Catch & release of alkyne-tagged molecules in water by polymer-supported cobalt complex

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1. General

$^1$H, $^{13}$C and $^{31}$P NMR spectra were measured on a JEOL JNM-AL400 spectrometer at 400, 100 and 160 MHz, respectively. All chemical shifts were recorded in $\delta$ (ppm) relative to tetramethylsilane (TMS), chloroform or H$_3$PO$_4$. Infrared spectra were measured on a Thermo Nicolet iS5, and only diagnostic absorptions are listed below. HPLC analysis was performed on DIONEX UltiMate3000 consisting of the followings: pump, DGP-3600; auto-sampler, WPS-3000; flow manager, FLM-3x00; detector, VWD-3x00 measured at 280 nm. ESI-MS was taken on a Bruker micrOTOF-QII-RSL. Column chromatography was performed with silica gel N-60 (40-100 $\mu$m) purchased from Kanto Chemical Co., Inc. or Chromatorex (N-H type silica gel) purchased from Fuji Silysia Chemical, Ltd. TLC analysis was performed on Silica gel 60 F$_{254}$-coated glass plates (Merck). Visualization was accomplished by means of ultraviolet (UV) irradiation at 254 nm or by spraying 12-molybdo(VI)phosphoric acid ethanol solution as the developing agent. Water was prepared with a Millipore Milli-Q Advantage A10. Dehydrated tetrahydrofuran (THF), methanol (MeOH) and dichloromethane (CH$_2$Cl$_2$) were purchased from Wako Pure Chemical Industries, Ltd. PS-PEG was purchased from HiPep Laboratories Co., Inc. Phosphine ligand supported by PS-PEG was synthesized according to the literature.$^{[1]}$ Co$_2$(CO)$_8$ was purchased from Kanto Chemical Co., Inc. Co$_2$(CO)$_7$(PPh$_3$) was synthesized according to the literature.$^{[2]}$
2. Synthesis of 1, 2, 4, 5 and 9

2.1. Synthesis of 1, 2 and 4

2.1.1. Synthesis of S1

To a solution of dansyl chloride (312 mg, 1 mmol) in CH$_2$Cl$_2$ was added Et$_3$N (560 ml, 4 mmol) and 11-azido-3,6,9-trioxaundec-1-amine (220 µl, 1 mmol) at room temperature. The mixture was stirred for 9 h, then aqueous NaHCO$_3$ was added. After extraction with CH$_2$Cl$_2$, the combined organic layer was dried over Na$_2$SO$_4$ and filtered. The filtrate was concentrated under reduced pressure and the residue was subjected to column chromatography on N-H type silica gel (hexane/ethyl acetate = 3/1) to give S1 as a yellow viscous oil (418.3 mg, 93%). IR (neat): 2869, 2098, 1588, 1574, 1454, 1407, 1307, 1140, 1091, 941, 790 cm$^{-1}$; $^1$H NMR (CDCl$_3$, 400 MHz): $\delta$ 8.53 (d, 1H, $J = 8.5$ Hz), 8.30 (d, 1H, $J = 8.8$ Hz), 8.24 (dd, 1H, $J = 1.2$, 7.3 Hz), 7.58-7.49 (m, 2H), 7.18 (d, 1H, $J = 7.6$ Hz), 5.44 (t, 1H, $J = 5.8$ Hz), 3.68-3.60 (m, 6H), 3.52-3.48 (m, 2H), 3.40-3.37 (m, 6H), 2.89 ppm (s, 6H); $^{13}$C NMR (CDCl$_3$, 100 MHz): $\delta$ 151.8, 135.0, 130.3, 129.8, 129.6, 129.2, 128.2, 123.1, 118.9, 115.1, 70.6, 70.6, 70.4, 70.3, 70.0, 69.2, 50.7, 45.4, 43.1 ppm; HRMS (ESI$^+$), Calcd for [C$_{20}$H$_{29}$O$_5$SNa$^+$]: $m/z = 474.1782$, Found: $m/z = 474.1784$.
2.1.2. Synthesis of 2

