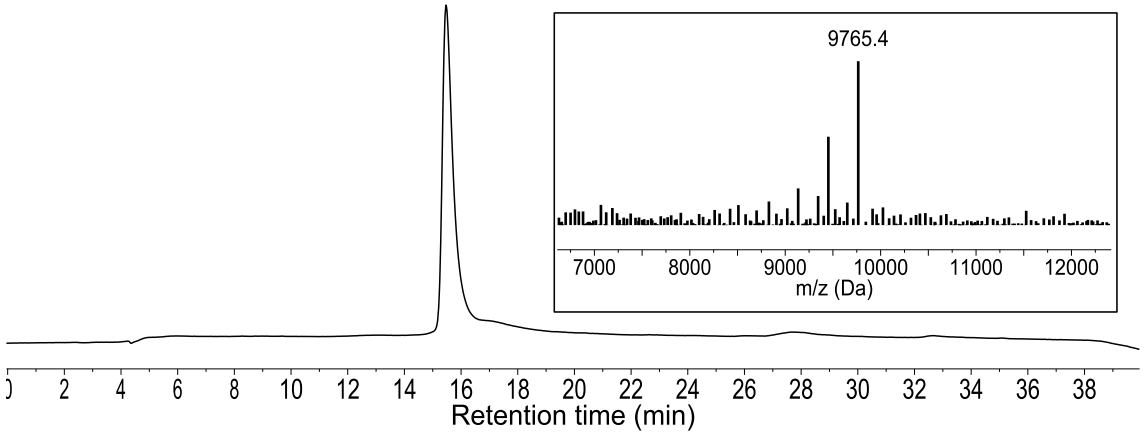

Figure S9. Analytical RP-HPLC trace of the pure $rSMDH_4$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 7-bp alkyne-DNA (3′-AAGGAAA) on the CPGs. This material was from the 31.4-33.4 min fraction collected during an initial purification of the reaction mixture using a semi-preparative RP-HPLC column (Figure S8). The trace is the signal from the diode detector set at 260 nm. Inset shows the MALDI-ToF spectra of the pure product: m/z: 9765.4 (9774.9 theoretical).

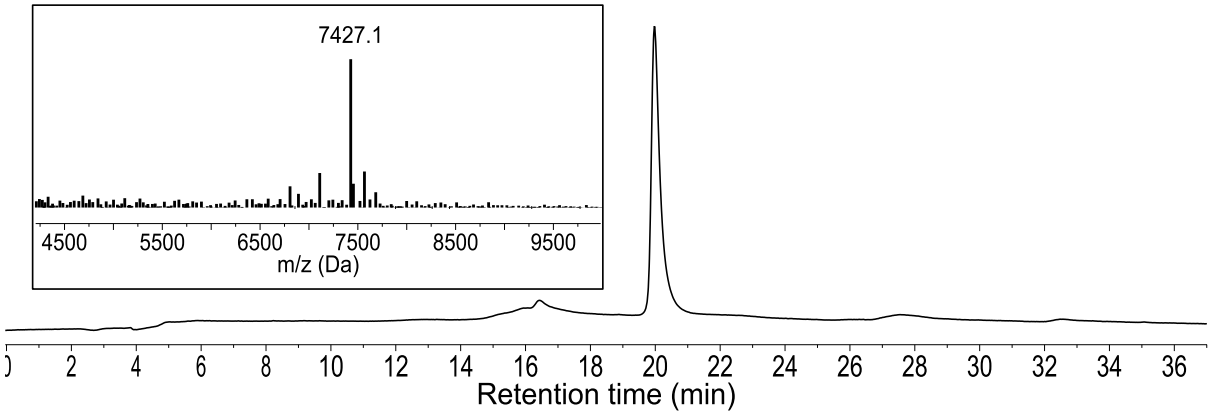

Figure S10. Analytical RP-HPLC trace of the pure $rSMDH_3$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 7-bp alkyne-DNA (3′-AAGGAAA) on the CPGs. This material was from the 40.5-42.2 min fraction collected during an initial purification of the reaction mixture using a semi-preparative RP-HPLC column (Figure S8). The trace is the signal from the diode detector set at 260 nm. Inset shows the MALDI-ToF spectra of the pure product: m/z: 7427.1 (7452.3 theoretical).

