

Supporting Information

Convergent synthesis of trifunctional molecules by three sequential azido-type-selective cycloadditions

Suguru Yoshida,^a Kimiyuki Kanno,^b Isao Kii,^{c,d} Yoshihiro Misawa,^a Masatoshi Hagiwara^c and Takamitsu Hosoya^{*,a}

^aLaboratory of Chemical Bioscience, Institute of Biomaterials and Bioengineering,
Tokyo Medical and Dental University (TMDU),
2-3-10 Kanda-Surugadai, Chiyoda-ku, Tokyo 101-0062, Japan

^bDepartment of Biological Information, Graduate School of Bioscience and Biotechnology,
Tokyo Institute of Technology, 4259 Nagatsuta-cho, Midori-ku, Yokohama 226-8501, Japan

^cDepartment of Anatomy and Developmental Biology, Kyoto University Graduate School of Medicine,
Yoshida-Konoe-cho, Sakyo-ku, Kyoto 606-8501, Japan

^dPresent address: Common Facilities Unit, Compass to Healthy Life Research Complex Program,
RIKEN Cluster for Science and Technology Hub,
6-7-3 Minatojima-minamimachi, Chuo-ku, Kobe 650-0047, Japan

Contents

General Remarks	S1
Chemical Experiments	S3
Biological Experiments	S17
Characterization Data of New Compounds	S20
References for Supporting Information	S36
¹H and ¹³C NMR Spectra of Compounds	S37

General Remarks

All reactions were performed in a dry glassware under atmosphere of argon, unless otherwise noted. Analytical thin-layer chromatography (TLC) was performed on precoated (0.25 mm) silica-gel plates (Merck Chemicals, Silica Gel 60 F₂₅₄, Cat. No. 1.05715). Column chromatography was conducted using silica-gel (Kanto Chemical Co., Inc., Silica Gel 60N, spherical neutral, particle size 40–50 μm, Cat. No. 37563-85). Preparative thin-layer chromatography (PTLC) was performed on silica-gel (Wako Pure Chemical Industries Ltd., Wakogel[®] B-5F, Cat. No. 230-00043). Melting points (Mp) were measured on a YANACO MP-J3 instrument or an OptiMelt MPA100 (Stanford Research Systems), and are uncorrected. ¹H NMR spectra were obtained with a Bruker AVANCE 400 or 500 spectrometer at 400 or 500 MHz, respectively. ¹³C NMR spectra were obtained with a Bruker AVANCE 500 at 126 MHz. CDCl₃ (Acros Organics, Cat. No. 368651000), DMSO-*d*₆ (CIL, Cat. No. DLM-10), or CD₃OD (Acros Organics, Cat. No. 321280100) was used as a solvent for obtaining NMR spectra. Chemical shifts (δ) are given in parts per million (ppm) downfield from (CH₃)₄Si (δ 0.00 for ¹H NMR in CDCl₃) or the solvent peak (δ 3.31 for ¹H NMR in CD₃OD, δ 2.49 for ¹H NMR in DMSO-*d*₆, δ 77.0 for ¹³C NMR in CDCl₃, and δ 49.2 for ¹³C NMR in CD₃OD) as an internal reference with coupling constants (*J*) in hertz (Hz). The abbreviations s, d, t, q, sept, m, and br signify singlet, doublet, triplet, quartet, septet, multiplet, and broad, respectively. IR spectra were measured by diffuse reflectance method on a Shimadzu IRPrestige-21 spectrometer attached with DRS-8000A with the absorption band given in cm⁻¹. High-resolution mass spectra (HRMS) were measured on a Bruker micrOTOF mass spectrometer under positive electrospray ionization (ESI⁺) or negative electrospray ionization (ESI⁻) conditions. Elemental analyses were carried out at the Center for Advanced Material Analysis (Ookayama) of Tokyo Institute of Technology. High-performance liquid chromatography (HPLC) was performed on a Shimadzu Prominence HPLC system (CBM-20A lite, LC-20AD × 2, DGU-20A3R, SUS316L, and CTO-20A) equipped with a Shimadzu SPD-20A UV/Vis detector. Fluorescence spectra (FL) were recorded on a JASCO FP-8500 spectrofluorophotometer (emission and excitation bandwidth, 1 nm) at 20 °C using a quartz cuvette (10 mm light path).

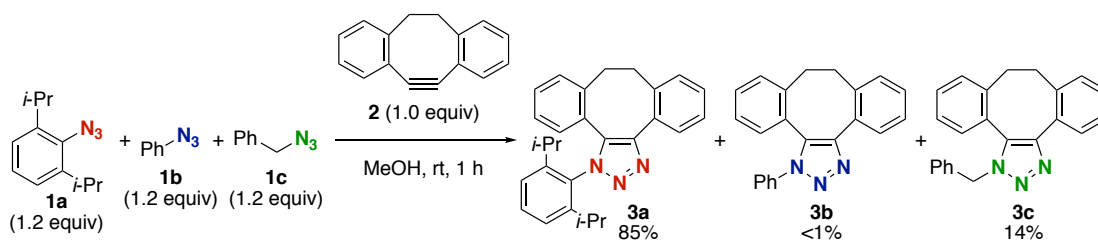
Unless otherwise noted, materials obtained from commercial suppliers were used without further purification. benzyl azide (**1c**) (Cat. No. 327-79632), acetylacetone (**7**) (Cat. No. 013-00493), potassium carbonate (Cat. No. 162-03495), sodium azide (Cat. No. 197-11091), sodium nitrite (Cat. No. 196-02575), acetic acid (Cat. No. 017-00256), triethylamine (Cat. No. 202-02646), methanol (Cat. No. 137-01823), benzene (Cat. No. 024-00706), toluene (Cat. No. 204-01861), *N,N*-dimethylformamide (DMF) (Cat. No. 045-02911), dimethylsulfoxide (DMSO) (Cat. No. 043-07211), dichloromethane (Cat. No. 135-02441), 1,4-dioxane (Cat. No. 042-31655), tetrahydrofuran (THF) (Cat. No. 204-08745), and 2-butanol (Cat. No. 026-11212) were purchased from Wako Pure Chemical Industries Ltd. Tetrakis(acetonitrile)copper(I) tetrafluoroborate (Cat. No. T2666), diketene (stabilized with ethylene glycol) (Cat. No. K0002), ca. 25 wt % aqueous solution of tetramethylammonium hydroxide (Cat. No. T1460), *N*-benzylacetoacetamide (Cat. No. B2808), and 1,1,2,2-tetrachloroethane (Cat. No. T0063) were purchased from Tokyo Chemical Industry Co., Ltd. 2-Methyl-3-butyn-2-ol (**4b**) (Cat. No. 25500-30) and potassium acetate (Cat. No. 32299-30) were purchased from Kanto Chemical Co. Inc. Phenylacetylene (**4a**) (Cat. No. 117706), pentamethylcyclopentadienylbis(triphenylphosphine)ruthenium(II) chloride (Cat. No. 673293), dichlorobis(triphenylphosphine)-palladium(II) (Cat. No. 412740), and 11-azido-3,6,9-trioxundecan-1-amine (Cat. No. 17758) were purchased from Sigma-Aldrich Japan. 5-(+)-Biotinamido)pentylamine (Cat. No. 21345) and (+)-biotinyl-3,6-dioxaoctanediamine (Cat. No. 21346) were purchased from Thermo Fisher Scientific K.K. Bis(pinacolato)diboron ((Bpin)₂) (Cat. No. AK-47583) was purchased from Ark Pharm, Inc.