To a solution of **S1** (390 mg, 0.86 mmol) in MeOH (10 ml) was added Pd/C-ethylene diamine complex (39 mg) at room temperature under nitrogen. The nitrogen was replaced with hydrogen, and the mixture was stirred for 5 h, then filtered through a pad of Celite. The filtrate was evaporated and the residue was subjected to column chromatography on N-H type silica gel (CH₂Cl₂/MeOH = 95/5) to give 2 as a yellow viscous oil (350.8 mg, 95%). IR (neat): 2865, 1587, 1573, 1453, 1405, 1352, 1139, 1091, 941, 790, 750 cm⁻¹; ¹H NMR (CDCl₃, 400 MHz): δ 8.49 (d, 1H, J = 8.3 Hz), 8.34 (d, 1H, J = 8.8 Hz), 8.20 (dd, 1H, J = 1.2, 7.3 Hz), 7.54-7.45 (m, 2H), 7.15 (d, 1H, J = 7.6 Hz), 3.62-3.56 (m, 4H), 3.52-3.49 (m, 4H), 3.40-3.36 (m, 4H), 3.09 (t, 2H, J = 4.9 Hz), 2.85-2.84 ppm (m, 8H); ¹³C NMR (CDCl₃, 100 MHz): δ 151.7, 135.6, 129.9, 129.8, 129.6, 128.9, 127.9, 123.1, 119.2, 115.0, 73.1, 70.5, 70.4, 70.2, 70.1, 69.7, 45.4, 43.0, 41.5 ppm; HRMS (ESI⁺), Calcd for [C₂₀H₃₁O₅SN₃ + H]⁺: m/z = 426.2057, Found: m/z = 426.2053.

2.1.3. Synthesis of 3

To a solution of 4-nitrophenyl chloroformate (1 g, 5 mmol) in THF was added Et₃N (0.8 ml, 5.8 mmol) and 2-propyn-1-ol (290 μl, 4.98 mmol) at 0 °C. The mixture was stirred for 1 h, then the reaction was quenched by addition of aqueous NH₄Cl. After extraction with ethyl acetate, the combined organic layer was dried over Na₂SO₄ and filtered. The filtrate was concentrated in vacuo and the residue was subjected to column chromatography on silica gel (hexane/ethyl acetate = 7/1) to give 3 as a colorless solid (905 mg, 82%). IR (neat): 3271, 2134, 1751, 1595, 1519, 1375, 1349, 1262, 1229, 1060, 966, 957, 859, 730 cm⁻¹; ¹H NMR (CDCl₃, 400 MHz): δ 8.28 (d, 2H, J = 9.3 Hz), 7.40 (d, 2H, J = 9.3 Hz), 4.88 (d, 2H, J = 2.2 Hz), 2.62 ppm (t, 1H, J = 2.2 Hz); ¹³C NMR (CDCl₃, 100 MHz): δ 155.1, 151.8, 145.4, 125.2, 121.6, 76.7, 76.0, 56.5 ppm; HRMS (ESI⁺), Calcd for [C₁₀H₇NO₅ + Na]⁺: m/z = 244.0216, Found: m/z = 244.0218.
2.1.4. Synthesis of 1