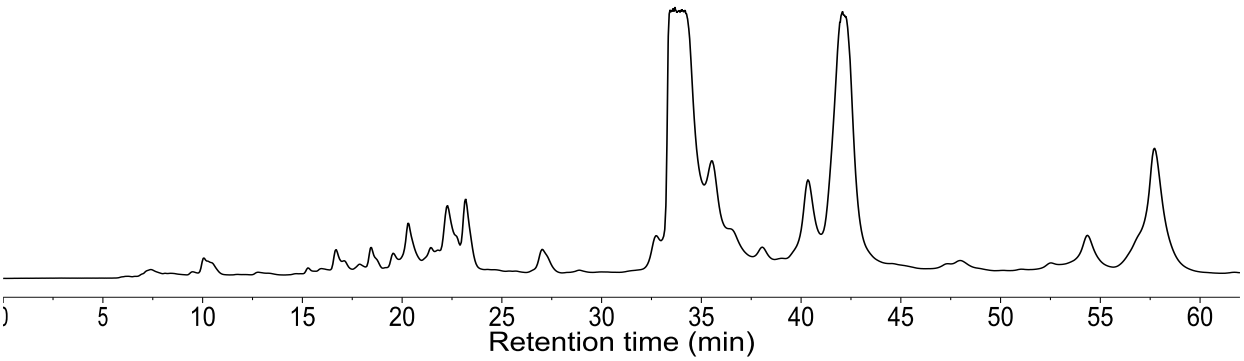

Figure S11. Semi-preparative RP-HPLC trace of the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 10-bp alkyne-DNA (3′-TTTCCTTTTT) on the CPGs. The trace is the signal from the diode detector set at 260 nm. The fractions between 33.2-34.7 min (rSMDH$_4$ product, see Figure S12 for analytical HPLC and MALDI-ToF data of the pure product) and 41.3-43 min (rSMDH$_3$ product, see Figure S13 for analytical HPLC and MALDI-ToF data of the pure product) were collected.

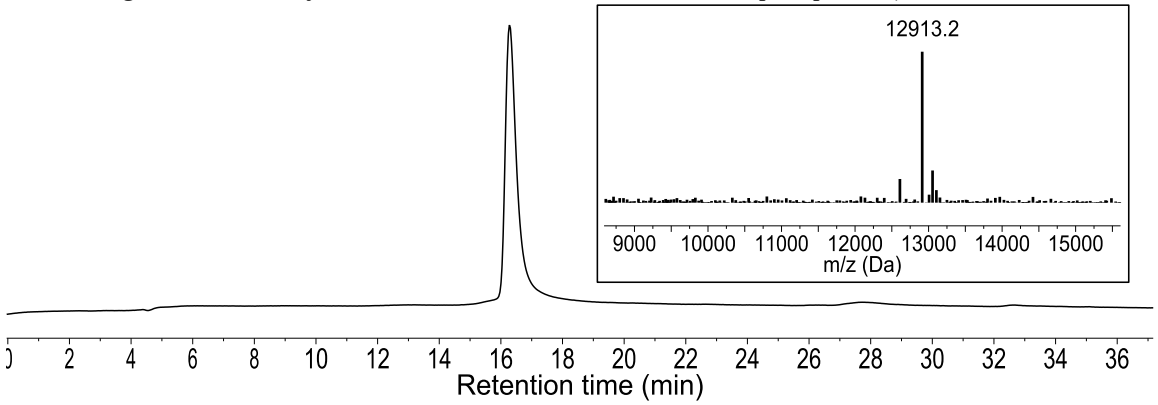

Figure S12. Analytical RP-HPLC trace of the pure rSMDH$_4$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 10-bp alkyne-DNA (3′-TTTCCTTTTT) on the CPGs. This material was from the 33.2-34.7 min fraction collected during an initial purification of the reaction mixture using a semi-preparative RP-HPLC column (Figure S11). The trace is the signal from the diode detector set at 260 nm. Inset shows the MALDI-ToF spectra of the pure product: m/z: 12913.2 (12924.9 theoretical).