2,6-Diisopropylphenyl azide (**1a**),^{S1} phenyl azide (**1b**),^{S2} 5,6-didehydro-11,12-dihydrodibenzo-*[a,e]*cyclooctene (**2**),^{S3} 11,12-didehydro-5,6-dihydrodibenzo-*[a,e]*cycloocten-5-yl 4-nitrophenyl carbonate,^{S3} 4-iodo-2,6-diisopropylaniline,^{S4} 1-azido-3-azidomethyl-5-iodobenzene (**9**),^{S5} 2-(2-((6-chlorohexyl)oxy)-ethoxy)ethylamine,^{S6} 2,5-dioxopyrrolidin-1-yl 4-ethynylbenzoate,^{S7} [3-ethyl-5-[(4-ethyl-3,5-dimethyl-2*H*-pyrrol-2-ylidene- κN)(4-ethynylphenyl)methyl]-2,4-dimethyl-1*H*-pyrrolato- κN]difluoroboron (**14**),^{S8} tris[(1-benzyl-1*H*-1,2,3-triazole-4-yl)methyl]amine (TBTA),^{S9} and (1,3-dimesitylimidazol-2-ylidene)copper bromide^{S10} were prepared according to the reported methods.

CAUTION! *Azido-containing compounds are presumed to be potentially explosive. Although we have never experienced such an explosion with azido compounds used in this study, all manipulations should be carefully carried out behind a safety shield in a hood.*

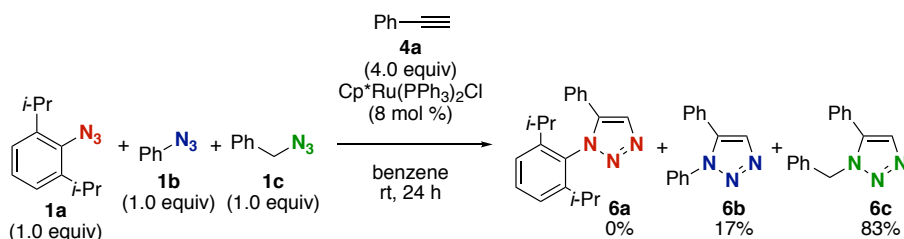
Chemical Experiments

Competition reaction A: concerted cycloaddition with cyclooctyne **2** (Table 1, entry 1)



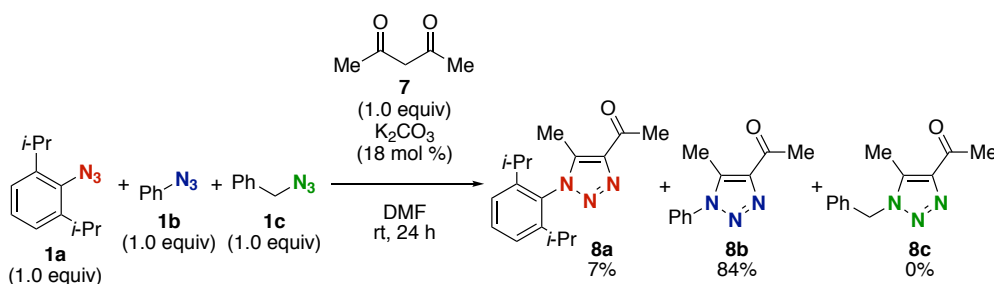
To a solution of 5,6-didehydro-11,12-dihydrodibenzo[*a,e*]cyclooctene (**2**) (20.4 mg, 99.9 μ mol, 1.00 equiv) dissolved in methanol (11.0 mL) was added dropwise a mixture of 2,6-diisopropylphenyl azide (**1a**) (24.4 mg, 0.120 mmol, 1.20 equiv), phenyl azide (**1b**) (14.3 mg, 0.120 mmol, 1.20 equiv), and benzyl azide (**1c**) (16.0 mg, 0.120 mmol, 1.20 equiv) dissolved in methanol (1.5 mL) at room temperature. After stirring for 1 h at the same temperature, the mixture was concentrated under reduced pressure. To the residue was added 1,1,2,2-tetrachloroethane (14.9 mg, 88.8 μ mol) as an internal standard, dissolved in CDCl_3 , and ^1H NMR analysis (400 MHz) was performed. Yields of **3a**, **3b**, and **3c** were determined to be 85.2%, <1%, and 14.2%, respectively, by comparing the relative values of integration for the peaks observed at 7.67–7.68 ppm (m, 1H) for **3a**, 7.60–7.63 ppm (m, 1H) for **3b**, and 5.56 ppm (s, 2H) for **3c** with that of 1,1,2,2-tetrachloroethane observed at 5.95 ppm (s, 2H).

Competition reaction B: ruthenium-catalyzed cycloaddition with terminal alkyne **4a** (Table 1, entry 4)



To a solution of phenylacetylene (**4a**) (81.7 mg, 0.800 mmol, 4.00 equiv) dissolved in benzene (1.0 mL) was added pentamethylcyclopentadienylbis(triphenylphosphine)ruthenium(II) chloride (13.0 mg, 16.3 μ mol, 8.2 mol %) at room temperature. After stirring for 5 min at the same temperature, to the solution was added a mixture of 2,6-diisopropylphenyl azide (**1a**) (40.7 mg, 0.200 mmol, 1.00 equiv), phenyl azide (**1b**) (23.8 mg, 0.200 mmol, 1.00 equiv), and benzyl azide (**1c**) (26.6 mg, 0.200 mmol, 1.00 equiv) dissolved in benzene (1.0 mL) at room temperature. After stirring for 24 h at the same temperature, the mixture was filtered through a short pad of silica gel (100 mg) and the filtrate was concentrated under reduced pressure. To the residue was added 1,1,2,2-tetrachloroethane (16.1 mg, 95.9 μ mol) as an internal standard, dissolved in CDCl_3 , and ^1H NMR analysis (400 MHz) was performed. Yields of **6a**, **6b**, and **6c** were determined to be 0%, 16.7%, and 83.4%, respectively, by comparing the relative values of integration for the peaks observed at 7.85 ppm (s, 1H) for **6b** and 5.52 ppm (s, 2H) for **6c** with that of 1,1,2,2-tetrachloroethane observed at 5.95 ppm (s, 2H).

