Supplementary Information for

What is a "DNA-compatible" reaction?

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Methods

Materials Sources. All reagents were obtained from Sigma Aldrich (St. Louis, MO) unless otherwise specified. 1,3-Bis[tris(hydroxymethyl)methylamino]propane (Bis-Tris), trifluoroacetic acid (TFA), triisopropylsilane (TIPS), N-methyl-2-pyrrolidone (NMP), triphenylphosphine ($P(Ph)_3$), triethylamine trihydrofluoride (TEA・3HF), α-cyano-4-hydroxycinnamic acid (HCCA), 2′,4′,6′-Trihydroxyacetophenone monohydrate (THAP), Dynabeads M-270 Carboxylic Acid (Life Technologies, Carlsbad, CA), Dynabeads M-270 Streptavidin (Life Technologies, Carlsbad, CA), triethylamine acetate (TEAA, 2 M, Life Technologies), N,N'-diisopropylcarbodiimide (DIC, Acrōs Organics, Fair Lawn, NJ), L(+)-ascorbic acid (Acrōs Organics), 1-hydroxy-7-azabenzotriazole (HOAt, Accela ChemBio Inc., San Diego, CA), MB 160 230 Tentagel MB Rink Amide resin (Tentagel RAM resin, Rapp Polymere, Tuebingen, Germany), Fmoc-Tyr(tBu)-OH (CEM, Matthews, NC), Fmoc-Gly-OH (AnaSpec, Fremont, CA), Fmoc-Arg(Pbf)-OH (AnaSpec), Fmoc-Glu(OAll)-OH (AnaSpec), dimethylformamide (DMF, Thermo Fisher Scientific, Waltham, MA), dichloromethane (DCM, Thermo Fisher Scientific), N,N-diisopropylethylamine (DIEA, Thermo Fisher Scientific), tris(2-carboxyethyl)phosphine hydrochloride (TCEP HCl, Thermo Fisher Scientific), 7-azabenzotriazol-1-yloxy)tripyrrolidinophosphonium hexafluorophosphonate (PyAOP, Oakwood Products, Inc., West Columbia, SC), phenylsilane (Oakwood), 4-isopropylphenylboronic acid (Synthonix, Wake Forest, NC), 3,4-(methylenedioxy)phenylboronic acid (Synthonix), 4-(chloromethyl)phenyl isocyanate (Alfa Aesar, Ward Hill, MA), biotin N-hydroxysulfosuccinimidyl ester (biotin-sNHS, Pierce Biotechnologies, Rockford, IL), Taq DNA polymerase (Taq, New England Biolabs, Ipswich, MA), T4 DNA ligase (New England Biolabs), 2′-deoxyribonucleotide triphosphate (dNTP, set of dATP, dTTP, dGTP, dCTP, Promega Corp., Milwaukee, WI) were used as provided.

Tris[(1-benzyl-1H-1,2,3-triazol-4-yl)methyl]amine (TBTA) was recrystallized three times in t-BuOH/H_2O (1:1).[1] Substituted chloropentenoic acid monomers (2S, 3E)-5-chloro-2,4-dimethyl-3-pentenoic acid (S-Me-COPA) and (2R, 3E)-5-chloro-2,4-dimethyl-3-pentenoic acid (R-Me-COPA) were prepared according to previously published methods.[2]

Oligonucleotides (Integrated DNA Technologies, Inc. Coralville, IA) were purchased as desalted lyophilate and used without further purification. Oligonucleotide ligation substrates were 5′-phosphorylated (/5Phos/). Amino-modified headpiece DNA (NH_2-HDNA, /5Phos/GAGTCA/iSp9//iUniAmM//iSp9/TGACTCCC) was HPLC purified at the manufacturer and used without further purification.

Buffers. Bis-Tris propane wash buffer (BTPWB, 30 mM NaCl, 0.04% Tween-20, 10 mM Bis-Tris, pH 7.6), Bis-Tris propane breaking buffer (BTPBB, 10 mM NaCl, 1% SDS, 1% Tween-20, 10 mM Bis-Tris, pH 7.6), Bis-Tris propane breaking buffer with EDTA (BTPBBE, 10 mM NaCl, 1% SDS, 1% Tween-20, 10 mM EDTA, 10 mM Bis-Tris, pH 7.6), borate buffer (150BWB, 150 mM borate, 0.04% Tween-20, pH 9.4), borate buffer (250BWB, 250 mM borate, 0.04% Tween-20, pH 9.4), 10X PCR buffer (2 mM dATP, 2 mM dGTP, 2 mM dCTP, 2 mM dTTP, 15 mM $MgCl_2$, 500 mM KCl, 100 mM Tris, pH 8.3), 1X Tris-Acetate-EDTA-Buffer (TAE, 40 mM Tris, 20 mM acetic acid, 1 mM EDTA, pH 8.4), 10X T4 DNA Ligase Buffer (T4LB, 100 mM $MgCl_2$, 10 mM ATP, 100 mM DTT, 500 mM Tris, pH 7.5), were prepared in DI H_2O.

Synthesis resin linker synthesis (S1). TentaGel Rink amide resin (160 μm, 0.43 mmol/g, 100 mg, Rapp-polymere, Tuebingen, Germany) was transferred to a fritted syringe (2.5 mL, Torviq, Niles, NI) and

swelled in DMF (1 h, 40 °C). Linker synthesis proceeded via iterative cycles of solid-phase peptide synthesis. Each cycle included: (1) 9-fluorenylmethoxycarbonyl (Fmoc) deprotection (20% piperidine in DMF, 2 x 1 mL, 7 min each aliquot, RT, 8 rpm); (2) N-α-Fmoc-amino acid (215 μmol) activation with DIC/HOAt (301 μmol/215 μmol) and incubation (5 min, RT); (3) N-α-Fmoc-amino acid coupling to resin by transferring activated acid (1 mL) to resin and incubating (see conditions below); (4) capping (first cycle only, 20% acetic anhydride in DMF, 15 min, RT). After each deprotection, monomer coupling, and capping step, reactants were expelled and the resin washed (DMF, 3 x 2 mL; DCM, 3 x 2 mL; DMF, 3 x 2 mL). The following N-α-Fmoc-amino acid couplings were performed in order with incubation details noted: (1) Fmoc-Tyr(tBu)-OH (1 h, 50 °C, 13 rpm); (2) Fmoc-Gly-OH (1 h, 50 °C, 13 rpm); (3) Fmoc-Arg(Pbf)-OH (3 h, 50 °C); (4) Fmoc-Gly-OH (1 h, 50 °C, 13 rpm); (5) Fmoc-Arg(Pbf)-OH (3 h, 50 °C, 13 rpm); (6) Fmoc-Gly-OH (1 h, 50 °C, 13 rpm).

Linker characterization. Resin (0.5 mg) was transferred to a clean fritted spin column (Mobicol Classic, MoBiTec GmbH, Goettingen, Germany), washed (DCM, 3 x 0.5 mL), dried *in vacuo*. Cleavage cocktail (90% TFA, 5% DCM, 5% TIPS, 300 μL) was added to resin and incubated (1 h, RT). Cleaved linker was expelled, concentrated *in vacuo*, resuspended (20% DMSO and 0.1% TFA in H_2O, 100 μL), and analyzed by reversed-phase HPLC (XBridge BEH130 C18, 4.6 x 100 mm, 130 Å, 3.5 μm, Waters) with gradient elution (mobile phase A: ACN; mobile phase B: 0.1% TFA in H_2O; 2 - 52% A, 20 min). HPLC fraction aliquots (1 μL) were spotted to a MALDI-TOF MS target plate, dried, covered with HCCA matrix solution (1.5 mg/mL HCCA in 1:2 ACN:0.1% TFA in H_2O) and analyzed via MALDI-TOF MS (Microflex, Bruker Daltonics, Inc., Billerica MA).

Azido headpiece DNA synthesis, purification and characterization. NH_2-HDNA (300 nmol)[3] was dissolved in phosphate buffer (1 M, pH 8.0, 240 µL). 5-azidopentanoic acid NHS ester was prepared by dissolving NHS (9.6 µmoles), EDC (9.6 µmoles), and 5-azidopentanoic acid (7.2 µmoles) in DMF (20 µL) and incubating (30 min, 60 °C). NH_2-HDNA was sparged (N_2, 1 min), added to 5-azidopentanoic acid NHS-ester solution (22 µL), and incubated(2 h, RT). A fresh solution of 5-azidopentanoic acid NHS-ester was prepared as described above, added to the acylation reaction, and the reaction incubated (1 h, RT). The reaction was quenched (1 M Tris, pH 7.6, 100 µL) and incubated (5 min, 60 °C). Azido-HDNA (N_3-HDNA) product was precipitated twice in ethanol. The pellet was dried under N_2, resuspended (20 mM TEAA, pH 8.0), and purified at semi-preparative scale using reversed-phase HPLC (X-Bridge BEH C18 column, 10 mm x 150 mm, 130 Å, 5 µm, Waters) with gradient elution (mobile phase A: H_2O, 20 mM TEAA, pH 8; mobile phase B: ACN; 5% - 12% B, 24 min). A product fraction aliquot (1 µL) was spotted to a MALDI-TOF MS target plate, dried, and covered with THAP matrix solution (18 mg/mL THAP, 7 mg/mL ammonium citrate dibasic in 50:50 ACN:H_2O) and mass analyzed via MALDI-TOF MS (Microflex, Bruker Daltonics Inc., Supporting Information Figure 2).

Biotin-HDNA and biotin-HDNA magnetic resin preparation. NH_2-HDNA (56 nmol)[3] was dissolved in carbonate buffer (60 mM, pH 8.5, 300 µL). Biotin-NHS (2.25 µmoles) was dissolved in DI H_2O (225 µL), combined with NH_2-HDNA solution and incubated (16 h, 4 °C). The DNA was precipitated in ethanol, resuspended in buffer (10 mM Tris, pH 8, 200 µL), and used without further purification. Streptavidin-coated magnetic resin (0.5 mg, 100 pmol sites, Dynabeads M270) was washed (BTPWB, 2 x 200 µL), resuspended (BTPWB, 100 µL), combined with crude Biotin-HDNA (120 pmol) and incubated (15 min, RT). The Biotin-HDNA magnetic resin was washed (BTPWB, 3 x 200 µL) and resuspended (BTPWB,

200 µL).

N-propargyl magnetic resin preparation. Carboxylic acid-functionalized magnetic resin (Dynabeads M-270 Carboxylic acid, 9 mg) was washed (DMF, 3 x 1 mL). Propargylamine (2 M in DMF, 50 µL), DIC (2.8 M in DMF, 50 µL), and HOAt (1 M in DMF, 100 µL) were added to resin and incubated with rotation (3.75 h, 50 °C, 13 rpm). The resin was washed (DMF, 6 x 1 mL; DCM, 2 x 1 mL; 1% Tween 20 in HPLC H_2O, 2 x 1 mL), incubated in 1% Tween 20 (1 h, 50 °C), resuspended in 1% Tween 20, and stored (RT).

HDNA magnetic resin preparation. N-propargyl-functionalized magnetic resin (1.35 mg) was resuspended in copper-catalyzed azide-alkyne cycloaddition (CuAAC) reaction buffer (1.8 mM $CuSO_4$, 2.2 mM TBTA, 10 mM ascorbic acid, 1% Tween 20, 11.1 µM N_3-HDNA, 40% DMSO, 1 M TEAA, pH 7, 27 µL total), and incubated (1.5 h, 50 °C). Resin was washed (BTPBBE, 1 x 100 uL, 1 x 1 mL) and incubated (18 h, 50 °C).

Magnetic "sensor bead" preparation. HDNA magnetic resin (3 x 1.35 mg) was aliquoted into 1.5-mL tubes and washed (BTPBB, 2 x 400 µL; 1X T4LB, 1 x 400 µL). Biotin-HDNA magnetic resin (25 µg) was aliquoted into a 1.5-mL tube and washed (BTPWB, 2 x 100 µL; 1X T4LB, 1 x 1.4 mL). An enzymatic oligonucleotide ligation reaction, consisting of 5'-/5Phos/ GCCGCCCAGTCCTGCTCGCTTCGCTACATGGACAAAGAGCCGACGACGACTTCCCCGCGGTCTAAACCTCAA -3' (924 pmol), 5'-/5Phos/ AGGCTTGAGGTTTAGACCGCGGGGAAGTCGTCGTCGGCTCTTTGTCCATGTAGCGAAGCGAGCAGGACTG GGCGGCGG-3' (924 pmol), 5'-CACTGAGCATATCCCTGATACCG-3' (924 pmol), 5'-/5phos/ GCCTCGGTATCAGGGATATGCTCAGTG-3' (924 pmol), and T4 DNA ligase (1,200 U) was assembled in 1X

T4LB (185 µL) and incubated (20 min, RT). A second aliquot of T4 DNA Ligase (1,200 U) was added to the ligation mixture and the mixture was aliquoted to HDNA magnetic resin (55 µL) and biotin-HDNA magnetic resin (15 µL). Resin samples were incubated with rotation (15 h, RT, 8 rpm), washed (BTPBB , 1 x 500 µL), resuspended (BTPBB, 1 mL), incubated with rotation (1 h, 50 ºC, 13 rpm), then washed (BTPBB, 1 x 500 µL; BTPWB, 3 x 500 µL).

qPCR analysis. qPCR matrix contained *Taq* DNA Polymerase (0.05 U/µL,), oligonucleotide primers 5'-CACTGAGCATATCCCTGATACCG-3' and 5'-CCTGCTCGCTTCGCTACATGGACAAAG-3' (0.5 µM each), 5'-/5Cy5/CCG ACG ACG ACT TCC CCG CG/3BHQ_2/-3' 5'-exonuclease assay probe (0.35 µM), and PCR buffer (1X). Magnetic resin samples in BTPWB (~100, 1,000 and 5,000 particles/µL) were quantitated using a hemocytometer. Aliquots (1 µL) were added to separate amplification reactions (20 µL) with technical replicates (N = 3). Biotin-HDNA magnetic resin (1 µL, 25 pg) was added as a positive control amplification reaction (20 µL). Template standard solutions (1 µL, 500 amol, 50 amol, 5 amol, 500 zmol, 50 zmol, and 5 zmol in DI H_2O) were added to separate amplification reactions (20 µL, N = 3). Reactions were thermally cycled (95 ºC, 20 s; [65.8 ºC, 20s; 68 ºC, 15 s] x 26 cycles ; C1000 Touch Thermal Cycler, Bio-Rad, Hercules, CA) with fluorescence monitoring (channel 4, CFX96 Real-Time System, Bio-Rad). Samples were quantitated (CFX Manager, Version 3.1, Bio-Rad, baseline subtracted). The number of amplifiable tags per bead was calculated by dividing the qPCR result by the counted number of beads per µL. Replicates were averaged. The % DNA remaining was calculated by comparing to qPCR quantitation of untreated sensor beads. Amplification product was confirmed by agarose gel electrophoresis (3% agarose in 1X TAE, 150 V, 45 min).