To a solution of 2 (350 mg, 0.82 mmol) in CH$_2$Cl$_2$ (10 ml) was added Et$_3$N (460 µl, 3.3 mmol) and 3 (221 mg, 1 mmol) at room temperature. The solution was stirred for 1 h, followed by addition of aqueous NaHCO$_3$. After extraction with ethyl acetate, the combined organic layer was washed with brine, dried over Na$_2$SO$_4$ and filtered. The filtrate was evaporated in vacuo and the residue was subjected to column chromatography on silica gel (hexane/ethyl acetate = 3/1) to give 1 as a yellow viscous oil (368.3 mg, 88%). IR (neat): 3267, 2870, 1711, 1588, 1573, 1525, 1455, 1319, 1232, 1139, 1090, 941, 790 cm$^{-1}$; $^1$H NMR (CDCl$_3$, 400 MHz): $\delta$ 8.53 (d, 1H, $J$ = 8.5 Hz), 8.31 (d, 1H, $J$ = 8.5 Hz), 8.24 (dd, 1H, $J$ = 1.5, 7.3 Hz), 7.56-7.49 (m, 2H), 7.18 (d, 1H, $J$ = 7.6 Hz), 5.57-5.49 (m, 2H), 4.65 (d, 2H, $J$ = 2.4 Hz), 3.63-3.54 (m, 6H), 3.50-3.48 (m, 2H), 3.43-3.35 (m, 6H), 3.11 (d, 2H, $J$ = 4.9, 5.6 Hz), 2.89 (s, 6H), 2.43 ppm (t, 1H, $J$ = 2.4 Hz); $^{13}$C NMR (CDCl$_3$, 100 MHz): $\delta$ 155.3, 151.7, 134.9, 130.2, 129.7, 129.5, 129.2, 128.1, 123.0, 118.8, 115.0, 78.3, 74.4, 70.3, 70.2, 70.1, 70.1, 69.8, 69.2, 52.3, 45.4, 43.0, 40.8 ppm; HRMS (ESI$^+$), Calcd for [C$_{24}$H$_{33}$O$_7$SN$_3$ + H]$^+$: m/z = 508.2112, Found: m/z = 508.2113.

2.1.5. Synthesis of 4

To a solution of 1 (10 mg, 0.02 mmol) in THF (1 ml) was added Co$_2$(CO)$_7$(PPh$_3$) (11.4 mg, 0.02 mmol) at room temperature. The mixture was stirred for 1 h, then concentrated under reduced pressure. The residue was subjected to column chromatography on silica gel (CH$_2$Cl$_2$/MeOH = 95/5) and GPC to give 4 [6.1 mg, 75% (based on recovered 1)] as a dark brown viscous liquid, together with 1 (6.0 mg). IR (neat): 2060, 1997, 1958 cm$^{-1}$; $^1$H NMR (CDCl$_3$, 400 MHz): $\delta$ 8.52 (d, 1H, $J$ = 9.0 Hz), 8.30 (d, 1H, $J$ = 9.0 Hz), 8.23 (dd, 1H, $J$ = 1.2, 7.3), 7.56-7.40 (m, 17H), 7.17 (d, 1H, $J$ = 7.3 Hz), 5.48-5.44 (m, 1H), 5.25-5.16 (m, 2H), 4.74 (d, 1H, $J$ = 14.4 Hz), 4.24 (d, 1H, $J$ = 14.4 Hz), 3.61-3.35 (m, 14H), 3.12-3.06 (m, 2H), 2.88 ppm (s, 6H); $^{13}$C NMR (CDCl$_3$, 100 MHz): $\delta$ 201.4 (brs), 156.2, 151.9, 135.0, 134.6, 134.2, 132.9, 132.8, 130.4, 129.9, 129.7, 129.4, 128.6, 128.5, 128.3, 123.2, 118.9, 115.2, 71.0, 70.5, 70.3, 70.2, 70.1, 69.2, 65.3, 45.4, 43.0, 40.7 ppm; $^{31}$P NMR (CDCl$_3$, 160 MHz): $\delta$ 53.9 ppm; HRMS (ESI$^+$), Calcd for [C$_{24}$H$_{33}$O$_7$SN$_3$Co$_2$(PC$_{10}$H$_{15}$)(CO)$_8$ + Na]$^+$: m/z = 1050.1253, Found: m/z = 1050.1253.
2.2. Synthesis of 9