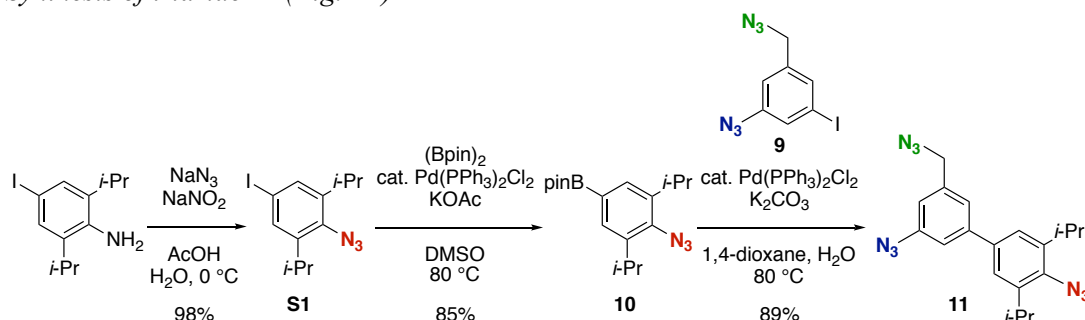
Competition reaction C: base-catalyzed cycloaddition with 1,3-diketone **7** (Table 1, entry 6)



To a mixture of 2,6-diisopropylphenyl azide (**1a**) (48.8 mg, 0.240 mmol, 1.00 equiv), phenyl azide (**1b**) (28.6 mg, 0.240 mmol, 1.00 equiv), benzyl azide (**1c**) (32.0 mg, 0.240 mmol, 1.00 equiv), and acetylacetone (**7**) (24.0 mg, 0.240 mmol, 1.00 equiv) dissolved in DMF (1.0 mL) was added potassium carbonate (6.0 mg,

43 μmol , 18 mol %) at room temperature. After stirring for 24 h at the same temperature, to the mixture was added water (10 mL). The mixture was extracted with EtOAc (20 mL $\times$ 3), and the combined organic extract was washed with water (10 mL $\times$ 3), brine (5 mL), dried (Na_2SO_4), and after filtration, the filtrate was concentrated under reduced pressure. To the residue was added 1,1,2,2-tetrachloroethane (15.9 mg, 94.5 μmol) as an internal standard, dissolved in CDCl_3 , and ^1H NMR analysis (400 MHz) was performed. Yields of **3a**, **3b**, and **3c** were determined to be 7.3%, 84.2%, and 0%, respectively, by comparing the relative values of integration for the peaks observed at 2.58 ppm (s, 3H) for **8a** and 2.34 ppm (s, 3H) for **8b** with that of 1,1,2,2-tetrachloroethane observed at 5.95 ppm (s, 2H).

Synthesis of triazide **11** (Fig. 2A)

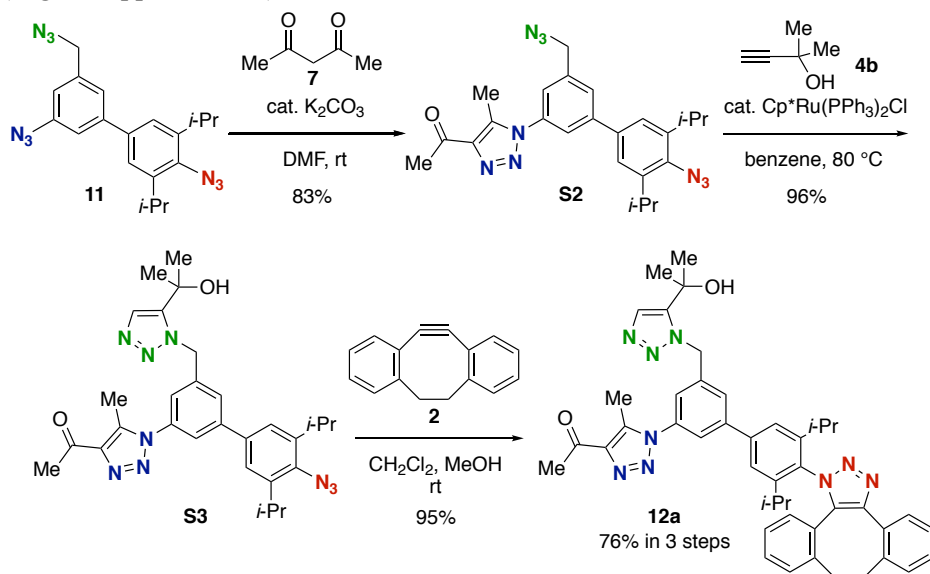


To a mixture of 4-iodo-2,6-diisopropylaniline (1.00 g, 3.30 mmol, 1.00 equiv) and sodium azide (429 mg, 6.60 mmol, 2.00 equiv) in acetic acid (20 mL) and distilled water (8.0 mL) was slowly added sodium nitrite (410 mg, 5.94 mmol, 1.80 equiv) at 0 °C. After stirring for 2 h at the same temperature, to the mixture was added a saturated aqueous solution of NaHCO_3 until the mixture became pH 10. The mixture was extracted with EtOAc (50 mL $\times$ 3), and the combined organic extract was washed with water (40 mL $\times$ 2) and brine (20 mL), and after filtration, the filtrate was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 30 g, *n*-hexane only) to give 4-iodo-2,6-diisopropylphenyl azide (**S1**) (1.06 g, 3.23 mmol, 97.9%) as a pale brown oil.

To a solution of 4-iodo-2,6-diisopropylphenyl azide (**S1**) (2.00 g, 6.08 mmol, 1.00 equiv) dissolved in DMSO (50 mL) were added bis(pinacolato)diboron (2.22 g, 8.74 mmol, 1.44 equiv), potassium acetate (1.77 g, 18.8 mmol, 3.09 equiv), and dichlorobis(triphenylphosphine)palladium(II) (220 mg, 0.313 mmol, 5.1 mol %) at room temperature and the mixture was heated at 80 °C with stirring for 2.5 h. After cooling to room temperature, to the mixture was added water (100 mL). The mixture was extracted with EtOAc (100 mL $\times$ 3), and the combined organic extract was washed with water (50 mL) and brine (20 mL $\times$ 2), and after filtration, the filtrate was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 50 g, *n*-hexane/EtOAc = 30/1 to 10/1) to give 2-(4-azido-3,5-diisopropylphenyl)-4,4,5,5-tetramethyl-1,3,2-dioxaborolane (**10**) (1.69 g, 5.14 mmol, 84.6%) as a pale brown oil.