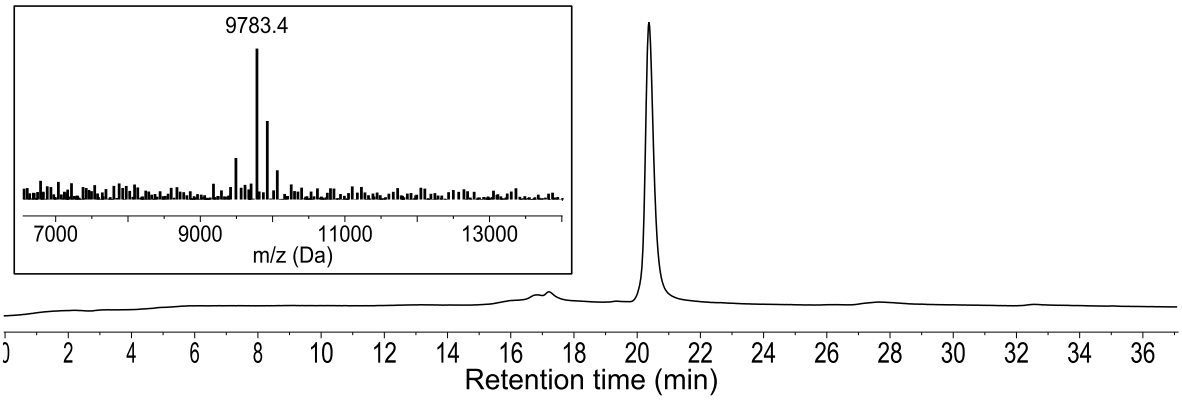

Figure S13. Analytical RP-HPLC trace of the pure rSMDH$_3$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 10-bp alkyne-DNA (3′-TTTCCTTTTT) on the CPGs. This material was from the 41.3-43 min fraction collected during an initial purification of the reaction mixture using a semi-preparative RP-HPLC column (Figure S11). The trace is the signal from the diode detector set at 260 nm. Inset shows the MALDI-ToF spectra of the pure product: m/z: 9783.4 (9814.8 theoretical).

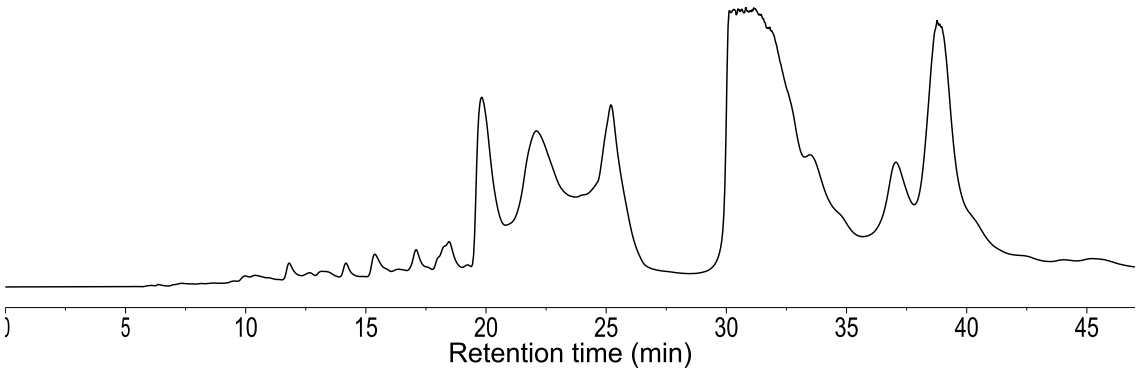

Figure S14. Semi-preparative RP-HPLC trace of the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 18-bp alkyne-DNA (3′-AACAATTATACTCAGCAA) on the CPGs. The trace is the signal from the diode detector set at 260 nm. The fractions between 30-31.8 min (rSMDH$_4$ product, see Figure S15 for analytical HPLC and MALDI-ToF data of the pure product) and 38-40 min (rSMDH$_3$ product, see Figure S16 for analytical HPLC and MALDI-ToF data of the pure product) were collected.