Linker modifications.

Linker S2. Linker resin **S1** (30 mg) was transferred to a clean fritted syringe. Linker synthesis proceeded via iterative cycles of solid-phase peptide synthesis. Each cycle included (1) 9-fluorenylmethoxycarbonyl (Fmoc) deprotection (20% piperidine in DMF, 2 x 1 mL, 7 min each aliquot, RT, 8 rpm); (2) N-α-Fmoc-amino acid (65 μmol) activation with DIC/HOAt (90 μmol/65 μmol) and incubation (5 min, RT); (3) N-α-Fmoc-amino acid coupling to resin by transferring activated acid (1.61 mL) to resin and incubating (1 h, 50 °C, 13 rpm). After each deprotection and monomer coupling step, reactants were expelled and the resin washed (DMF, 3 x 2 mL; DCM, 3 x 2 mL; DMF, 3 x 2 mL). The following N-α-Fmoc-amino acid couplings were performed in order: (1) Fmoc-Glu(OAll)-OH ; (2) Fmoc-NH-$(PEG)_2$-CH_2CH_2COOH; (3) Fmoc-Lys(Mtt)-OH. Resin was then Fmoc deprotected (20% piperidine in DMF, 2 x 1 mL, 7 min each aliquot, RT), capped with acetic anhydride (20% in DMF (v/v), 15 min, RT, 8 rpm), and washed (DMF, 3 x 2 mL; DCM, 3 x 2 mL; DMF, 3 x 2 mL).

Linker S3. Linker resin **S1** (10 mg) was transferred to a clean fritted syringe. Fmoc was removed (20% piperidine in DMF, 2 x 1 mL, 7 min each aliquot, RT, 8 rpm) and the resin washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). 2-Azidoacetic acid (43 μmol) was pre-activated with DIC/HOAt (61 μmol/43 μmol) in DMF (1.075 mL, 5 min, RT). Pre-activated acid was added to resin, incubated (2 h, 40 °C, 13 rpm), and washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL).

Linker S4. Linker resin **S1** (8 mg) was transferred to a clean fritted syringe. Fmoc was removed (20% piperidine in DMF, 2 x 1 mL, 7 min each aliquot, RT, 8 rpm) and the resin washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). Fmoc-Glu(OAll)-OH (34 μmol) was pre-activated with DIC/HOAt (49 μmol/ 34 μmol) in DMF (860 μL, 5 min, RT). Pre-activated acid was added to resin, incubated (1 h, 37 °C, 13 rpm), and washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL).

Linker S5. Linker resin **S1** (30 mg) was transferred to a clean fritted syringe. Linker synthesis proceeded

via iterative cycles of solid-phase peptide synthesis. Each cycle included (1) 9-fluorenylmethoxycarbonyl (Fmoc) deprotection (20% piperidine in DMF, 2 x 1 mL, 7 min each aliquot, RT, 8 rpm); (2) N-α-Fmoc-amino acid or 4-iodobenzoic acid (65 μmol) activation with DIC/HOAt (92 μmol/65 μmol) and incubation (5 min, RT); (3) N-α-Fmoc-amino acid or benzoic acid coupling to resin by transferring activated acid (1.61 mL) to resin and incubating (1 h, 50 °C, 13 rpm). After each deprotection and monomer coupling step, reactants were expelled and the resin washed (DMF, 3 x 2 mL; DCM, 3 x 2 mL; DMF, 3 x 2 mL). The following acid couplings were performed: (1) Fmoc-Leu-OH ; (2) Fmoc-Ser(TBDMS)-OH; (3) 4-iodobenzoic acid.

Linker S6. Linker resin **S1** (3 mg) was transferred to a clean fritted syringe. Fmoc was removed (20% piperidine in DMF, 2 x 1 mL, 7 min each aliquot, RT, 8 rpm) and the resin washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL).

Linker S7. Linker resin **S1** (40 mg) was transferred to a clean fritted syringe. Fmoc was removed (20% piperidine in DMF, 2 x 1 mL, 7 min each aliquot, RT, 8 rpm) and the resin washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). Bromoacetic acid (1 mmol) and DIC (0.5 mmol) were combined and incubated in DMF (1 mL, 2 min, RT). Pre-activated acid was added to syringe, incubated (20 min, 37 °C, 13 rpm), and washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). Benzylamine (1 mmol in DMF) was added to syringe, incubated (2 h, 50 °C, 13 rpm), and washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL).

Linker S8. Linker resin **S1** (30 mg) was transferred to a clean fritted syringe. Fmoc was removed (20% piperidine in DMF, 2 x 1 mL, 7 min each aliquot, RT, 8 rpm) and the resin washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). 4-Iodobenzoic acid (129 μmol) was combined with DIC/HOAt (184 μmol/129 μmol) in DMF and incubated(3.23 mL, 5 min, RT). Pre-activated acid was added to resin, incubated (1 h, 50 °C, 13 rpm), and washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL).

DNA-encoded reaction rehearsal. All reactions were performed in filtration microplate wells using the following protocol unless otherwise specified. Filtration microplates (Millipore MultiScreen Solvinert 0.45 μm Hydrophobic PTFE, EMD Millipore, Billerica, MA) were wetted using a vacuum manifold (DCM, 3 x 150 μL; DMF, 3 x 150 μL, Vac-Man® 96 Vacuum Manifold, Promega, Madison, WI). Linker resin (0.5 mg in DMF) and sensor beads (8.1 μg in DMF) were added and the solvent drained. Reagents for testing were added to wells (total volume 150 μL). Plates were covered with adhesive foil (VWR International, Radnor, PA) and incubated with agitation (800 rpm). Following incubation, mixtures were drained and resin was washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). Resin was resuspended and transferred by pipette to 1.5-mL tubes (DMF, 3 x 150 μL). Sensor beads were magnetically separated (Magna-Sep Magnetic Particle Separator, Life Technologies, Grand Island, NY), and linker resin was transferred to a clean fritted spin column (Mobicol Classic, MoBiTec GmbH, Goettingen, Germany), washed (DCM, 3 x 0.5 mL), and dried *in vacuo*. Linker cleavage and characterization were performed as described. Sensor beads were washed (BTPBB, 125 μL), resuspended (BTPBB, 500 μL), incubated with rotation (16 h, RT, 13 rpm), washed (BTPBB, 1 x 125 μL; DI H_2O, 3 x 125 μL, BTPWB, 2 x 125 μL), then resuspended for qPCR analysis (BTPWB, 120 μL) as described.

Fmoc deprotection. Piperidine (20% v/v in DMF, 150 μL) was added to resin **S1** and incubated (7 min, RT). The mixture was drained and a fresh aliquot was added and incubated as above. Linker resin and magnetic sensor beads were separated and washed as described

Mtt deprotection. Linker resin **S2** (0.5 mg in DCM) and magnetic sensor beads (8.1 μg in DCM) were added to 1.5-mL tubes and supernatant removed. TFA (1% v/v in DCM, 150 μL) was added and the mixture was incubated with rotation (15 min, RT, 8 rpm). Supernatant was removed and resin washed (DCM, 3 x 150 μL; DMF, 3 x 150 μL). Linker resin and magnetic sensor beads were separated and washed as described.

Staudinger reduction. Linker resin **S3** (0.5 mg in DCM), and magnetic sensor beads (8.1 μg in DCM) were added to 1.5-mL tubes and supernatant removed. TCEP HCl (100 mM in BTPWB, pH 7.6) was added (150 μL) and the mixture was incubated with rotation (16 h, 50 °C, 13 rpm). The mixture was drained and the resin was washed (BTPWB, 3 x 150 μL). Resin was resuspended and transferred to 1.5 mL-tubes (BTPWB, 3 x 150 μL). Linker resin and magnetic sensor beads were separated and washed as described.

O-Allyl deprotection. Linker resin **S4** (0.5 mg in DCM) and magnetic sensor beads (8.1 μg in DCM) were added to 1.5-mL tubes and supernatant removed. Phenylsilane (122 μmol in DCM, 100 μL) was added followed by $Pd(PPh_3)_4$ (8.7 μmol in DCM, 100 μL). The suspension was vortexed and incubated with rotation (30 min, RT, 8 rpm). Supernatant was removed and resin was washed (DCM, 3 x 150 μL; DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). Linker resin and magnetic sensor beads were separated and washed as described.

TBDMS deprotection. Linker resin **S5** (0.5 mg in DCM), and magnetic sensor beads (8.1 μg in DCM) were added to 1.5-mL tubes and supernatant removed. TEA・3HF (920 μmol, 150 μL) was added, vortexed, and incubated with rotation (16 h, RT, 8 rpm).[4] Supernatant was removed and resin was washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). Sensor beads were magnetically separated, and linker resin was transferred to a clean fritted spin columns. Magnetic sensor beads were washed as described. Linker resin was acetylated (20% v/v acetic anhydride in DMF, 300 μL, 1 h, RT, 8 rpm), washed (DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL; DCM, 3 x 150 μL), and dried *in vacuo*.

Amino acid coupling to primary amine. Fmoc-Pro-OH (6 μmol), DIC (8.55 μmol), and HOAt (6 μmol) were combined in DMF (150 μL), incubated (5 min, RT), added to resin **S6** (0.5 mg), and incubated (1 h, 37 °C).

Amino acid coupling to secondary amine. Fmoc-Ser(tBu)-OH (12 μmol), DIC (15 μmol), OxymaPure(12 μmol), and TMP (12 μmol) were combined in DMF (150 μL), incubated (5 min, RT), added to

resin **S7** (0.5 mg), and incubated (3 h, 37 °C).

Haloacid coupling. Chloroacetic acid or bromoacetic acid (6 μmol), DIC (8.55 μmol), and HOAt (6 μmol) were combined in DMF (150 μL), incubated (5 min, RT), added to resin **S6** (0.5 mg), and incubated (1 h, 37 °C).

(R/S)-5-chloro-2,4-dimethyl-3-pentenoic acid coupling. R- or S-5-chloro-2,4-dimethyl-3-pentenoic acid was prepared and characterized as described.[2] Chloroacid monomer (6 μmol), DIC (8.55 μmol), and HOAt (6 μmol) were combined in DMF (150 μL), incubated (5 min, RT), added to resin **S6** (0.5 mg), and incubated (1 h, 37 °C).

Nucleophilic displacement. Benzylamine (1 M in DMF, 150 μL) was added to resin **S7** (0.5 mg) and incubated (1 h, 37 °C).

Acetylation. Acetic anhydride (20% v/v in DMF, 150 μL) was added to resin **S6** (0.5 mg) and incubated (15 min, RT).

Nucleophilic addition of isocyanates. 4-(chloromethyl)phenyl isocyanate (40 mM in DMF, 150 μL) was added to resin **S6** (0.5 mg) and incubated (15 min, RT).

Aldehyde reductive amination. Linker resin **S7** (0.5 mg) and magnetic sensor beads (8.1 μg in DMF) were added to 1.5-mL tubes and supernatant removed. 4-Iodobenzaldehyde (75 μmol in 1% v/v acetic acid in DMF, 150 μL) was added to resin, vortexed, and incubated with rotation (10 min, RT, 8 rpm). The suspension was centrifuged, and sodium cyanoborohydride (75 μmol in 1% v/v acetic acid in MeOH, 150 μL) was added, the suspension vortexed, and incubated with rotation (1 h, RT, 8 rpm).[5] Supernatant was removed and resin washed (MeOH, 150 μL; DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). Linker resin and magnetic sensor resin were separated and washed as described.

Suzuki-Miyaura cross-coupling. Linker resin **S8** (0.5 mg in NMP), and magnetic sensor resin (8.1 μg in NMP) were added to 1.5-mL Safe-Lock tubes (Eppendorf North America, Inc., Hauppauge, NY) and

supernatant removed. NMP and all stock solutions were sparged (Ar, 1 min) prior to use. 4-Isopropylphenylboronic acid (172 μmol), DIEA (344 μmol), and $Pd(PPh_3)_4$ (64.5 nmol) were combined in NMP (500 μL), added to resin, sparged (Ar, 1 min), vortexed, and incubated with rotation (7 h, 70 °C, 13 rpm). Supernatant was removed and resin washed (NMP, 3 x 150 μL; DMF, 3 x 150 μL; DCM, 3 x 150 μL; DMF, 3 x 150 μL). Linker resin and magnetic sensor beads were separated and washed as described.

Macrocyclization. Following Mtt and O-Allyl deprotection, PyAOP (7 μmol), HOAt (14 μmol), and DIEA (21 μmol) were combined in DMF (150 μL), added to resin **S2** (0.5 mg), and incubated (3 h, 37 °C).

Solution-Phase Reaction rehearsal

Solution-phase oligonucleotide characterization. Oligonucleotides were resuspended (DI H_2O, 50 μL) and analyzed by reversed-phase HPLC (XTerra MS C18, 4.6 x 50 mm, 2.5 μm, Waters) with gradient elution (mobile phase A: 5% ACN in 0.1 M TEAA pH 6.0; mobile phase B: 25% ACN in 0.1 M TEAA pH 6.0; mobile phase C: ACN; 0-100% B, 1-20 min; 100-20% B, 0-80% C, 20-28 min). HPLC fraction aliquots (1 μL) were spotted to a MALDI-TOF MS target plate, dried, covered with THAP matrix solution (18 mg/mL THAP, 7 mg/mL ammonium citrate dibasic in 1:1 ACN:0.1% TFA in H_2O) and analyzed via MALDI-TOF MS (Microflex, Bruker Daltonics, Inc.).

Amino acid coupling.[3] 5'-aminohexyl-modified oligonucleotide **S34** (5'-/5AmMC6/ACTGGCATT-3', 50 nmol) was aliquoted into 1.5-mL tubes and dried *in vacu*o. Magnetic sensor beads (8.1 μg), Fmoc-Lys(Alloc)-OH (1.88 μmol in DMF, 12.6 μL), DMT-MM (1.9 μmol in DI H_2O, 7.6 uL) and buffer (150BWB, 50 μL) were combined, vortexed, and incubated with rotation (18 h, 4 °C, 13 rpm). Sensor beads were magnetically separated, and oligonucleotide-containing supernatant transferred to a clean 1.5-mL tube. Oligonucleotides were precipitated in ethanol, centrifuged (1 min, 10,000 rcf), and supernatant was

removed. The pellet was resuspended (DI H_2O, 100 µL), precipitated in ethanol, pelleted (1 min, 10,000 rcf), supernatant was removed, the pellet washed (80% EtOH in DI H_2O, 3 x 100 µL), and air dried. Oligonucleotide-Lys(Alloc)-Fmoc conjugate was used for further reactions without purification. Sensor beads were washed (150 BWB, 3 x 150 µL), resuspended (BTPBB, 500 µL), incubated with rotation (16 h, RT, 13 rpm), washed (BTPBB, 125 µL; DI H_2O, 3 x 125 µL, BTPWB, 2 x 125 µL), then resuspended (BTPWB, 120 µL) for qPCR analysis.