To a solution of 1-azido-3-azidomethyl-5-iodobenzene (**9**) (150 mg, 0.500 mmol, 1.00 equiv) dissolved in 1,4-dioxane (2.5 mL) and water (0.5 mL) were added 2-(4-azido-3,5-diisopropylphenyl)-4,4,5,5-tetramethyl-1,3,2-dioxaborolane (**10**) (247 mg, 0.750 mmol, 1.50 equiv), potassium carbonate (140 mg, 1.01 mmol, 2.02 equiv), dichlorobis(triphenylphosphine)palladium(II) (18.0 mg, 25.6 μmol , 5.1 mol %) at room temperature and the mixture was heated at 80 °C with stirring for 2 h. After cooling to room temperature, to the mixture was added water (20 mL). The mixture was extracted with EtOAc (20 mL $\times$ 3), and the combined organic extract was washed with water (10 mL) and brine (5 mL $\times$ 2), and after filtration, the filtrate was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 10 g, *n*-hexane only) to give 3',4-diazido-5'-azidomethyl-3,5-diisopropyl-1,1'-biphenyl (**11**) (167 mg, 0.444 mmol, 88.8%) as a pale brown oil.

Three sequential cycloadditions of triazide **11** with three different azidophiles (**1**):
 1) base-catalyzed cycloaddition, 2) ruthenium-catalyzed cycloaddition, and 3) strain-promoted cycloaddition
 (Fig. 2B upper scheme)

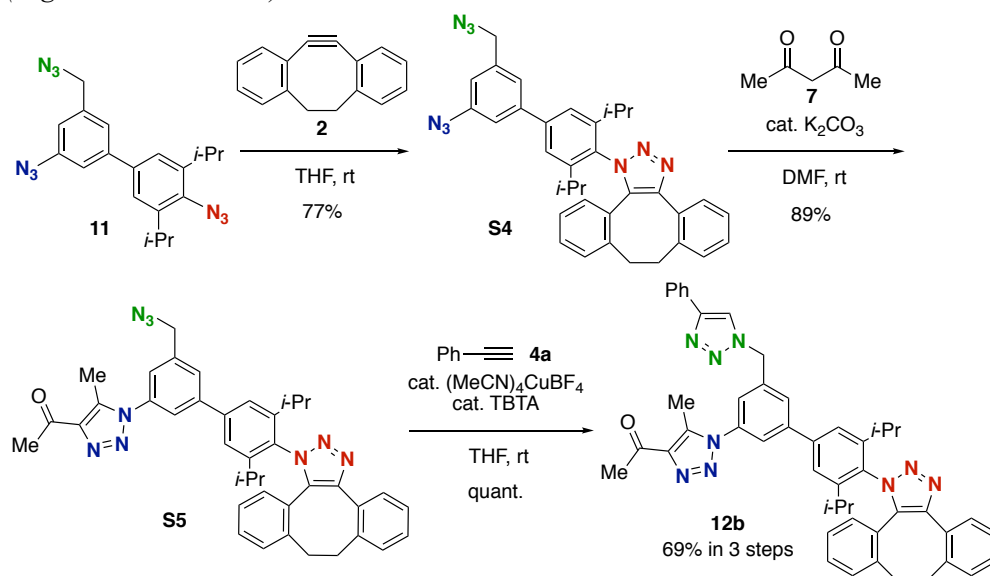


To a solution of 3,4'-diazido-5'-azidomethyl-3,5-diisopropyl-1,1'-biphenyl (**11**) (80.0 mg, 0.213 mmol, 1.00 equiv) dissolved in DMF (2.1 mL) were added acetylacetone (**7**) (25.6 mg, 0.256 mmol, 1.20 equiv) and potassium carbonate (5.9 mg, 43 μ mol, 20 mol %) at room temperature. After stirring for 24 h at the same temperature, to the mixture was added water (10 mL). The mixture was extracted with EtOAc (20 mL $\times$ 3), and the combined organic extract was washed with water (10 mL $\times$ 3), brine (5 mL), dried (Na_2SO_4), and after filtration, the filtrate was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, *n*-hexane/EtOAc = 10/1 to 3/1) to give 1-(1-(4'-azido-5'-azidomethyl-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**S2**) (80.6 mg, 0.176 mmol, 82.7%) as a pale yellow solid.

To a solution of 1-(1-(4'-azido-5'-azidomethyl-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**S2**) (34.1 mg, 74.5 μ mol, 1.00 equiv) dissolved in benzene (0.80 mL) were added 2-methyl-3-butyn-2-ol (**4b**) (9.4 mg, 0.12 mmol, 1.6 equiv) and (pentamethylcyclopentadienyl)bis(triphenylphosphine)ruthenium(II) chloride (3.0 mg, 3.8 μ mol, 5.1 mol %) at room temperature and the mixture was heated at 80 °C with stirring for 2 h. After cooling to room temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, *n*-hexane only; *n*-hexane/EtOAc = 3/1 to 1/1; EtOAc only) to give 1-(1-(4'-azido-5'-((5-(2-hydroxypropan-2-yl)-1*H*-1,2,3-triazol-1-yl)methyl)-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**S3**) (38.7 mg, 71.4 μ mol, 95.9%) as a pale yellow solid.

To a solution of 1-(1-(4'-azido-5'-((5-(2-hydroxypropan-2-yl)-1*H*-1,2,3-triazol-1-yl)methyl)-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**S3**) (15.3 mg, 28.2 μ mol, 1.00 equiv) dissolved in dichloromethane (0.50 mL) and methanol (0.50 mL) was added 5,6-didehydro-11,12-dihydrodibenzo[*a,e*]cyclooctene (**2**) (6.9 mg, 34 μ mol, 1.2 equiv) at room temperature. After stirring for 2 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, *n*-hexane/EtOAc = 3/1 to 1/1; EtOAc only) to give 1-(1-(4'-(8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazol-1-yl)-5'-((5-(2-hydroxypropan-2-yl)-1*H*-1,2,3-triazol-1-yl)methyl)-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**12a**) (20.0 mg, 26.8 μ mol, 94.9%) as a colorless solid.