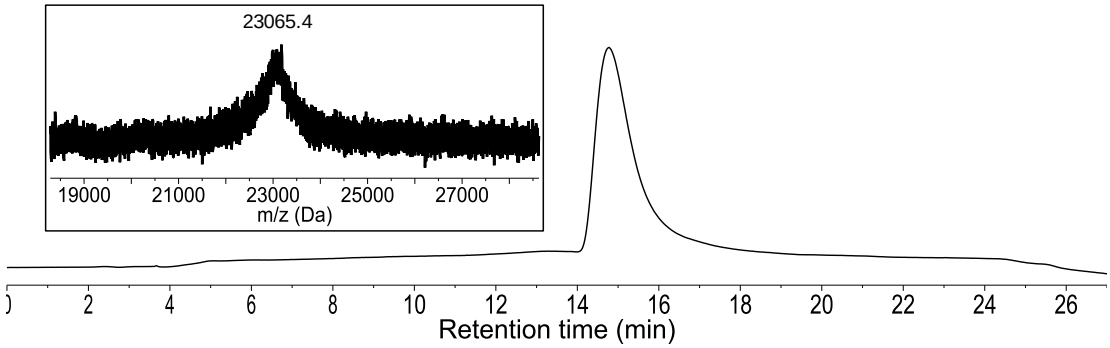

Figure S15. Analytical RP-HPLC trace of the pure rSMDH$_4$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 18-bp alkyne-DNA (3′-AACAATTATA-CTCAGCAA) on the CPGs. This material was from the 30-31.8 min fraction collected during an initial purification of the reaction mixture using a semi-preparative RP-HPLC column (Figure S14). The trace is the signal from the diode detector set at 260 nm. Inset shows the MALDI-ToF spectra of the pure product: m/z: 23065.4 (22963.7 theoretical).

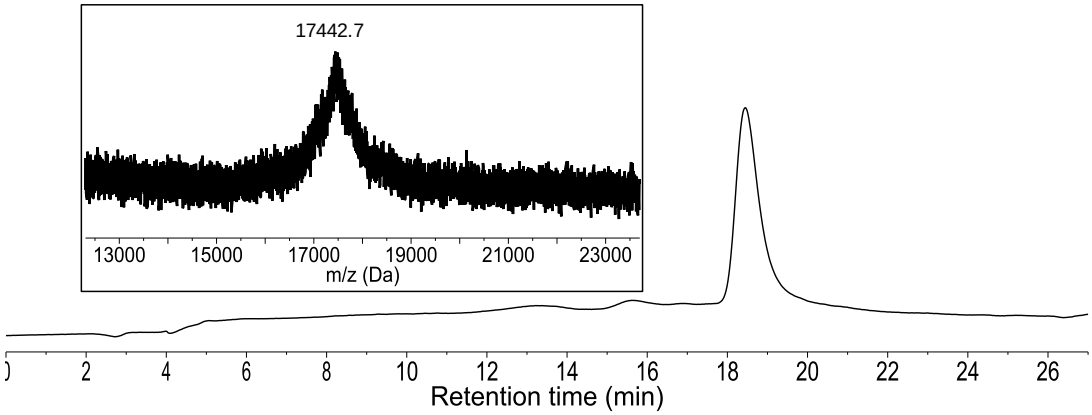

Figure S16. Analytical RP-HPLC trace of the pure rSMDH$_3$ product from the coupling reaction of the *tetrakis*(4-azidophenyl) methane core with 18-bp alkyne-DNA (3′-AACAATTATA-CTCAGCAA) on the CPGs. This material was from the 38-40 min fraction collected during an initial purification of the reaction mixture using a semi-preparative RP-HPLC column (Figure S14). The trace is the signal from the diode detector set at 260 nm. Inset shows the MALDI-ToF spectra of the pure product: m/z: 17442.7 (17343.9 theoretical).