Fmoc deprotection.[3] Magnetic sensor beads (8.1 µg in diH_2O, 67.5 µL) were added to oligonucleotide-Lys(Alloc)-Fmoc conjugate **S35** (50 nmol) in 1.5-mL tubes. Piperidine (20% in DI H_2O, 67.5 µL) was added, and the reaction was vortexed, then incubated with rotation (4 h, RT, 8 rpm). Sensor beads were magnetically separated, and oligonucleotide-containing supernatant transferred to a clean 1.5-mL tube. Oligonucleotides were precipitated in ethanol and centrifuged (2 min, 7,600 rpm). The supernatant was removed, the pellet was washed (80% EtOH in DI H_2O, 3 x 2 mL), and air dried. Sensor beads were washed (BTPBB, 3 x 150 µL), resuspended (BTPBB, 500 µL), incubated with rotation (16 h, RT, 13 rpm), washed (BTPBB, 1 x 125 µL; DI H_2O, 3 x 125 µL, BTPWB, 2 x 125 µL), then resuspended (BTPWB, 120 µL) for qPCR analysis.

Alloc deprotection.[6] Magnetic sensor beads (8.1 µg in 250 BWB, 35 µL) were added to oligonucleotide-Lys(Alloc)-Fmoc conjugate **S35** (50 nmol) in 1.5-mL tubes. Buffer (250BWB, 15 µL), $NaBH_4$ (2.0 µmol in ACN, 10 µL) and $Pd(PPh_3)_4$ (0.10 µmol in DMA, 10 µL) were added, the reaction was vortexed, then incubated with rotation (2 h, RT, 8 rpm). Oligonucleotides were precipitated in ethanol, centrifuged (1 min, 10,000 rcf), and supernatant was removed. The pellet was resuspended (DI H_2O, 100 µL, 5 min, 60 °C), sensor beads were magnetically separated, and oligonucleotide-containing supernatant was transferred to a

clean 1.5-mL tube. Oligonucleotides were precipitated in ethanol and centrifuged (1 min, 10,000 rcf). The supernatant was removed, the pellet was washed (80% EtOH in DI H_2O, 3 x 100 µL; DCM, 1 x 100 µL, 80% EtOH in DI H_2O, 1 x 100 µL), and air dried. Sensor beads were washed (BTPBBE, 2 x 150 µL), resuspended (BTPBBE, 500 µL), incubated with rotation (16 h, RT, 13 rpm), washed (BTPBB, 1 x 125 µL; DI H_2O, 3 x 125 µL, BTPWB, 2 x 125 µL), then resuspended (BTPWB, 120 µL) for qPCR analysis.

Boc deprotection.[7] Magnetic sensor beads (8.1 µg) were aliquoted to 1.5-mL boil-proof tubes and supernatant removed. NaOAc (750 nmol in DI H_2O, 37.5 µL) and $MgCl_2$ (5 µmol in diH_2O, 5 µL) were added, and the reaction was vortexed, then incubated with rotation (16 h, 70 °C, 13 rpm). Supernatant was removed, and sensor beads were washed (BTPBB, 2 x 150 µL), resuspended (BTPBB, 500 µL), incubated with rotation (16 h, RT, 13 rpm), washed (BTPBB, 1 x 125 µL; DI H_2O, 3 x 125 µL, BTPWB, 2 x 125 µL), then resuspended (BTPWB, 120 µL) for qPCR analysis.

Boc deprotection.[6] Magnetic sensor beads (8.1 µg) were aliquoted to 1.5-mL boil-proof tubes and supernatant removed. Buffer (250BWB, 50 µL) was added, and the reaction was vortexed, then incubated with shaking (16 h, 70 °C, 1,000 rpm). Supernatant was removed, and sensor beads were washed (BTPBB, 2 x 150 µL), resuspended (BTPBB, 500 µL), incubated with rotation (16 h, RT, 13 rpm), washed (BTPBB, 1 x 125 µL; DI H_2O, 3 x 125 µL, BTPWB, 2 x 125 µL), then resuspended (BTPWB, 120 µL) for qPCR analysis.

Quinazolinone formation.[6] Magnetic sensor resin (8.1 µg) was aliquoted to boil-proof 1.5-mL tubes and supernatant removed. The sensor beads were combined with buffer (250BWB, 50 µL) and NaOH (25 µmol in DI H_2O, 25 µL), vortexed, and incubated with shaking (16 h, 70 °C, 1,000 rpm). Supernatant was removed, and sensor beads were washed (BTPBB, 2 x 150 µL), resuspended (BTPBB, 500 µL), incubated

with rotation (16 h, RT, 13 rpm), washed (BTPBB, 1 x 125 µL; DI H_2O, 3 x 125 µL, BTPWB, 2 x 125 µL), then resuspended (BTPWB, 120 µL) for qPCR analysis.

Suzuki-Miyaura cross-coupling.[8] 5'-aminohexyl-modified oligonucleotide **S34** (5'-/5AmMC6/ ACTGGCATT-3', 50 nmol) was aliquoted into 1.5-mL tubes and dried *in vacuo*. Buffer (250BWB, 17.5 µL), 4-iodobenzoic acid (200 nmol in DMF, 3.5 µL) and DMT-MM (200 nmol in DI H_2O, 3.5 µL) were added, chilled on ice (10 min), and incubated with rotation (17 h, RT, 8 rpm). Oligonucleotides were precipitated, centrifuged (1 min, 13,000 rcf), and supernatant removed. The pellet was washed (80% EtOH in DI H_2O, 2 x 100 µL), resuspended (DI H_2O, 50 µL), precipitated in ethanol, centrifuged (1 min, 13,000 rcf), supernatant removed, and air dried. Oligonucleotide-conjugate **S39** was resuspended (0.04% Tween-20 in DI H_2O), quantitated, and transferred (20 nmol) to a 1.5-mL boil-proof tube containing magnetic sensor beads (8.1 µg in 0.04% Tween-20, 20 µL). 3,4-(Methylenedioxy)phenylboronic acid (400 nmol in 1:1 ACN/ H_2O, 1.0 µL), Na_2CO_3 (800 nmol in H_2O, 1.25 µL), and $Pd(PPh_3)_4$ (20 nmol in 1:2:2 DCM/ACN/toluene, 1.0 µL) were added, and the reaction was vortexed, then incubated with shaking (1.5 h, 80 °C, 1,000 rpm). Sensor beads were magnetically separated, and oligonucleotide-containing supernatant transferred to a fresh 1.5-mL tube. Oligonucleotides were precipitated in ethanol, centrifuged (1 min, 13000 rcf), and supernatant removed. The pellet was washed (80% EtOH in DI H_2O, 100 µL) and air dried. Sensor beads were washed (1:2:2 DCM/ACN/toluene, 125 µL; 0.04% Tween-20, 125 µL; BTPBBE, 125 µL), resuspended (BTPBBE, 500 µL), incubated with rotation (16 h, RT, 13 rpm), washed (BTPBB, 125 µL; DI H_2O, 3 x 125 µL, BTPWB, 2 x 125 µL), then resuspended (BTPWB, 120 µL) for qPCR analysis.

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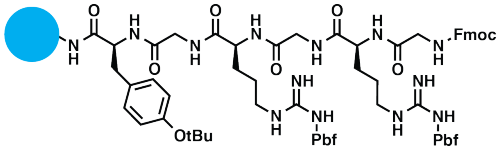
Supporting Information Figures

Figure S1. Synthesis resin linker modifications. S1 was modified as described to investigate Mtt-deprotection using **S2**, O-allyl deprotection using **S2** and **S4**, macrocyclization using **S2**, Staudinger reduction using **S3**, TBDMS-deprotection using **S5**, amide bond forming reactions using **S6** and **S7**, urea-bond forming using **S6**, reductive amination using **S7**, and Suzuki-Miyaura cross-coupling using **S8**.

S1



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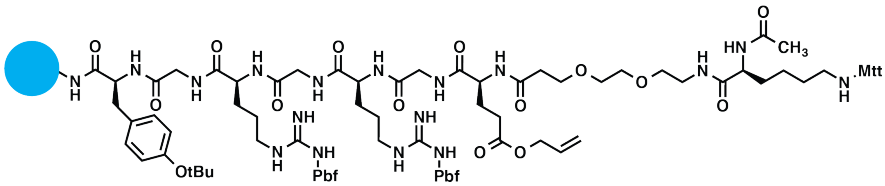

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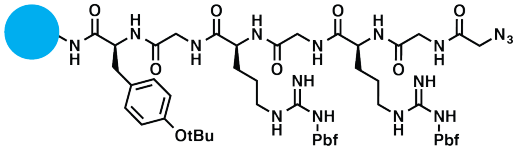

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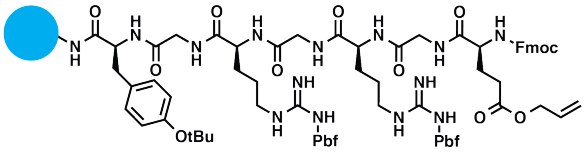

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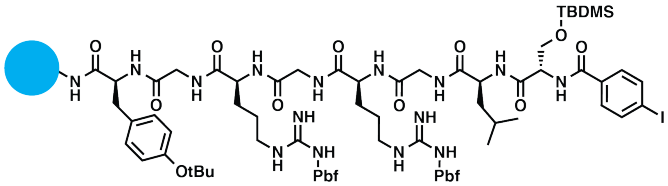
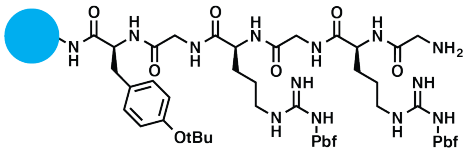
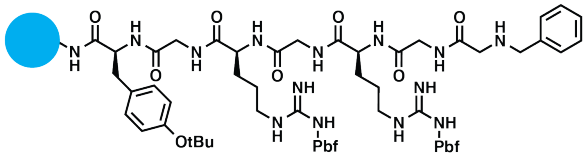


S6



S7



S8

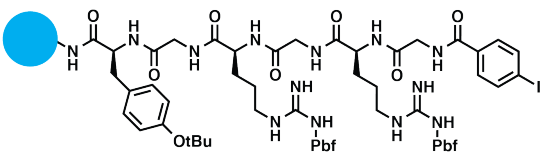


Figure S2. Preparation of azido-HDNA. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of NH_2-HDNA **S9** starting material, and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of N_3-HDNA **S10** support correct conversion to the expected azido product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

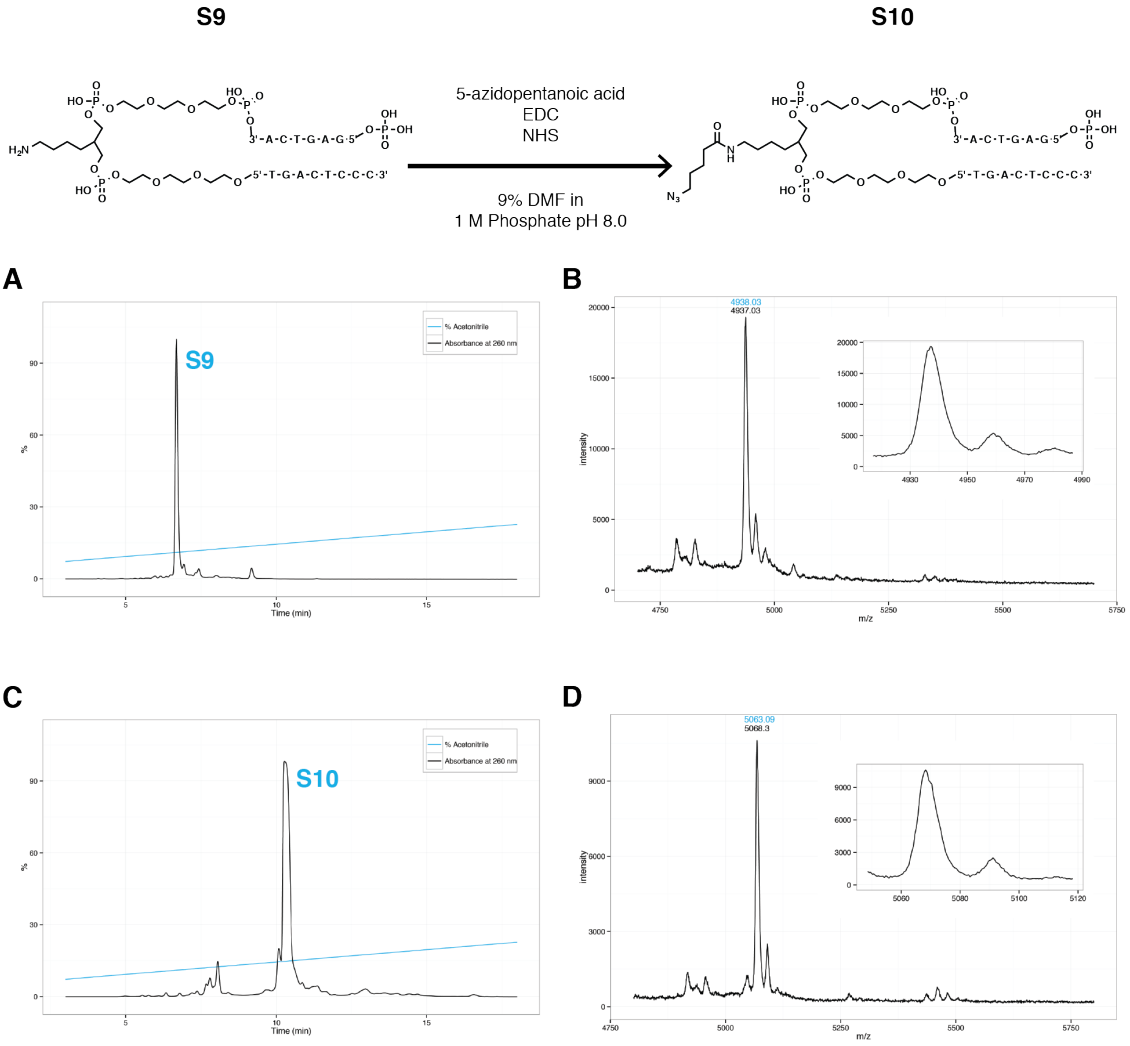


Figure S3. Fmoc deprotection of synthesis linker S1. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of Fmoc-protected starting linker **S11** (TFA cleavage product of synthesis linker **S1**), and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of Fmoc deprotected product **S12** support correct generation of amine product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