Three sequential cycloadditions of triazide **11** with three different azidophiles (**2**):
 1) strain-promoted cycloaddition, 2) base-catalyzed cycloaddition, and 3) copper-catalyzed cycloaddition
 (Fig. 2B lower scheme)

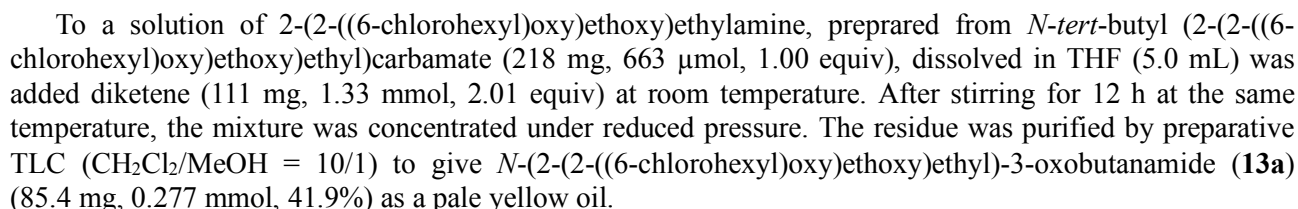


To a solution of 3,4'-diazido-5'-azidomethyl-3,5-diisopropyl-1,1'-biphenyl (**11**) (113 mg, 0.301 mmol, 3.01 equiv) dissolved in THF (6.5 mL) was added 5,6-didehydro-11,12-dihydrodibenzo[*a,e*]cyclooctene (**2**) (20.4 mg, 99.9 μ mol, 1.00 equiv) at room temperature. After stirring for 8 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by preparative TLC (*n*-hexane/EtOAc = 3/1) to remove the remaining **11**, followed by flash column chromatography (silica-gel 5 g, *n*-hexane/EtOAc = 10/1 to 3/1) to give 1-(3'-azido-5'-azidomethyl-3,5-diisopropyl-1,1'-biphenyl-4-yl)-8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazole (**S4**) (44.3 mg, 76.4 μ mol, 76.5%) as a colorless solid.

To a solution of 1-(3'-azido-5'-azidomethyl-3,5-diisopropyl-1,1'-biphenyl-4-yl)-8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazole (**S4**) (18.2 mg, 31.4 μ mol, 1.00 equiv) dissolved in DMF (0.30 mL) were added acetylacetone (**7**) (3.8 mg, 38 μ mol, 1.2 equiv) and potassium carbonate (1.0 mg, 7.2 μ mol, 23 mol %) at room temperature. After stirring for 24 h at the same temperature, to the mixture was added water (5 mL). The mixture was extracted with EtOAc (10 mL $\times$ 3), and the combined organic extract was washed with water (5 mL $\times$ 3), brine (5 mL), dried (Na₂SO₄), and after filtration, the filtrate was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, *n*-hexane/EtOAc = 10/1 to 1/1) to give 1-(5-azidomethyl-4'-(8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazol-1-yl)-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**S5**) (18.5 mg, 28.0 μ mol, 89.0%) as a colorless solid.

To a solution of 1-(5-azidomethyl-4'-(8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazol-1-yl)-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**S5**) (6.0 mg, 9.1 μ mol, 1.0 equiv) dissolved in THF (3.0 mL) were added phenylacetylene (**4a**) (1.5 mg, 15 μ mol, 1.6 equiv), TBTA (1.0 mg, 1.9 μ mol, 21 mol %), and tetrakis(acetonitrile)copper(I) tetrafluoroborate (1.0 mg, 3.2 μ mol, 35 mol %) at room temperature. After stirring for 24 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by preparative TLC (*n*-hexane/EtOAc = 1/1) to give 1-(1-(4'-(8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazol-1-yl)-3',5'-diisopropyl-5-((4-phenyl-1*H*-1,2,3-triazol-1-yl)methyl)-1,1'-biphenyl)-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**12b**) (6.9 mg, 9.0 μ mol, quant.) as a colorless solid.

Preparation of β -ketoamide **13a** bearing a HaloTag ligand moiety



The reaction scheme illustrates the synthesis of compound **13b** through two parallel pathways starting from a common azide-terminated poly(ethylene glycol) (PEG) chain.

Left Pathway: The starting material (azide-terminated PEG) reacts with 2-methylenedioxirane in THF at room temperature (rt) to yield intermediate **S6** in 75% yield. **S6** is a PEG chain with an azide terminus and a 2-oxo-2-methylpropanamide group.

Right Pathway: The starting material reacts with 4-ethynylbenzoic acid and N-methylsuccinimide in the presence of NEt_3 in CH_2Cl_2 at room temperature (rt) to yield intermediate **S7** in 99% yield. **S7** is a PEG chain with a terminal chloride and a 4-ethynylbenzamide group.

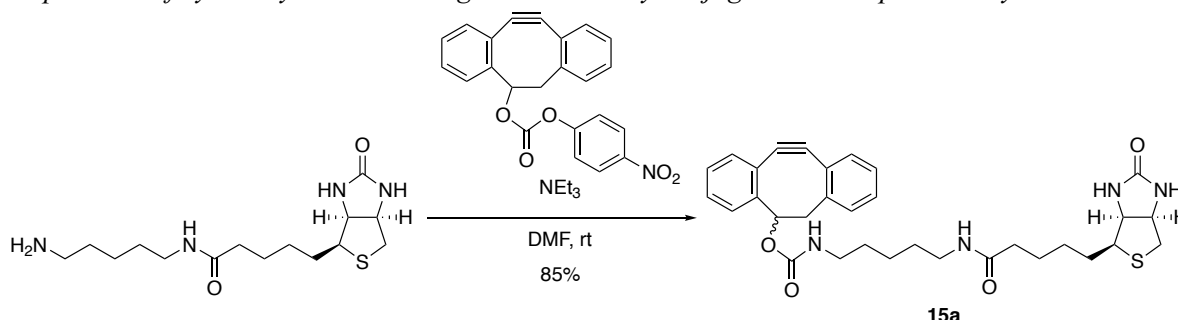
Coupling: Intermediates **S6** and **S7** are coupled using $\text{cat. (MeCN)}_4\text{CuBF}_4$ and cat. TBTA in THF at room temperature (rt) to form the final product **13b** in 92% yield. **13b** is a PEG chain containing both the 2-oxo-2-methylpropanamide and the 4-ethynylbenzamide moieties.

To a solution of 2-(2-((6-chlorohexyl)oxy)ethoxy)ethylamine, prepared from *N*-*tert*-butyl 2-(2-((6-chlorohexyl)oxy)ethoxy)ethylcarbamate (99.8 mg, 0.308 mmol, 1.07 equiv), dissolved in dichloromethane (3.0 mL) were added triethylamine (87.8 mg, 0.868 mmol, 3.01 equiv) and 2,5-dioxopyrrolidin-1-yl 4-ethynylbenzoate (70.0 mg, 0.288 mmol, 1.00 equiv) at room temperature. After stirring for 24 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 10 g, *n*-hexane/EtOAc = 2/1 to 1/1) to give *N*-(2-(2-((6-chlorohexyl)oxy)ethoxy)ethyl)-4-ethynylbenzamide (**S7**) (101 mg, 0.286 mmol, 99.3%) as a colorless solid.