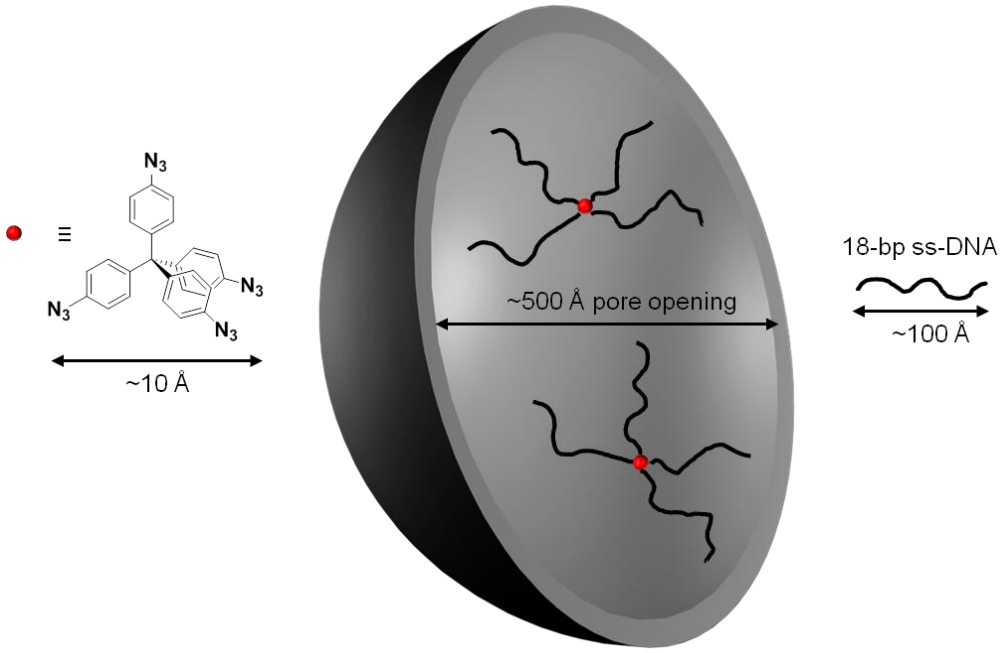

Figure S17. A schematic representation of the solid-phase click coupling reaction of the *tetrakis*(4-azidophenyl) methane core (red sphere) with 18-bp alkyne-DNA (black curved lines) on a CPG pore with 500 Å diameter (pores were shown as open half spheres). To the best of our ability to illustrate, the objects were drawn to scale to show that the formation of the $SMDH_4$ product is possible given the large difference in length scales between the ssDNA and core (10:1), the high flexibility of the ssDNA strand on the solid support; and the relatively "high" curvature of the CPG pores and DNA density.

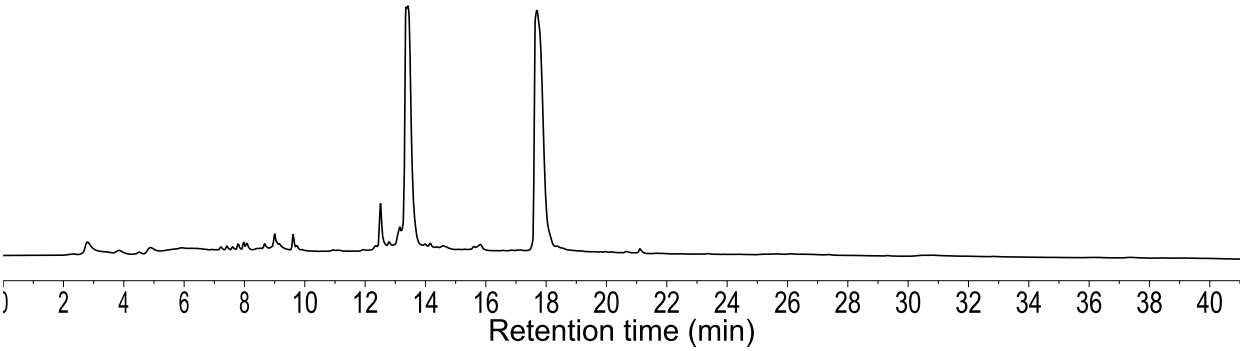

Figure S18. Analytical RP-HPLC trace of the coupling reaction of the 1,6-diazidohexane core with 6-bp alkyne-DNA (3′-TTCCTT) on the CPGs. The trace is the signal from the diode detector set at 260 nm. The fractions between 13.2-13.7 min ($fSMDH_2$ product, see Figure S18 for analytical RP-HPLC and MALDI-ToF data of the pure product) and 17.5-18.1 min (fSMH product, see Figure S19 for analytical HPLC and MALDI-ToF data of the pure product) were collected.