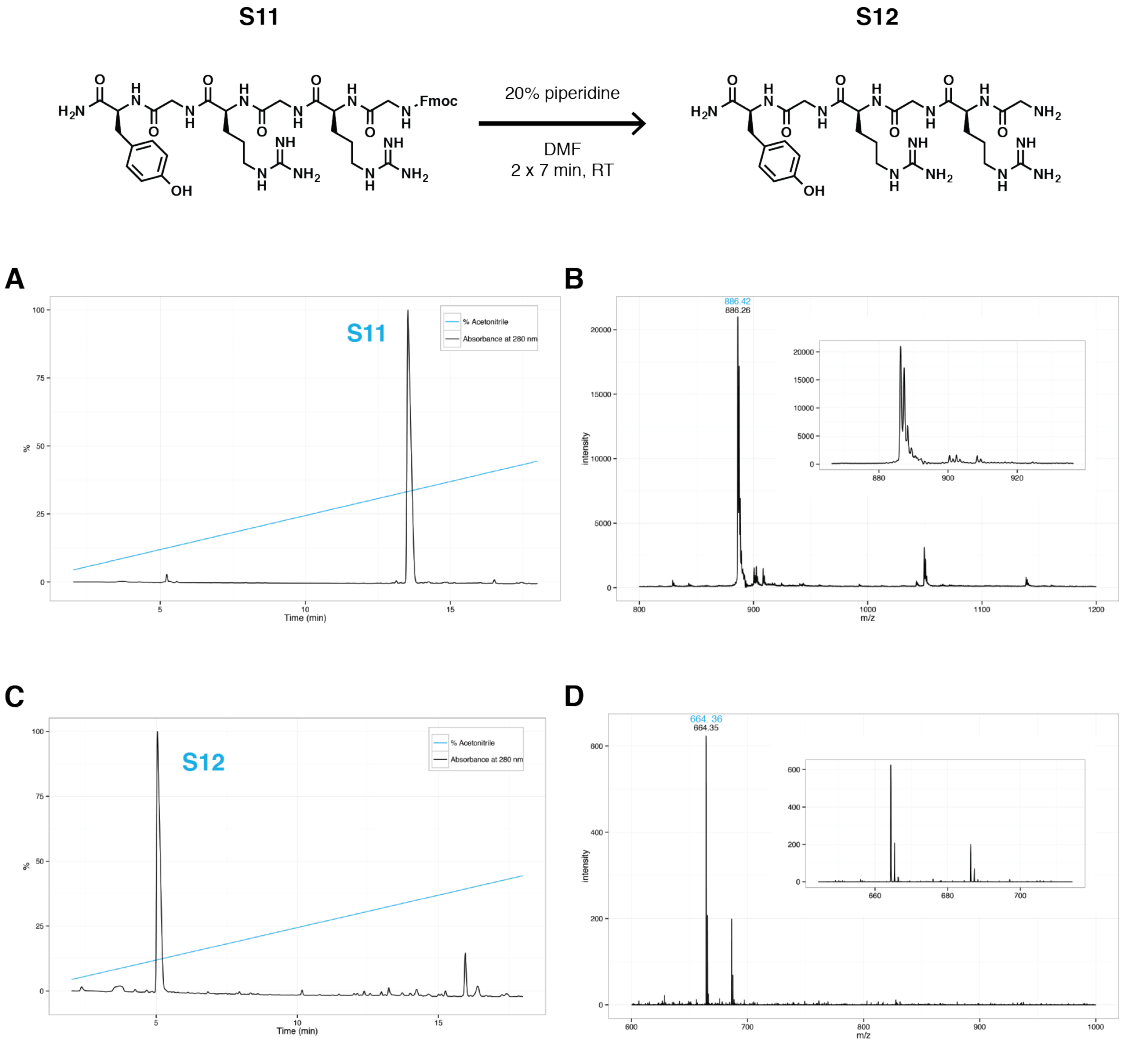
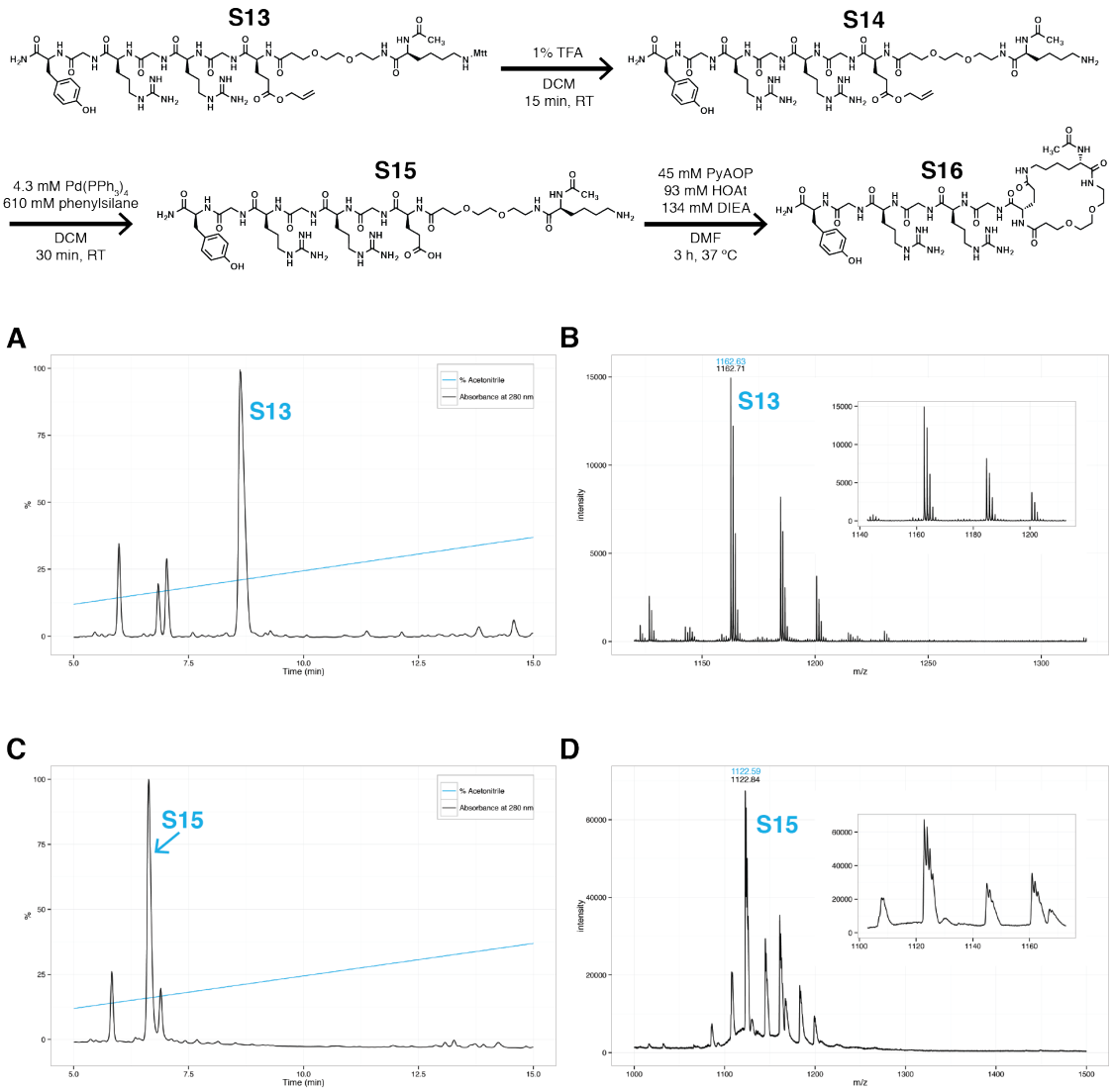
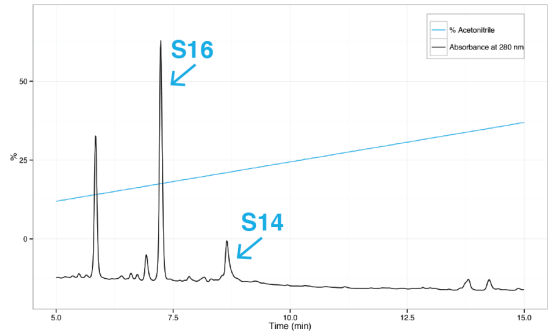


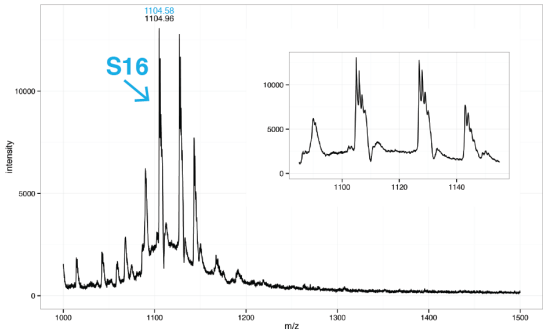
Figure S4. Mtt deprotection, O-allyl deprotection and cyclization of synthesis linker S2. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of starting linker **S13** (TFA cleavage product of synthesis linker **S2**), HPLC (**C**) and MALDI-TOF MS (**D**) analysis of Mtt- and O-Allyl-deprotected product **S15**, and HPLC (**E**) and MALDI-TOF MS analysis of cyclized product **S16** (**F**) and starting linker **S13** that did not react under Mtt or O-Allyl-deprotection conditions **S14** (**G**) support correct conversion to the expected deprotected and cyclized products as well as the presence of starting material. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.



E


% Acetonitrile
Absorbance at 280 nm
S16
S14
%
Time (min)

F


1104.58
1104.96
S16
intensity
m/z

G

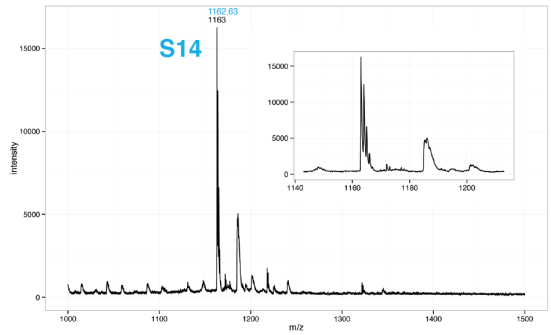

1162.63
1163
S14
intensity
m/z

Figure S5. Staudinger reduction of synthesis linker S3. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of azido-starting linker **S17** (TFA cleavage product of synthesis linker **S3**), and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of amine **S18** support correct generation of product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

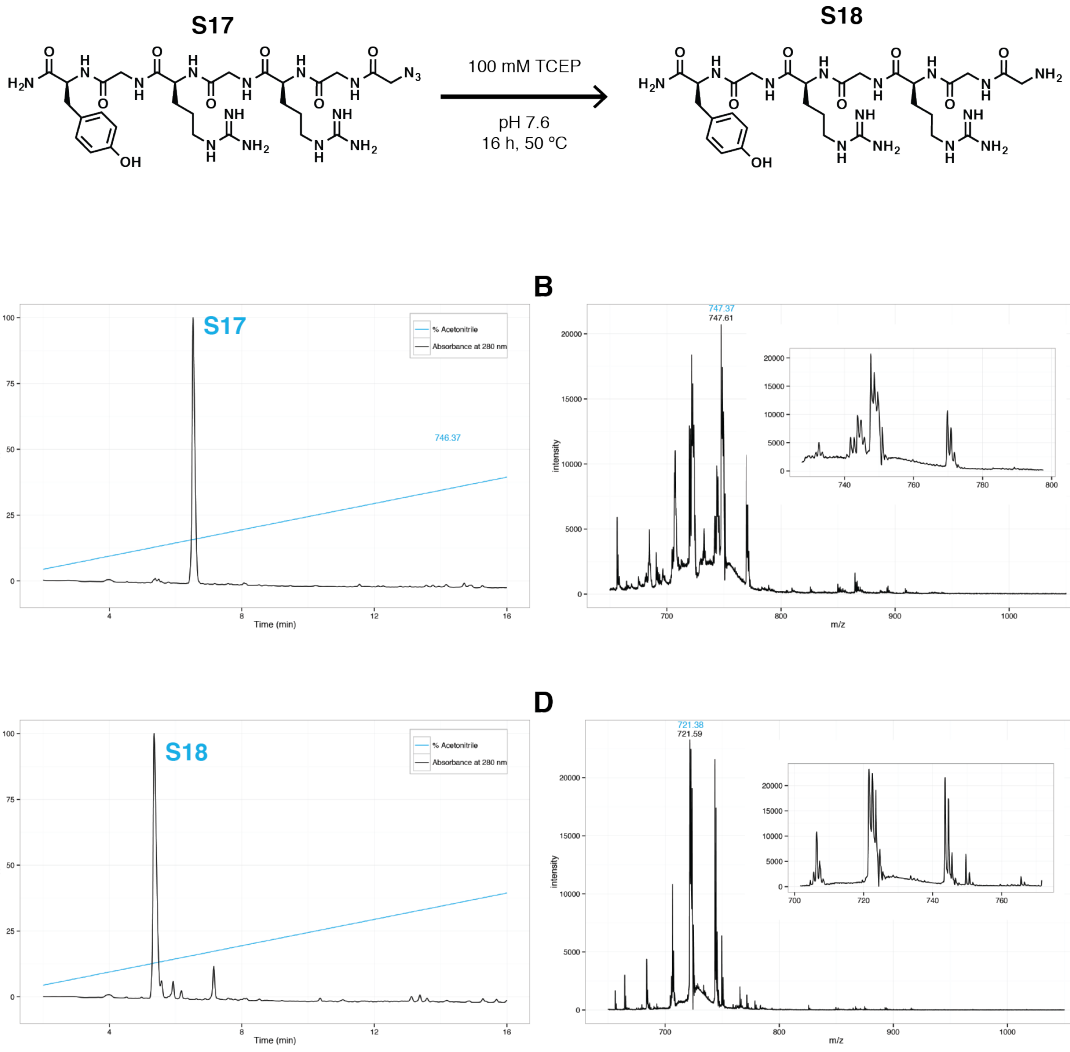


Figure S6. O-Allyl deprotection of synthesis linker S4. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of O-Allyl protected starting linker **S19** (TFA cleavage product of synthesis linker **S4**), and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of carboxylic acid **S20** support correct generation of product **S20**. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