To a solution of 2 (85 mg, 0.2 mmol) in dioxane/H₂O (3 ml/3 ml) were added NaHCO₃ (51 mg, 0.6 mmol) and benzyl chloroformate (34 µl, 0.24 mmol) at 0 °C. The solution was stirred for 2 h at 0 °C, followed by addition of aqueous NH₄Cl. After extraction with ethyl acetate, the combined organic layer was washed with brine, dried over MgSO₄ and filtered. The filtrate was concentrated under reduced pressure and the residue was subjected to column chromatography on silica gel (ethyl acetate) to give 9 as a yellow viscous oil (93.6 mg, 84%). IR (neat): 3311, 2969, 1701, 1587, 1573, 1521, 1454, 1321, 1231, 1140, 1091, 941, 790, 750 cm⁻¹; ¹H NMR (CDCl₃, 400 MHz): δ 8.52 (d, 1H, J = 8.5 Hz), 8.30 (d, 1H, J = 8.5 Hz), 8.23 (d, 1H, J = 7.3 Hz), 7.56-7.48 (m, 2H), 7.36-7.28 (m, 5H), 7.17 (d, 1H, J = 7.3 Hz), 5.47 (brs, 1H), 5.37 (brs, 1H), 5.07 (s, 2H), 3.61-3.34 (m, 14H), 3.08-3.04 (m, 2H), 2.88 ppm (s, 6H); ¹³C NMR (CDCl₃, 100 MHz): δ 156.3, 151.7, 136.5, 135.0, 130.1, 129.7, 129.5, 129.1, 128.3, 128.1, 127.9, 127.9, 123.0, 118.9, 115.0, 70.3, 70.2, 70.0, 69.9, 69.2, 66.5, 45.4, 42.9, 40.8 ppm; HRMS (ESI⁺), Calcd for [C₂₈H₃₇N₃O₇S + Na⁺]: m/z =582.2244, Found: m/z = 582.2247.

2.3. Synthesis of 5

To a solution of 2-propyn-1-ol (58 µl, 1 mmol) in THF (5 ml) was added Co₂(CO)₈ (342 mg, 1 mmol) at room temperature. The mixture was stirred for 1 h, then concentrated under reduced pressure. The residue was dissolved in benzene (5 ml) and triphenylphosphine (262 mg, 1 mmol) was added to the solution under nitrogen atmosphere. After stirring for 2 h at 60 °C, the reaction mixture was concentrated under reduced pressure. The residue was subjected to column chromatography on silica gel (hexane/ethyl acetate = 3/1) to give 5 (240 mg, 41%) as a brown viscous liquid. IR (neat): 2059, 1994 cm⁻¹; ¹H NMR (CDCl₃, 400 MHz): δ 7.49-7.40 (m, 15H),
5.21 (d, 1H, J = 3.7 Hz), 4.18 (dd, 1H, J = 5.0, 14.2 Hz), 3.90-3.85 (m, 1H), 1.34 ppm (dd, 1H, J = 5.0, 7.8 Hz); $^{13}$C NMR (CDCl$_3$, 100 MHz): δ 205.7, 204.6, 201.6, 134.5 (d, J = 42 Hz), 132.9 (d, J = 11 Hz), 130.4, 128.6 (d, J = 11 Hz), 92.1, 70.9, 62.8 ppm; $^{31}$P NMR (CDCl$_3$, 160 MHz): δ 53.9 ppm; LC-MS (ESI$^+$) [GL Sciences Inertsil ODS-3, H$_2$O/MeCN (0.1% TFA) = 84/16 (0-10 min) → 75/25 (10-25 min) → 23/77 (25-35 min), 100 μl/min, 280 nm, τ = 33.8 min], Calcd for [\((C_3H_4O)Co_2(PC_{18}H_{15})(CO) + Na]^+\): m/z = 486.9679, Found: m/z = 486.9662, Calcd for [\((C_3H_4O)Co_2(PC_{18}H_{15})(CO)_2 + Na]^+\): m/z = 514.9628, Found: m/z = 514.9604, Calcd for [\((C_3H_4O)Co_2(PC_{18}H_{15})(CO)_3 + Na]^+\): m/z = 542.9577, Found: m/z = 542.9562, Calcd for [\((C_3H_4O)Co_2(PC_{18}H_{15})(CO)_4 + Na]^+\): m/z = 570.9526, Found: m/z = 570.9509, Calcd for [\((C_3H_4O)Co_2(PC_{18}H_{15})(CO)_5 + Na]^+\): m/z = 598.9475, Found: m/z = 598.9459.