S7

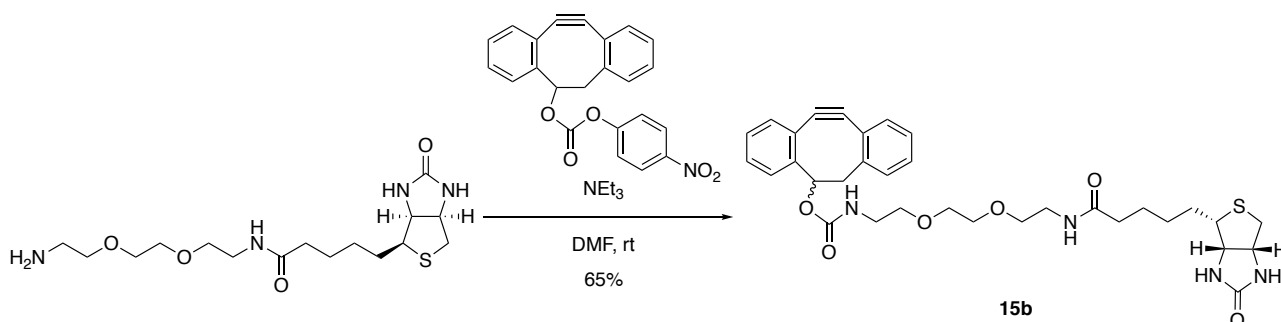
for 48 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, CH₂Cl₂ only; then CH₂Cl₂/MeOH = 30/1) to give *N*-(2-(2-((6-chlorohexyl)oxy)ethoxy)ethyl)-4-(1-(13,15-dioxo-3,6,9-trioxa-12-azahexadecyl)-1*H*-1,2,3-triazol-4-yl)benzamide (**13b**) (100 mg, 0.153 mmol, 92.4%) as a colorless solid.

Preparation of cyclooctyne 15a bearing a biotin moiety conjugated with a pentamethylene linker



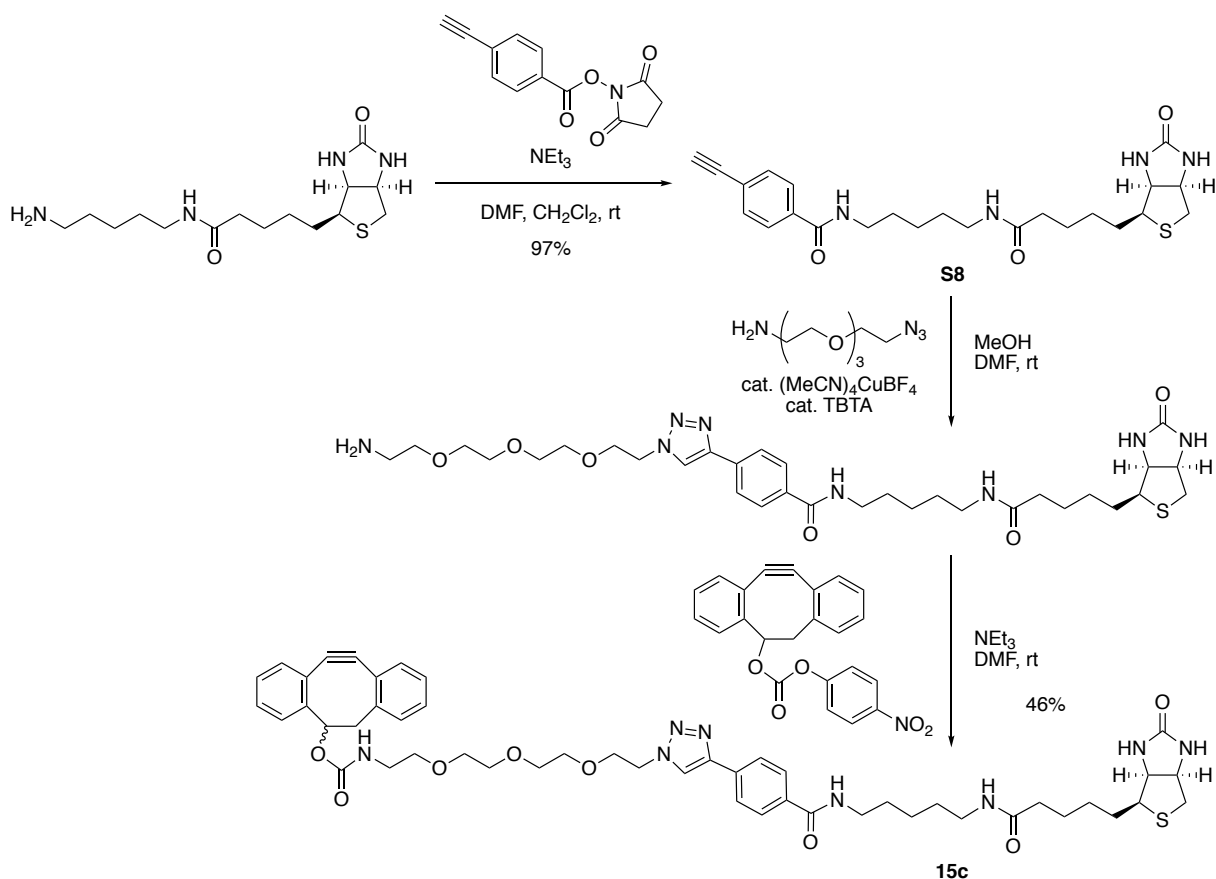
To a solution of 5-((+)-biotinamido)pentylamine (46.2 mg, 99.9 μ mol, 1.00 equiv) dissolved in DMF (10 mL) were added 11,12-didehydro-5,6-dihydrodibenzo[*a,e*]cycloocten-5-yl 4-nitrophenyl carbonate (46.2 mg, 0.120 mmol, 1.20 equiv) and triethylamine (30.4 mg, 0.300 mmol, 3.00 equiv) at room temperature. After stirring for 24 h at the same temperature, to the mixture was added 11,12-didehydro-5,6-dihydrodibenzo[*a,e*]cycloocten-5-yl 4-nitrophenyl carbonate (18.0 mg, 54.8 μ mol, 0.549 equiv) again at room temperature. After stirring for 4 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, CH₂Cl₂ only; then CH₂Cl₂/MeOH = 30/1 to 10/1) to give cyclooctyne **15a** (48.9 mg, 85.1 μ mol, 85.2%) as a colorless solid.