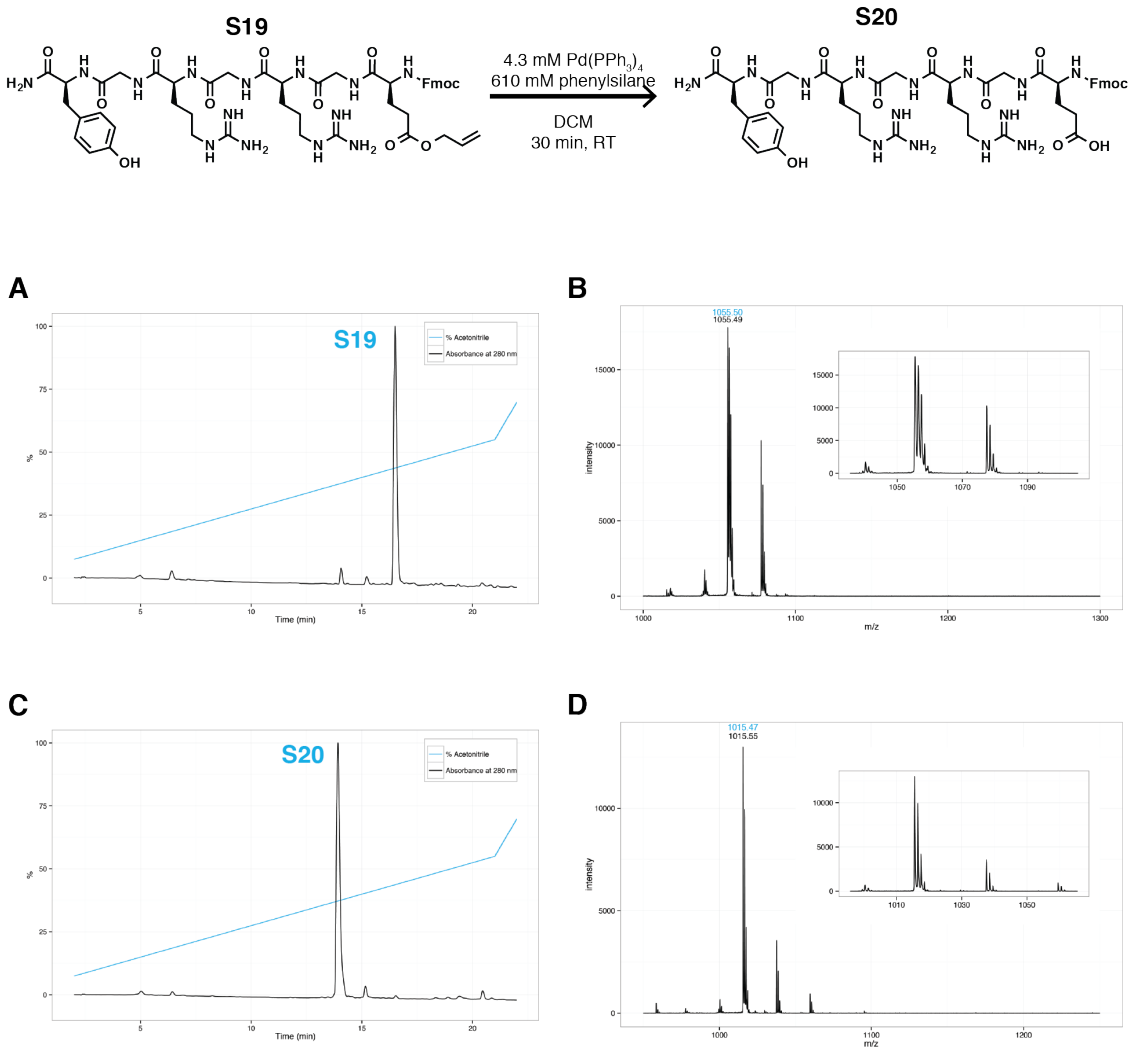


Figure S7. TBDMS deprotection and acetylation of synthesis linker S5. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of TBDMS-protected starting linker **S21** (TFA cleavage product of synthesis linker **S5**) and TFA cleavage product **S21b**, and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of TBDMS-deprotected and acetylated product **S22** support correct generation of alcohol product and subsequent acetic anhydride capping. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

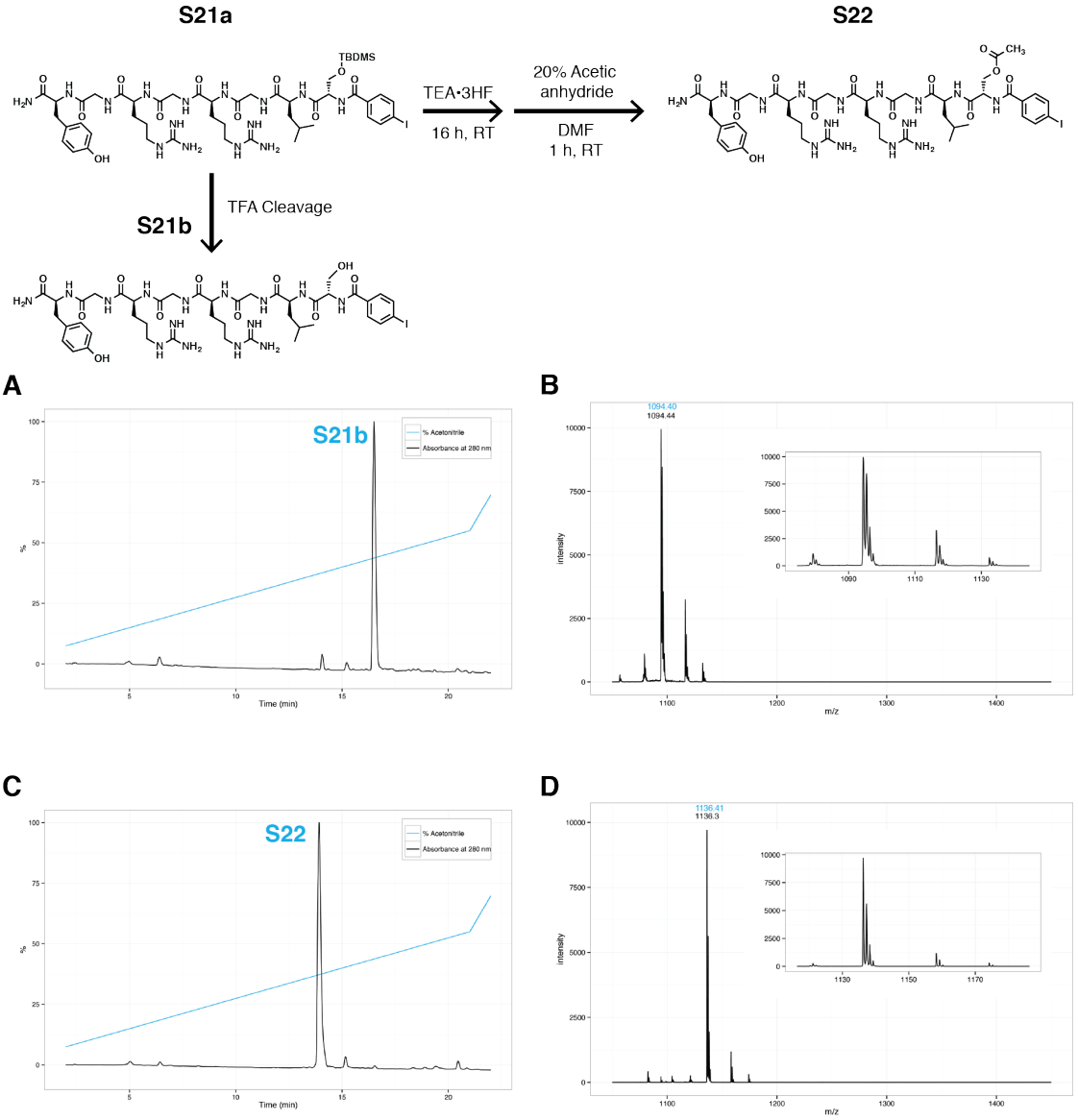


Figure S8. Amino acid coupling to primary amine linker S6. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of primary amine starting linker **S12** (TFA cleavage product of synthesis linker **S6**), and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of Fmoc-Pro-OH-coupled and Fmoc-deprotected product **S23** support correct conversion to amino acid-coupled product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

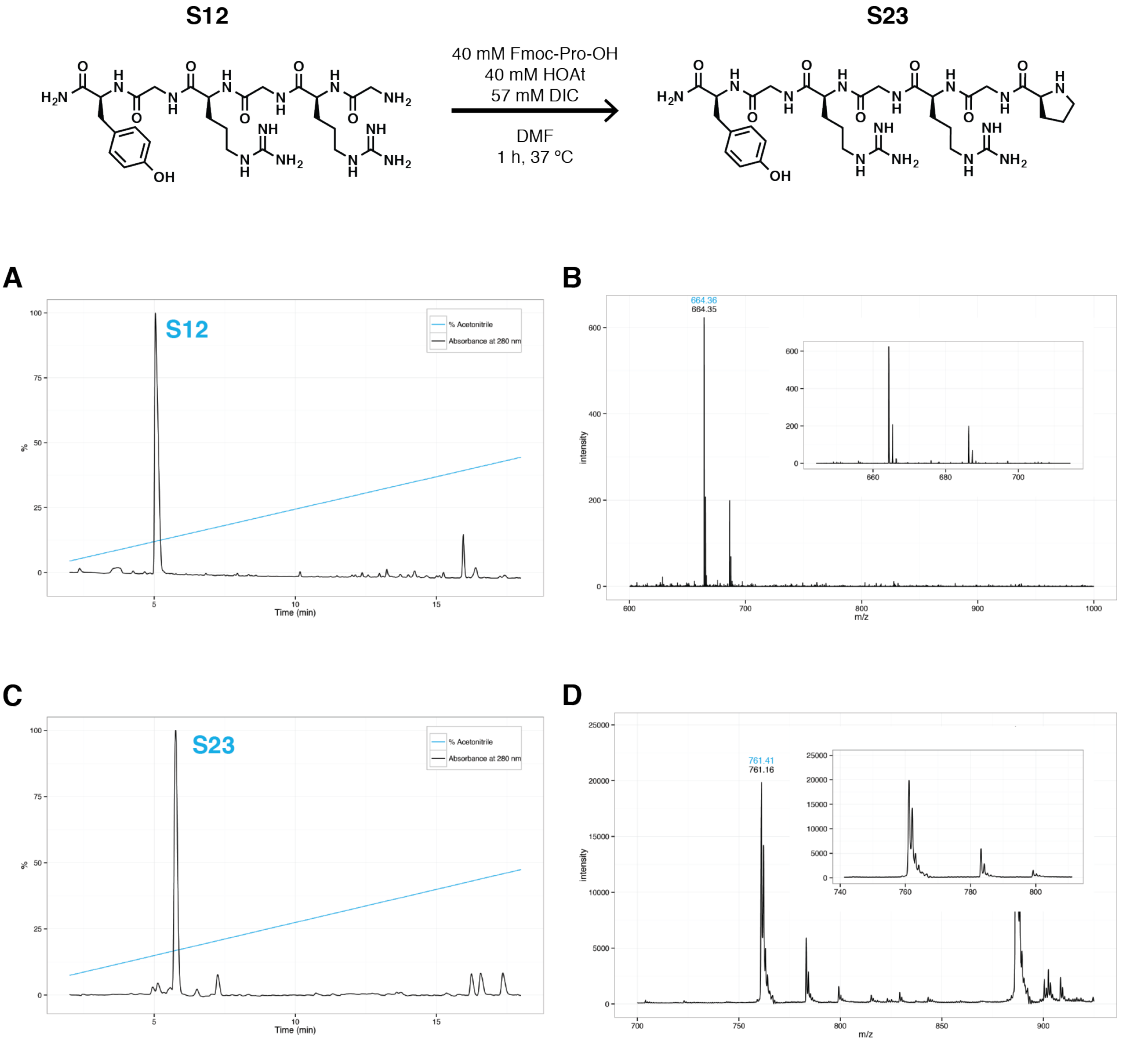


Figure S9. Amino acid coupling to secondary amine linker S7. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of secondary amine starting linker **S24** (TFA cleavage product of synthesis linker **S7**), and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of Fmoc-Ser(tBu)-OH-coupled product **S26** support correct conversion to amino acid-coupled product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

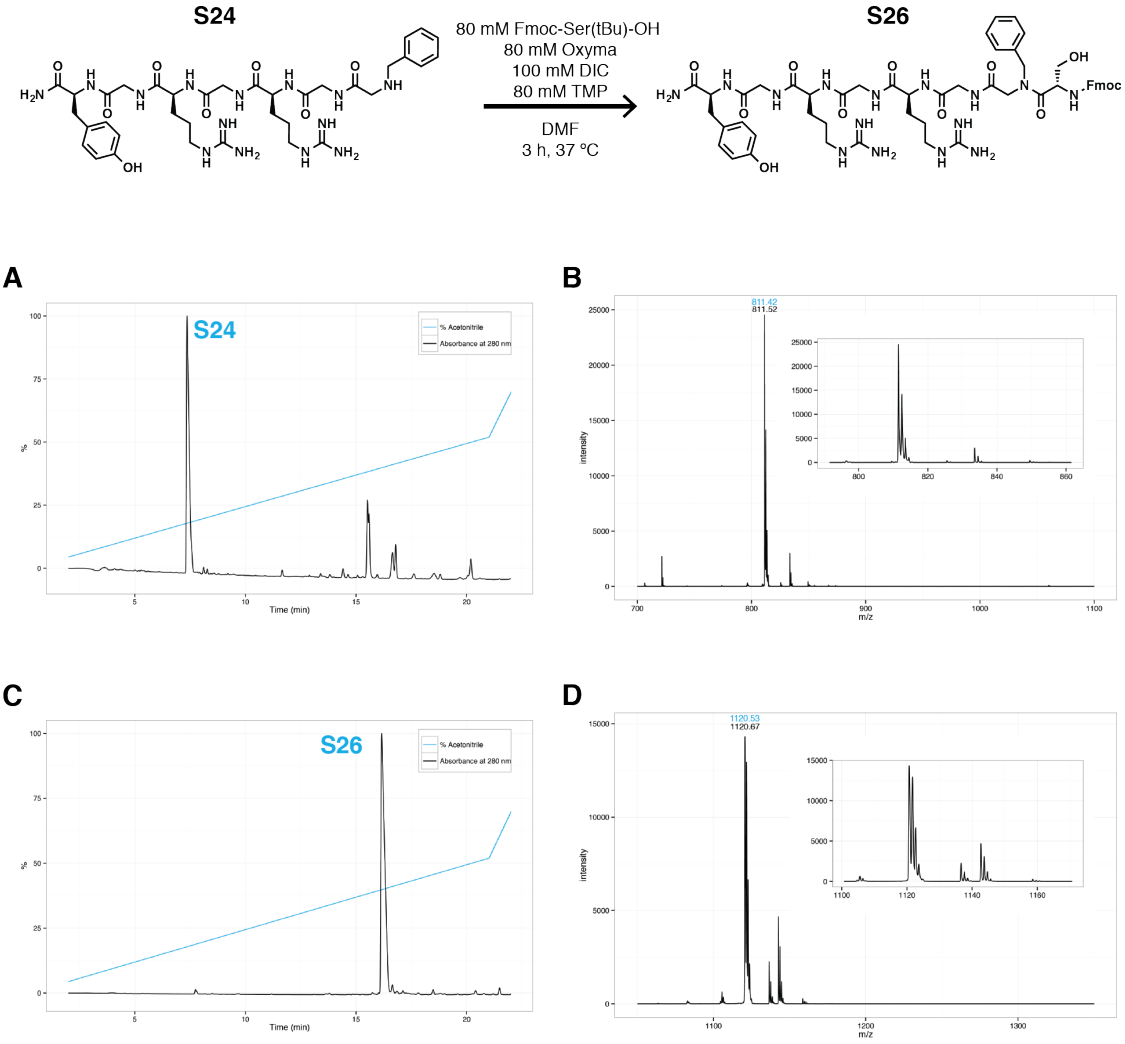


Figure S10. Haloacid coupling to starting linker S6 and nucleophilic displacement. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of primary amine starting linker **S12** (TFA cleavage product of synthesis linker **S6**), and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of chloroacetic acid coupling and nucleophilic displacement with benzylamine **S24** support correct conversion to the expected haloacid-coupled, amine-displaced product. HPLC (**E**) and MALDI-TOF MS (**F**) analysis of bromoacetic acid coupling and nucleophilic displacement with benzylamine to yield **S25** indicate hydrolysis of the haloacid to yield undesired side-product **S25**. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