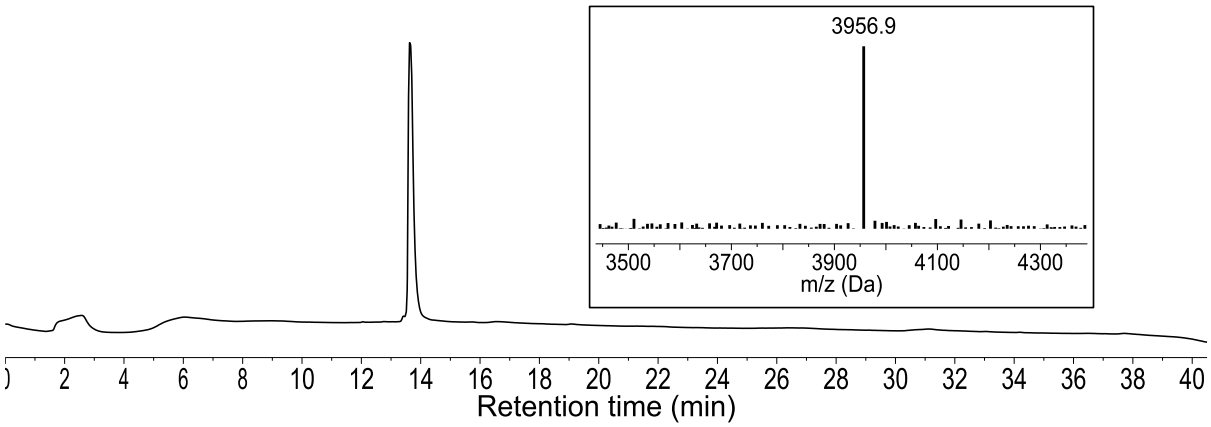

Figure S19. Analytical RP-HPLC trace of the pure $fSMDH_2$ product from the coupling reaction of the 1,6-diazidohexane core with 6-bp alkyne-DNA (3′-TTCCTT) on the CPGs. This material was from the 13.2-13.7 min fraction collected during an initial purification of the reaction mixture using an analytical RP-HPLC column (Figure S17). The trace is the signal from the diode detector set at 260 nm. The inset shows the MALDI-ToF spectra of the pure product: m/z = 3956.9 (3956.8 theoretical).

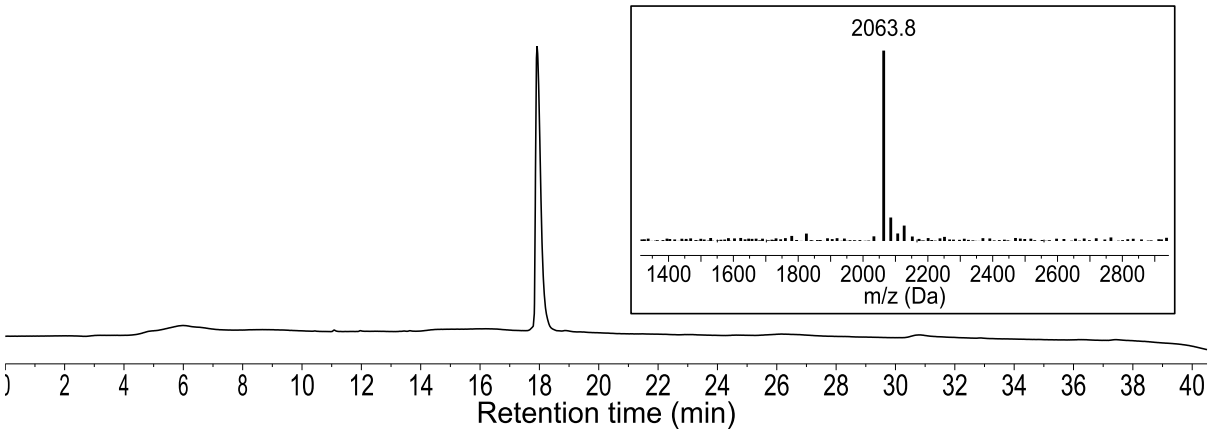

Figure S20. Analytical RP-HPLC trace of the pure fSMDH product from the coupling reaction of the 1,6-diazidohexane core with 6-bp alkyne-DNA (3′-TTCCTT) on the CPGs. This material was from the 17.5-18.1 min fraction collected during an initial purification of the reaction mixture using an analytical RP-HPLC column (Figure S17). The trace is the signal from the diode detector set at 260 nm. The inset shows the MALDI-ToF spectra of the pure product: m/z = 2063.8 (2063.5 theoretical).