Preparation of cyclooctyne 15b bearing a biotin moiety conjugated with a 3,6-dioxaoctamethylene (PEO₂) linker



To a solution of 11,12-didehydro-5,6-dihydrodibenzo[*a,e*]cycloocten-5-yl 4-nitrophenyl carbonate (25.9 mg, 67.2 μ mol, 1.20 equiv) dissolved in DMF (5.0 mL) were added (+)-biotinyl-3,6-dioxaoctanediamine (21.0 mg, 56.1 μ mol, 1.00 equiv) and triethylamine (15.2 mg, 0.150 mmol, 2.67 equiv) at room temperature. After stirring for 24 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, CH₂Cl₂/MeOH = 20/1 to 6/1) to give cyclooctyne **15b** (22.7 mg, 36.6 μ mol, 65.2%) as a colorless solid.

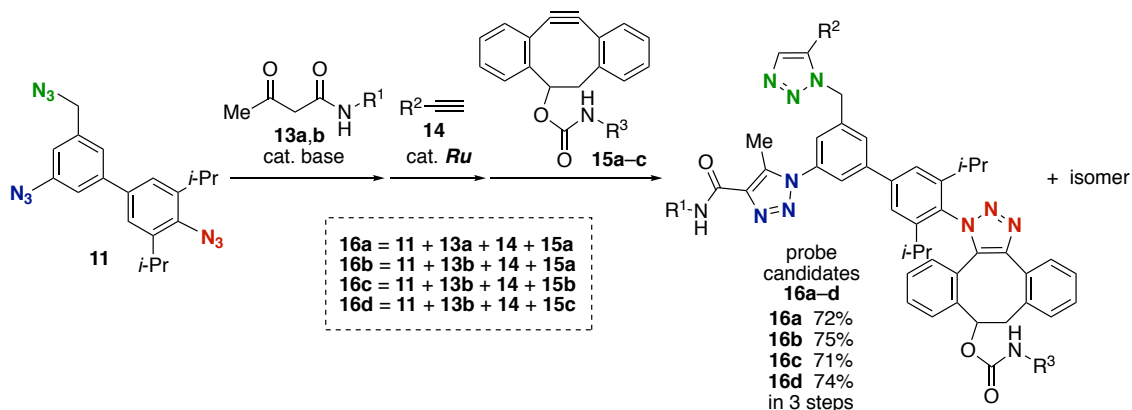
Preparation of cyclooctyne **15c** bearing a biotin moiety conjugated with 3,6,9-trioxaundecamethylene (PEO₃) and pentamethylene linkers linked by a 1,2,3-triazole skeleton



To a solution of 5-(+)-biotinamido)pentylamine (20.0 mg, 60.9 μ mol, 1.00 equiv) dissolved in dichloromethane (1.0 mL) and DMF (1.0 mL) were added 2,5-dioxopyrrolidin-1-yl 4-ethynylbenzoate (17.8 mg, 73.0 μ mol, 1.20 equiv) and triethylamine (18.2 mg, 0.180 mmol, 2.96 equiv) at room temperature. After stirring for 9 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, CH₂Cl₂/MeOH = 10/1 to MeOH only) to give 4-ethynyl-*N*-(5-(5-((3*aS*,4*S*,6*aR*)-2-oxohexahydro-1*H*-thieno[3,4-*d*]imidazol-4-yl)pentanamido)pentyl)benzamide (**S8**) (25.4 mg, 59.3 μ mol, 97.3%) as a colorless solid.

To a solution of 4-ethynyl-*N*-(5-(5-((3*aS*,4*S*,6*aR*)-2-oxohexahydro-1*H*-thieno[3,4-*d*]imidazol-4-yl)pentanamido)pentyl)benzamide (**S8**) (25.4 mg, 59.3 μ mol, 1.01 equiv) dissolved in MeOH (1.5 mL) and DMF (0.20 mL) were added 11-azido-3,6,9-trioxaundecan-1-amine (12.8 mg, 58.6 μ mol, 1.00 equiv), TBTA (3.2 mg, 6.0 μ mol, 10 mol %), and tetrakis(acetonitrile)copper(I) tetrafluoroborate (1.9 mg, 6.0 μ mol, 10 mol %) at room temperature. After stirring for 24 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was dissolved in DMF (2.0 mL) and to the solution were added 11,12-didehydro-5,6-dihydrodibenzo[*a,e*]cycloocten-5-yl 4-nitrophenyl carbonate (38.5 mg, 99.9 μ mol, 1.70 equiv) and triethylamine (18.2 mg, 0.180 mmol, 3.07 equiv) at room temperature. After stirring for 24 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, CH₂Cl₂/MeOH = 10/1 to MeOH only) to give cyclooctyne **15c** (25.0 mg, 27.1 μ mol, 46.3%) as a colorless solid.

Synthesis of trifunctional probe candidates **16a–d**



General procedures for triazole formation reactions conducted for the synthesis of trifunctional probe candidates **16a–d**

General procedure A: base-catalyzed cycloaddition at the sterically unhindered aromatic azido group of triazide **11** using a β -ketoamide

To a solution of 3,4'-diazido-5'-azidomethyl-3,5-diisopropyl-1,1'-biphenyl (**11**) (0.10 mmol) dissolved in DMF (1.0 mL) were added β -ketoamide **13a** or **13b** (0.12 mmol) and potassium carbonate (20 μ mol) at room temperature. After stirring for 24 h at the same temperature, to the mixture was added water (10 mL). The mixture was extracted with EtOAc (20 mL $\times$ 3), and the combined organic extract was washed with water (10 mL $\times$ 3), brine (5 mL), dried (Na_2SO_4), and after filtration, the filtrate was concentrated under reduced pressure. The residue was purified by flash column chromatography to give the corresponding triazole.