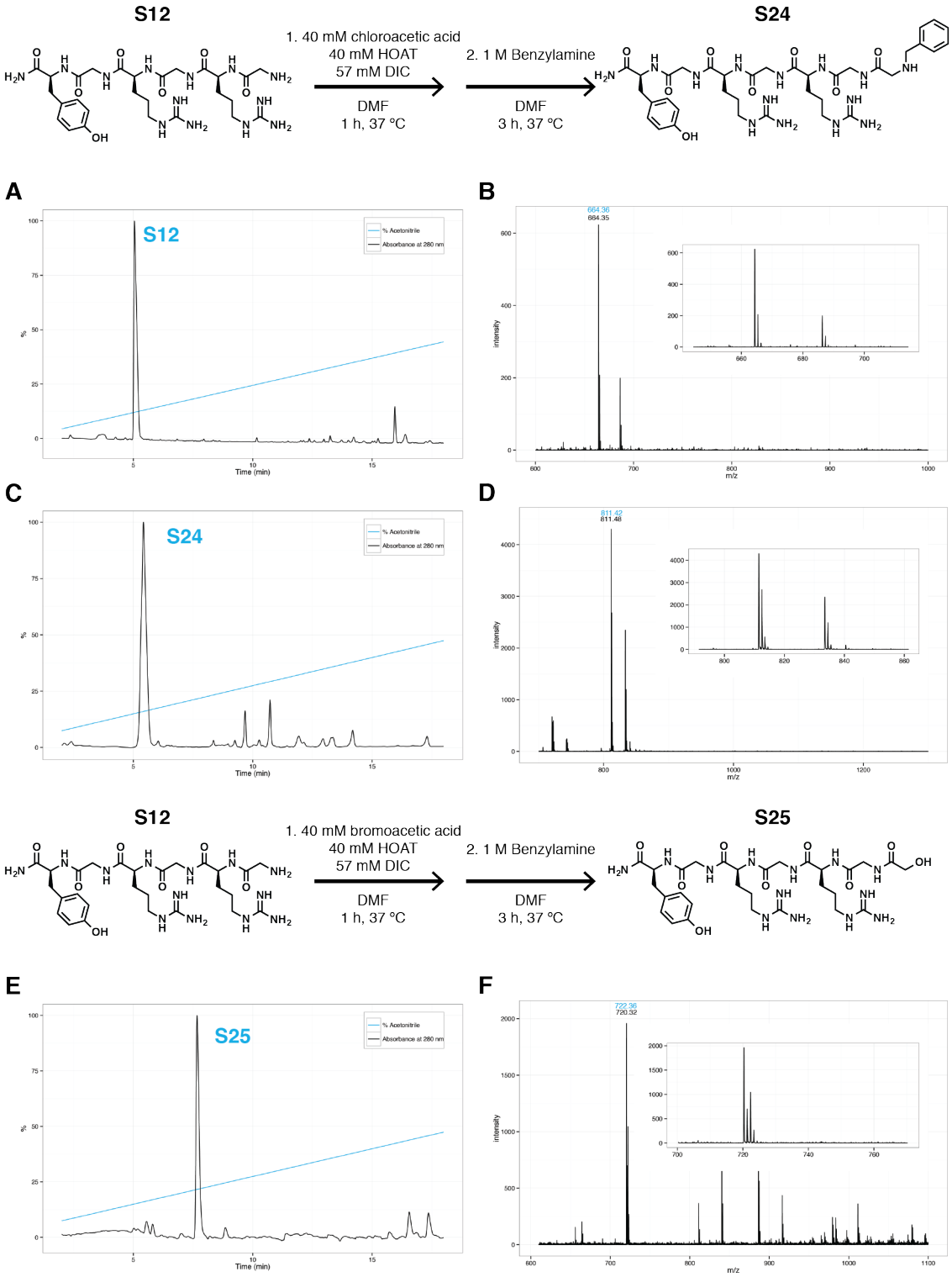


Figure S11. (R/S)-5-chloro-2,4-dimethyl-3-pentenoic acid coupling and nucleophilic displacement. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of primary amine starting linker **S12** (TFA cleavage product of synthesis linker **S6**), HPLC (**C**) and MALDI-TOF MS (**D**) analysis of R-5-chloro-2,4-dimethyl-3-pentenoic acid coupling and nucleophilic displacement with benzylamine to yield **S27**, and HPLC (**E**) and MALDI-TOF MS (**F**) analysis of S-5-chloro-2,4-dimethyl-3-pentenoic acid coupling and nucleophilic displacement with benzylamine to yield **S28** support correct haloacid coupling and amine displacement to the expected products. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

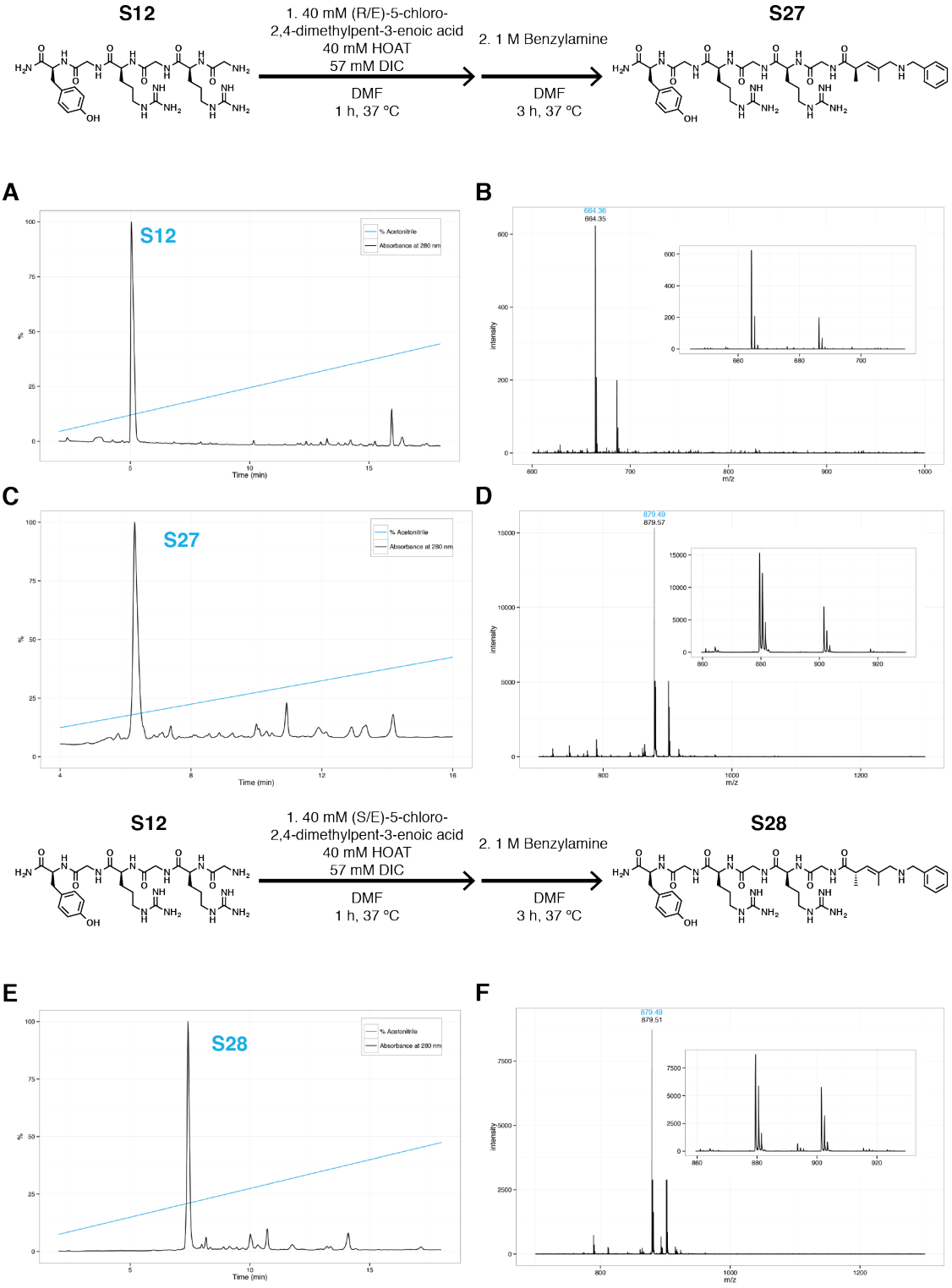


Figure S12. Acetylation of starting linker S6. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of starting linker **S12** (TFA cleavage product of synthesis linker **S6**), and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of **S29** support correct conversion to the expected acetylated product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

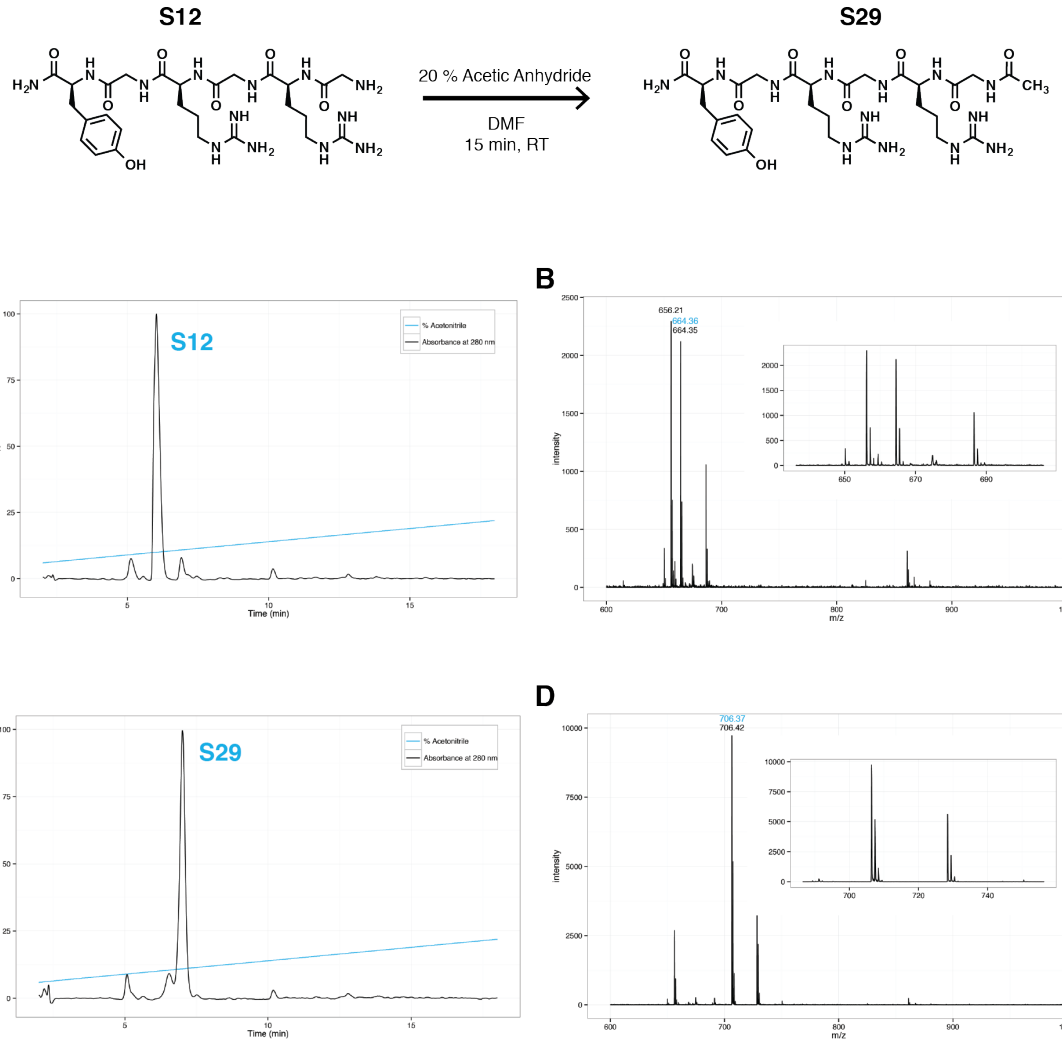


Figure S13. Nucleophilic addition of isocyanates to starting linker S6. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of the nucleophilic addition of 4-(chloromethyl)phenyl isocyanate to starting linker **S12** (TFA cleavage product of synthesis linker **S6**) followed by nucleophilic displacement with benzylamine to yield **S30** support the correct conversion to the expected isocyanate-substituted, amine-displaced product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

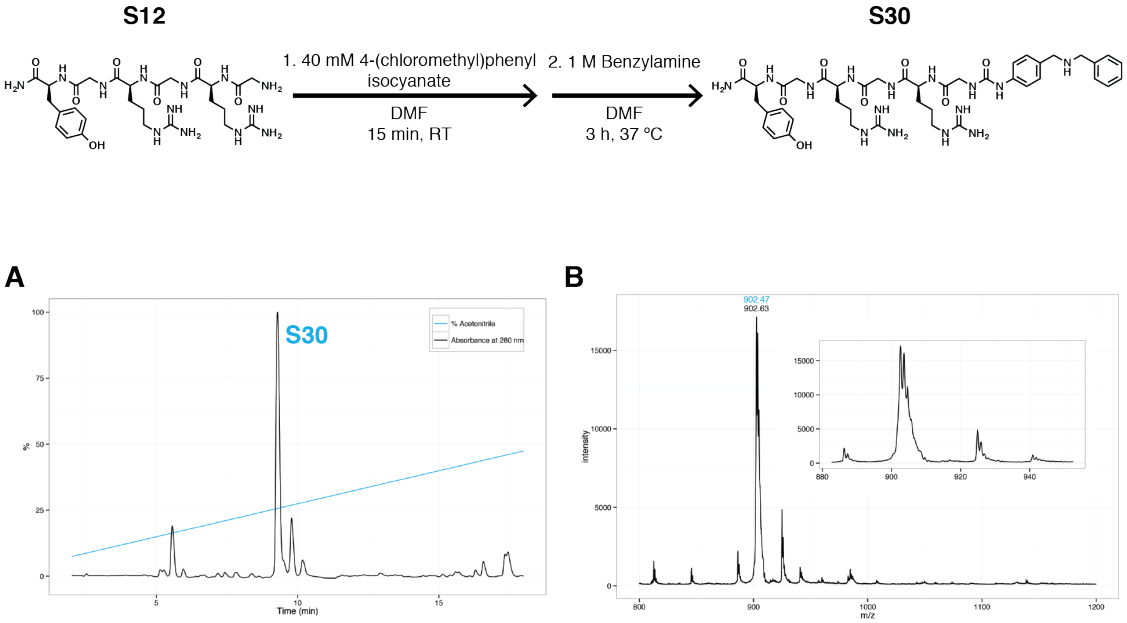


Figure S14. Reductive amination of 4-iodobenzaldehyde with secondary amine linker S7. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of secondary amine starting linker **S24** (TFA cleavage product of synthesis linker **S7**), and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of aryl iodide **S31** support correct conversion to the expected product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for$[M+H]^+$.