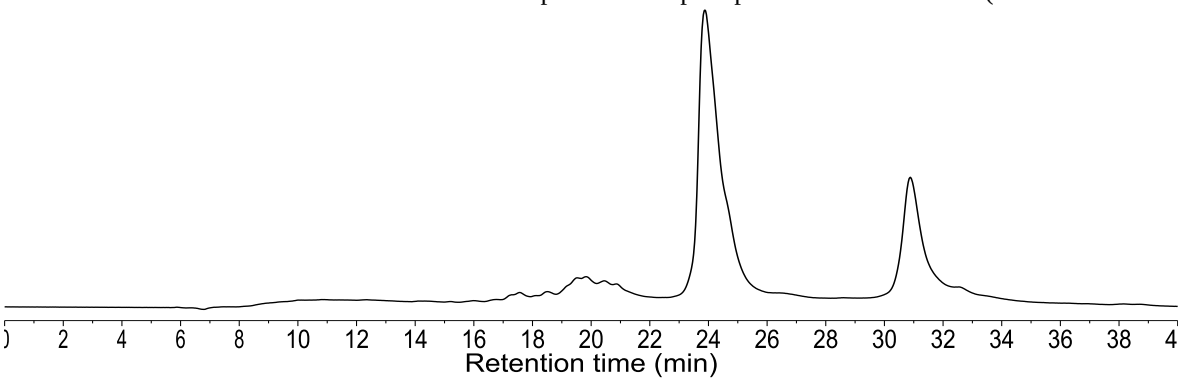

Figure S21. Semi-preparative RP-HPLC trace of the coupling reaction of $rSMDH_3$ (3′-TTTCCTTTTT, see Figure S13) with an 18-bp alkyne-DNA (3′-TTGCTGAGTATAATTGTT) strand on the CPGs. The trace is the signal from the diode detector set at 260 nm. The fraction between 30-32 min was collected and is the desired product.