3. Reaction of 4 with TFA (Figure 1)

To a solution of triphenylphosphine-cobalt-alkyne complex 4 in 5% MeCN/H$_2$O (50 μM, 1 ml) was added TFA (50 μl) at 4 °C. The reaction mixture was stirred at 4 °C. Aliquots (5 μl) were taken from the reaction mixture at intervals and subjected to HPLC analysis to monitor the reaction. The conversions and yields of all materials were calculated with reference to standard curves. HPLC [GL Sciences Inertsil ODS-3, H$_2$O/MeCN (0.1% TFA) = 84/16 (0-10 min) → 75/25 (10-25 min) → 23/77 (25-35 min), 100 μl/min, 280 nm, τ (compound 2) = 7.0 min, τ (compound 1) = 19.9 min, τ (compound 4) = 32.4 min, τ (compound 5) = 33.8 min].

4. Synthesis of cobalt complexes 7 and 8 (Scheme 3)$^{[3]}$

4.1. Preparation of complex 7

To a solution of Co$_2$(CO)$_8$ (30 mg, 0.088 mmol) in THF (0.5 ml) was added phosphine supported on PS-PEG (30 mg, <0.28 mmol/g). After rotation for 3 h in a tube rotator, the mixture was filtered on a glass filter and the beads were washed with THF (10 ml) and MeOH (10 ml). The IR spectrum was directly measured. IR (neat): 2077, 2021, 1985, 1956 cm$^{-1}$. 


4.2. Reaction of alkyne 1 and cobalt complex 7

Cobalt complex 7 supported on PS-PEG (30 mg, <0.28 mmol/g) was added to a solution of 1 (9 mg, 0.018 mmol) in Hepes buffer (pH 7.0) containing 150 mM NaCl, 10 mM Hepes-Na, 0.5% and Triton X-100 (9 ml) at room temperature. The mixture was rotated for 6 h, then centrifuged, and the supernatant was collected. The beads were washed with the same buffer (9 ml × 3), and the IR spectrum was directly measured in a wet condition. IR (neat): 2064, 2012, 2002, 1960 cm⁻¹.

5. Catch & release reaction of alkyne 1 at 4 °C under highly diluted conditions (Scheme 4)

Cobalt complex 7 (10 mg) was added to a solution of 1 (50 μM, 1 ml) in Hepes buffer (pH 7.0) containing 150 mM NaCl, 10 mM Hepes-Na, and 0.5% Triton X-100 at 4 °C. The mixture was rotated for 12 h at the same temperature, then centrifuged. After removal of the supernatant, the beads were washed with Hepes buffer (1 ml × 10). The supernatant and the washing solutions were subjected to fluorescence analysis (excitation: 355 nm, emission: 538 nm) to determine the conversion. To the beads was added 5% TFA solution (1 ml) and the mixture was rotated for 24 h. An aliquot of the supernatant was subjected to HPLC analysis to determine the yields of products. The conversions and yields of all materials were calculated with reference to standard curves. HPLC [GL Sciences Inertsil ODS-3, H₂O/MeCN (0.1% TFA) = 86/14 (0-10 min) → 77/23 (10-25 min), 100 μl/min, 280 nm, τ (compound 2) = 10.1 min, τ (compound 1) = 23.8 min].

6. Selective reaction of 1 in the presence of control material 9 (Scheme 5)

Cobalt beads 7 (10 mg) were added to a solution (1 ml) of 1 (50 μM) and 9 (50 μM) in Hepes buffer (pH 7.0) containing 150 mM NaCl, 10 mM Hepes-Na, and 0.5% Triton X-100 at 4 °C. The mixture was rotated for 12 h at the same temperature, then centrifuged. After removal of supernatant, the beads were washed with buffer (1 ml × 10). The supernatant and the washing solutions were subjected to HPLC analysis to determine conversions. To the beads was added 5% TFA in 5% MeCN/H₂O (1 ml) and the mixture was rotated for 24 h. A part of the supernatant was subjected to the HPLC analysis and the yields of all materials were calculated. The conversions and yields of all materials were calculated with reference to standard curves. HPLC [GL Sciences Inertsil ODS-3, H₂O/MeCN (0.1% TFA) = 86/14 (0-10 min) → 77/23 (10-25 min) → 61/39.
(25-35 min), 100 µl/min, 280 nm, τ (compound 2) = 10.1 min, τ (compound 1) = 23.8 min, τ (compound 9) = 31.0 min.

7. References


8. NMR spectra

[Diagram of NMR spectra with chemical structures]