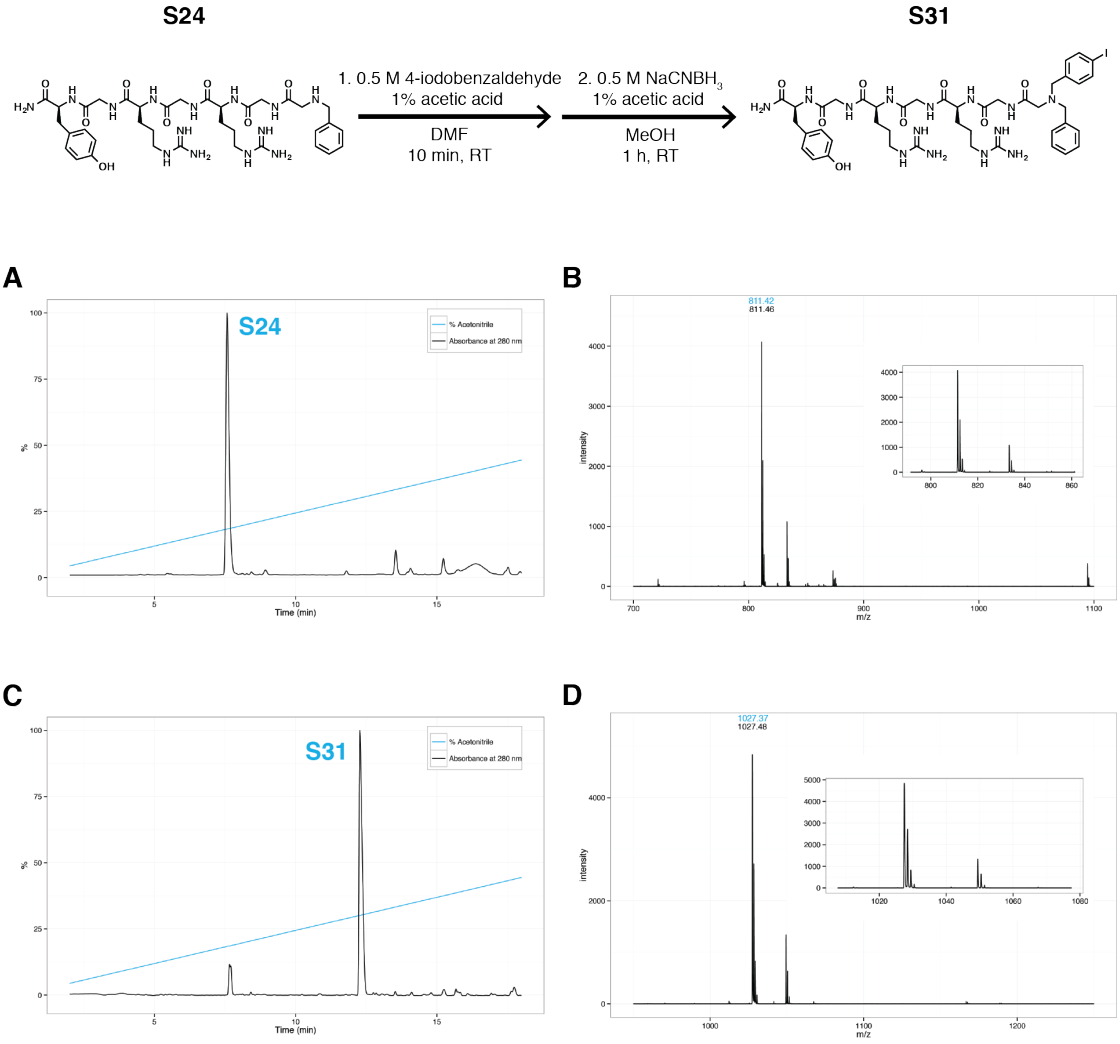
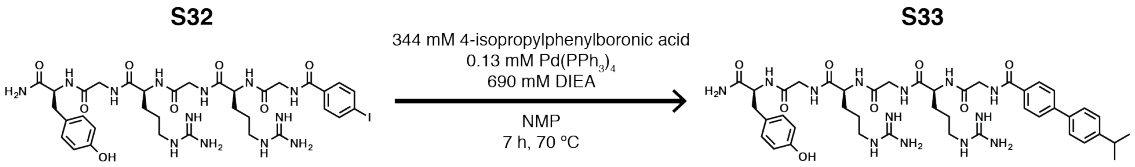


Figure S15. Suzuki-Miyaura cross-coupling to starting linker S8. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of aryl iodide linker **S32** (TFA cleavage product of synthesis linker **S8**), and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of **S33** support correct conversion to the expected Suzuki-Miyaura product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.



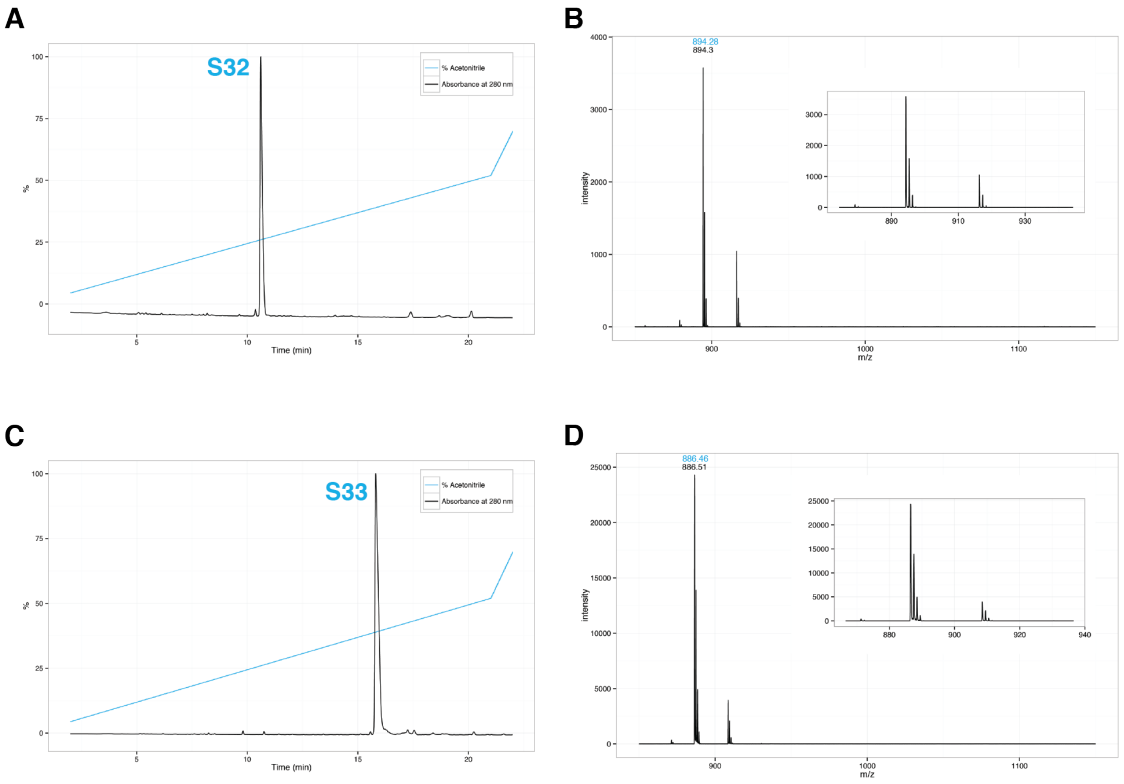


Figure S16. Solution-phase amino acid coupling. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of 5'-aminohexyl-modified oligonucleotide **S34**, HPLC (**C**) and MALDI-TOF MS analysis of unreacted starting material **S34** (**D**) and Fmoc-Lys(Alloc)-modified oligonucleotide **S35** (**E**) support correct conversion to the expected amino acid-coupled product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

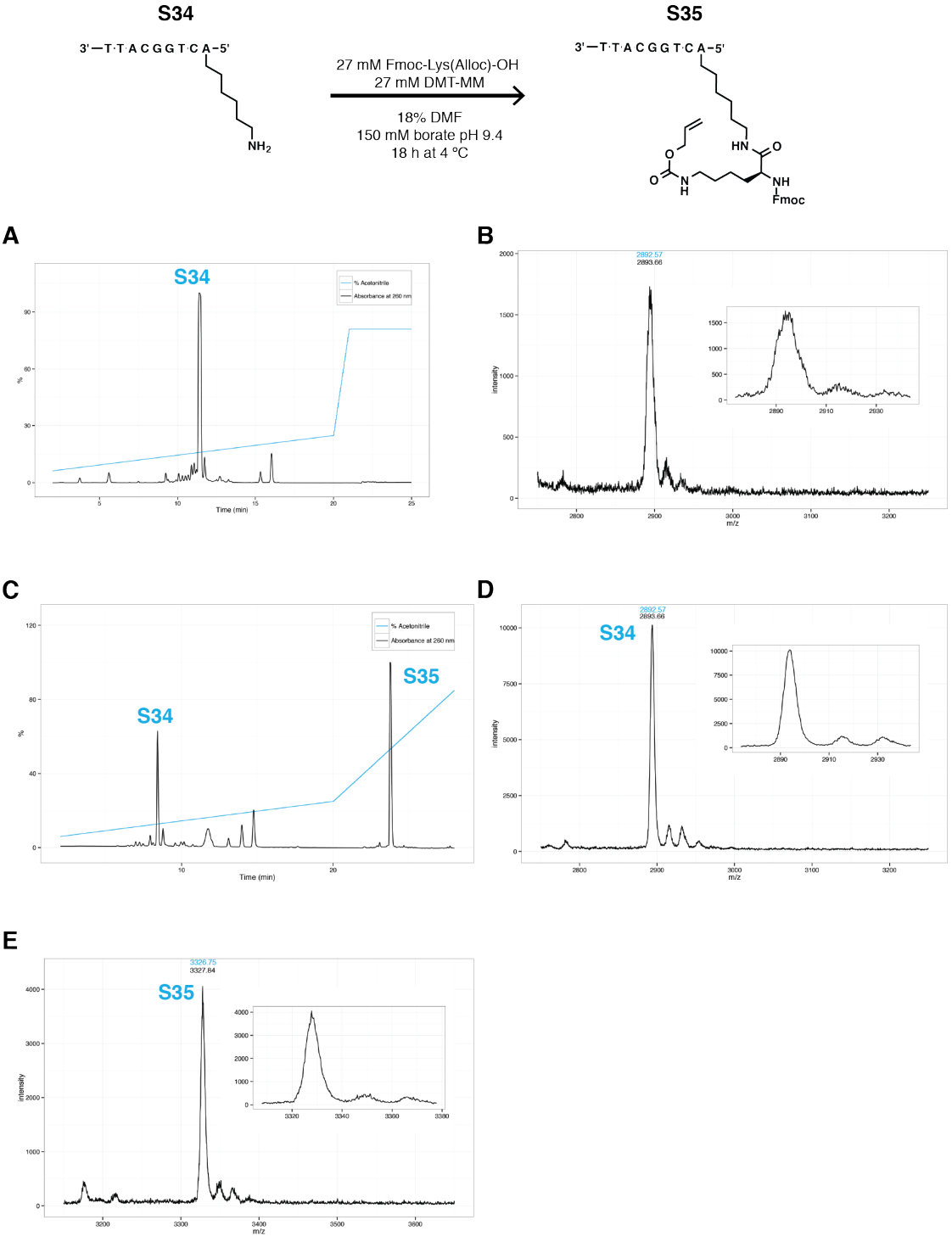


Figure S17. Solution-phase Fmoc deprotection. HPLC (**A**) analysis of Fmoc-Lys(Alloc)-modified oligonucleotide **S35**, MALDI-TOF MS analysis of unreacted 5'-aminohexyl-modified oligonucleotide **S34** (**B**) and Fmoc-Lys(Alloc)-modified oligonucleotide **S35** (**C**), and HPLC (**D**) and MALTI-TOF MS (**E**) analysis of Fmoc-deprotected oligonucleotide **S36** support the generation of the expected Fmoc-deprotected product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