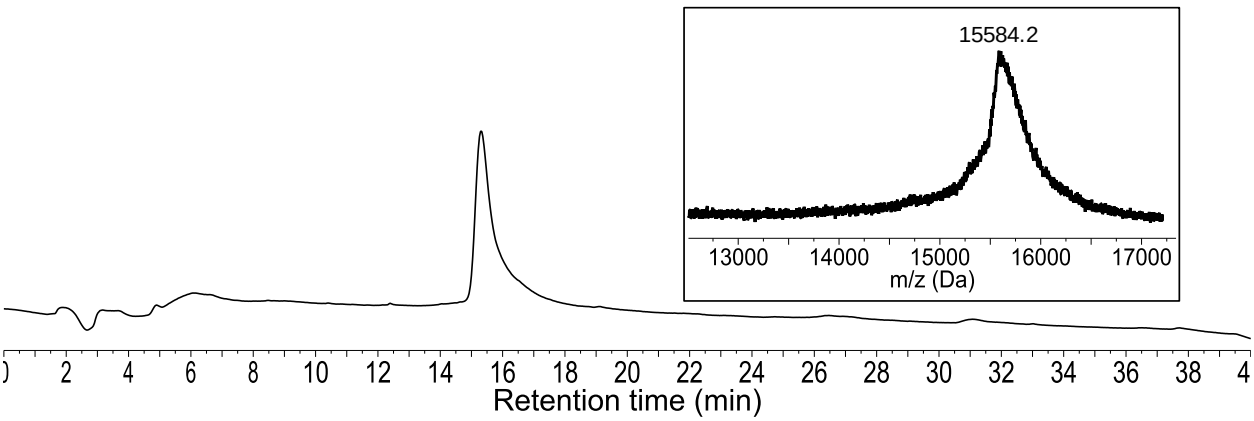

Figure S22. Analytical RP-HPLC trace of pure mixed-rSMDH$_4$ consisting of three identical strands (3′-TTTCCTTTTT) and one different strand (3′ -TTGCTGAGTATAATTGTT). This material was from the 30-32 min fraction collected during an initial purification of the reaction mixture using a semi-preparative RP-HPLC column (Figure S21). The trace is the signal from the diode detector set at 260 nm. Inset shows the MALDI-ToF spectra of the pure product (m/z): 15584.2 (15509.6 theoretical).

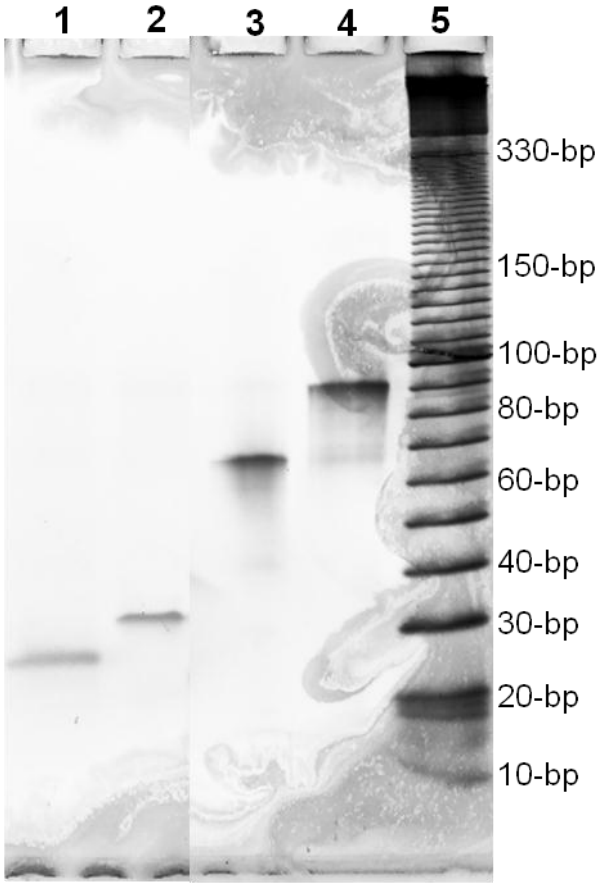

Figure S23. Denaturing PAGE-gel image (8%) of rSMDHs (Gel was prepared in 8 M urea and 1X TBE buffer (89 mM TRIS-base, 89 mM boric acid, 2 mM EDTA), and run at RT for 30 min at a 350 V potential). From left to right: *lane 1* = rSMDH$_3$ with 6-bp ssDNA (3′-TTCCTT), *lane 2* = rSMDH$_4$ with 6-bp ssDNA (3′-TTCCTT), *lane 3* = rSMDH$_3$ with 18-bp ssDNA (3′-TTGCTGAGTATAATTGTT), *lane 4* = rSMDH$_4$ with 18-bp ssDNA (3′-TTGCTGAGTATAATTGTT), *lane 5* = 10 bp DNA ladder (Invitrogen). The whirly features at the top, right and bottom edges of the gel image are artifacts from the handling and staining of the gel.

Author contributions audit. I.E. and S.T.N. conceived the experiments presented herein. The tetraazide core was prepared by O.K.F. The remainder of the materials were prepared and characterized by I.E. and R.V.T, both of whom carried out all the experiments and wrote the initial draft of the paper with inputs from all the co-authors. S.T.N. and C.A.M. supervised the project. I.E., R.V.T., and S.T.N. finalized the manuscript.

References.

(S1) Pandey, P.; Farha, O. K.; Spokoyny, A. M.; Mirkin, C. A.; Kanatzidis, M. G.; Hupp, J. T.; Nguyen, S. T. *J. Mater. Chem.* **2011**, *21*, 1700.
(S2) Romuald, C.; Busseron, E.; Coutrot, F. *J. Org. Chem.* **2010**, *75*, 6516.