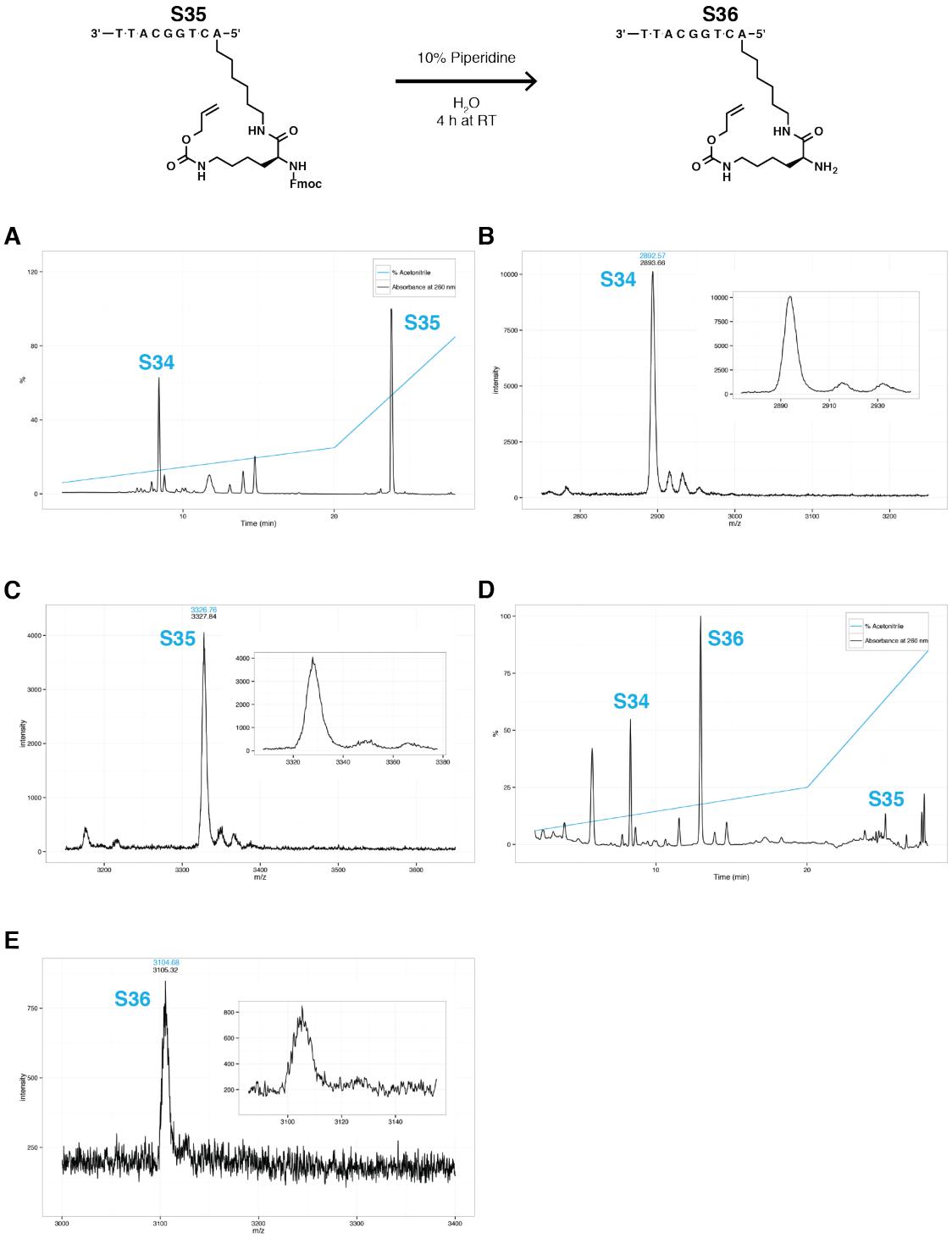


Figure S18. Solution-phase Alloc deprotection. HPLC (**A**) and MALDI-TOF (**B**) analysis of Fmoc-Lys(Alloc)-modified oligonucleotide **S35**, and HPLC (**C**) analysis of Alloc-deprotected product including MALDI-TOF MS analysis of unreacted starting material **S34** (**C**), Alloc- and Fmoc-deprotected product **S38** (**D**), and Alloc-deprotected product **S37** (**E**) support the generation of the expected Fmoc-deprotected product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

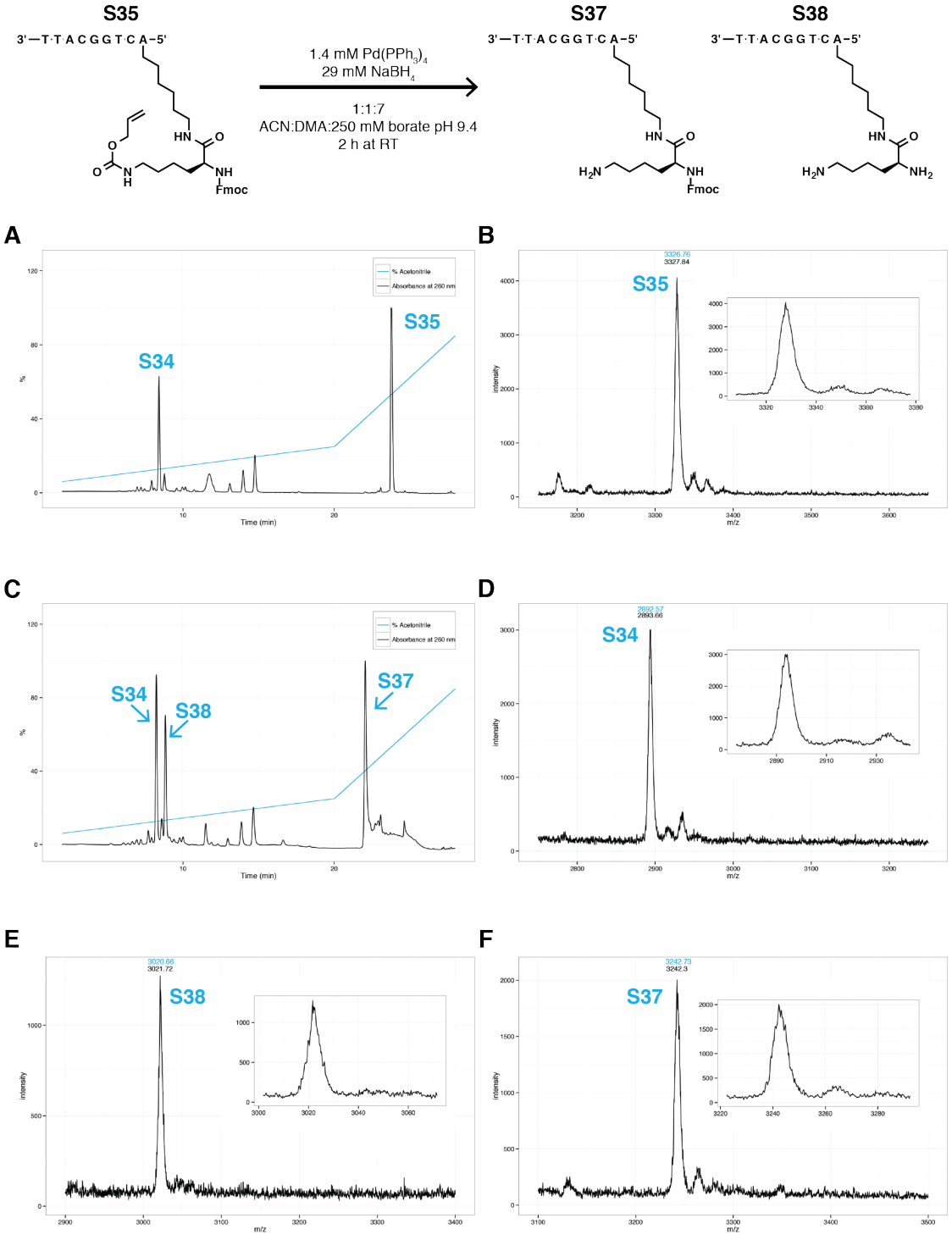


Figure S19. Solution-phase Suzuki-Miyaura coupling starting material preparation. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of 5'-aminohexyl-modified oligonucleotide **S34**, and HPLC (**C**) and MALDI-TOF MS (**D**) analysis of **S39** support correct conversion to the expected aryl iodide product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

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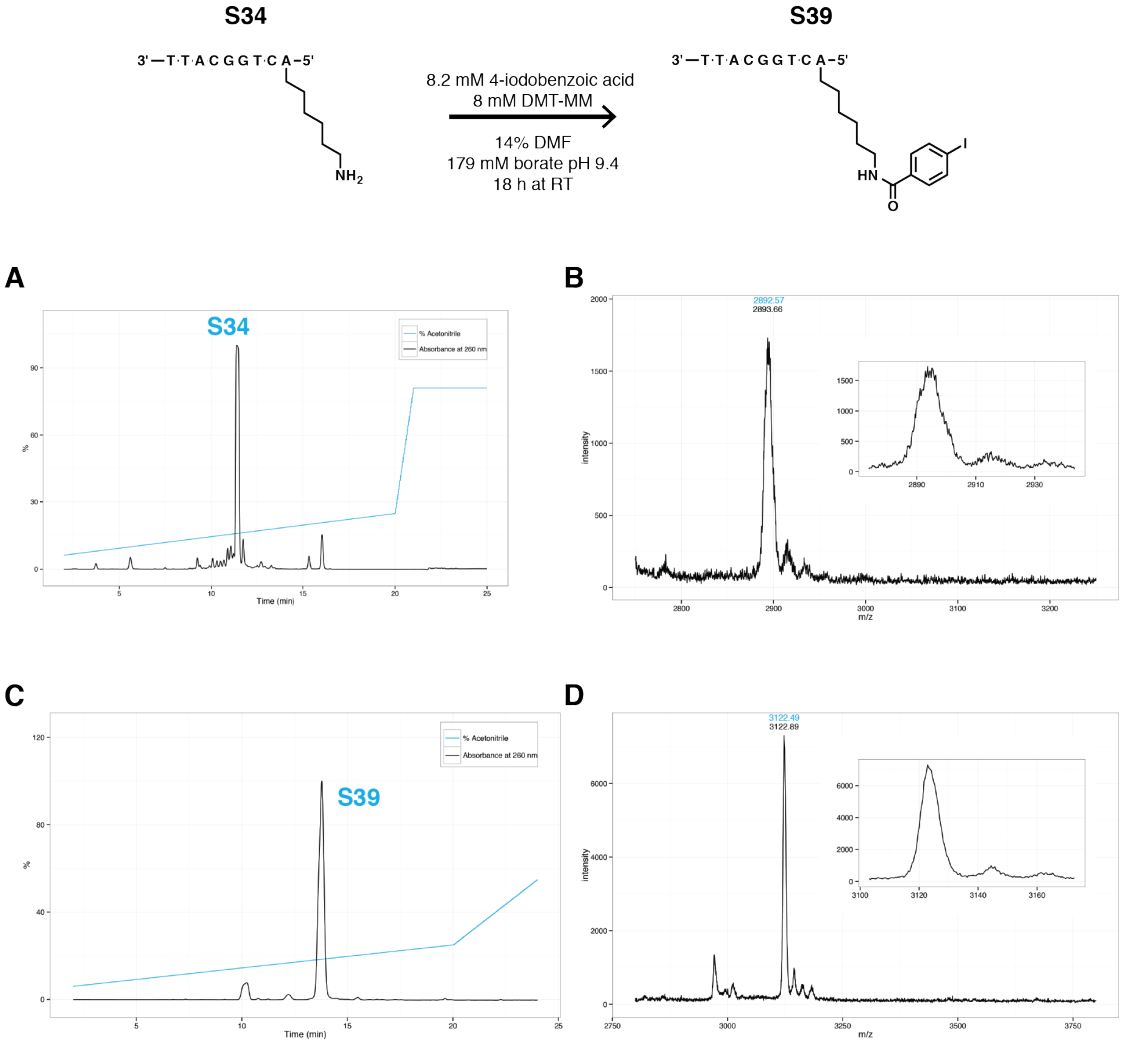

Figure S20. Solution-phase Suzuki-Miyaura cross-coupling. HPLC (**A**) and MALDI-TOF MS (**B**) analysis of aryl iodide **S39**, and HPLC (**C**) and MALDI-TOF MS analysis of Suzuki-Miyaura reaction, including oligonucleotide cleavage fragment **S41** (**D**), and **S40** (**E**) support correct conversion to the expected Suzuki-Miyaura product. Mass spectra display the theoretical exact mass (cyan) and observed mass (black) for $[M+H]^+$.

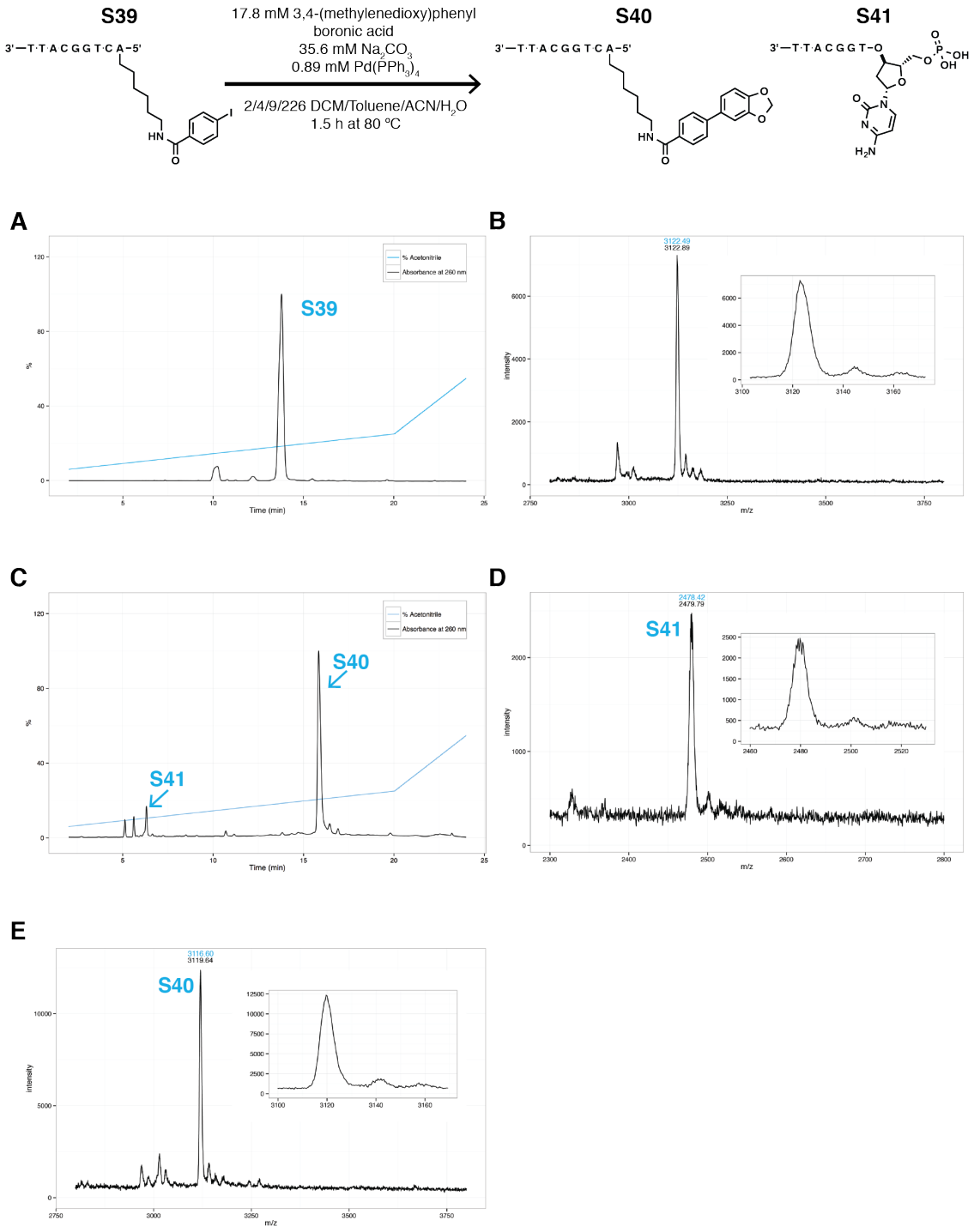

Figure S21. Example qPCR analysis of magnetic sensor beads. Fluorescence trace monitoring 5-Cy5 fluorescence (**A**) and standard curve (**B**) were typical of qPCR analysis.