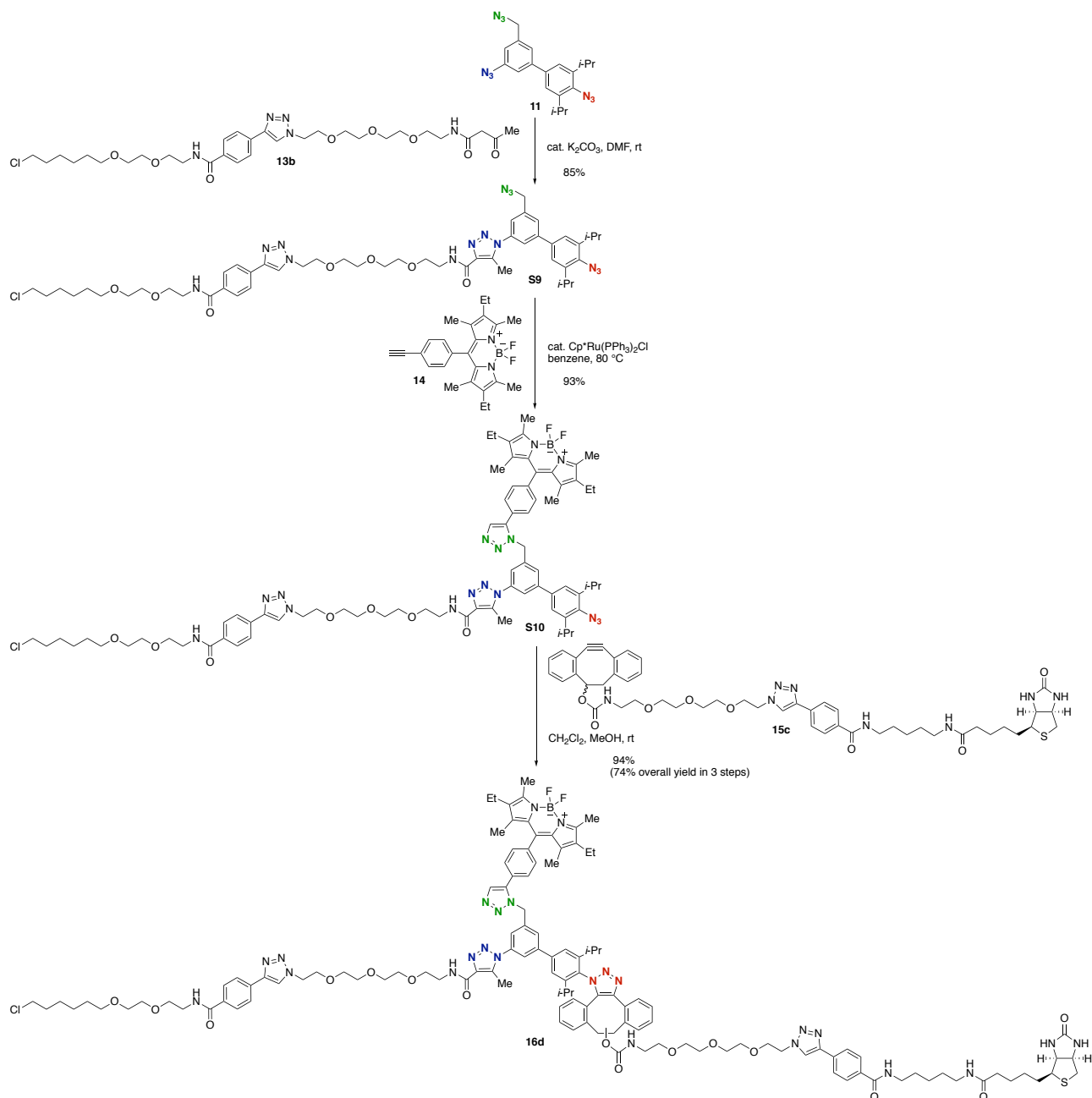
General procedure B: Ru-catalyzed cycloaddition at the aliphatic azido group of a diazido intermediate with terminal alkyne **14**

To a solution of diazide (75 μ mol) dissolved in toluene (0.80 mL) were added terminal alkyne **14** (0.120 mmol) and (pentamethylcyclopentadieny)bis(triphenylphosphine)ruthenium(II) chloride (4 μ mol) at room temperature and the mixture was heated at 80 $^\circ\text{C}$ with stirring for 2 h. After cooling to room temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography to give the corresponding bistriazole.

General procedure C: strain-promoted cycloaddition with a cyclooctyne

To a solution of azide (20 μ mol) dissolved in dichloromethane (0.50 mL) and methanol (0.50 mL) was added cyclooctyne **15a**, **15b**, or **15c** (22 μ mol) at room temperature. After stirring for 4 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography to give the corresponding tristriazole.

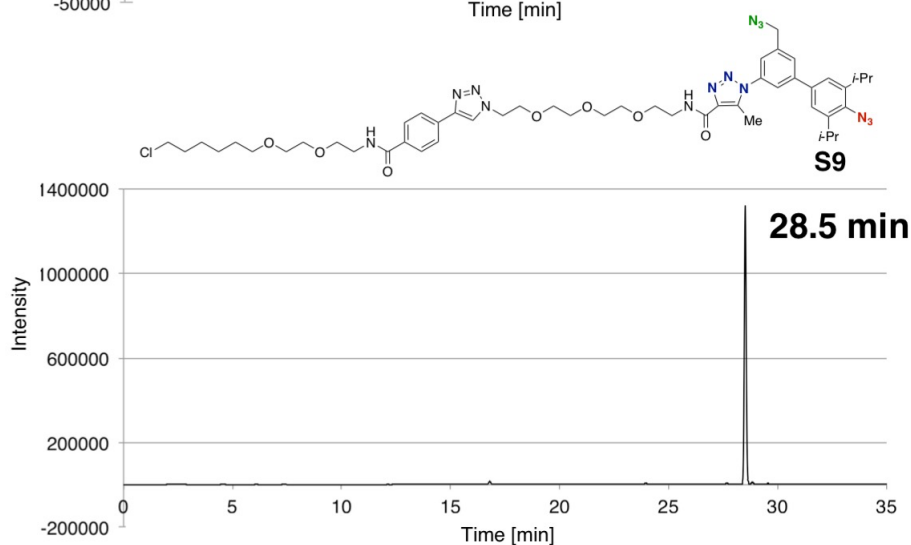
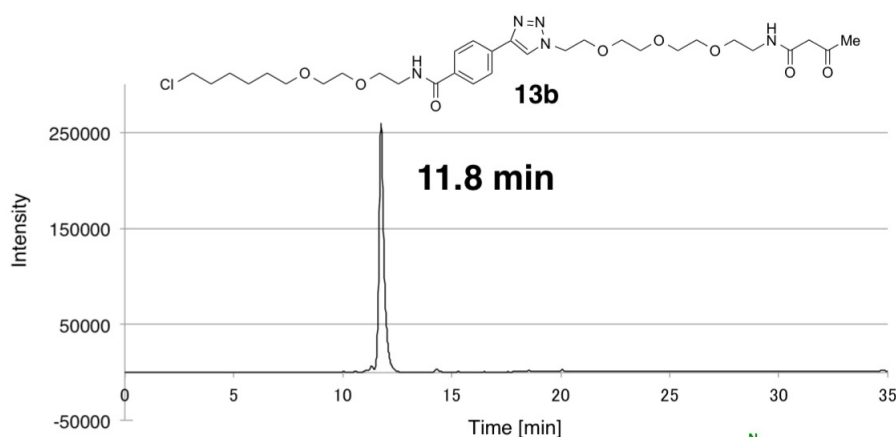
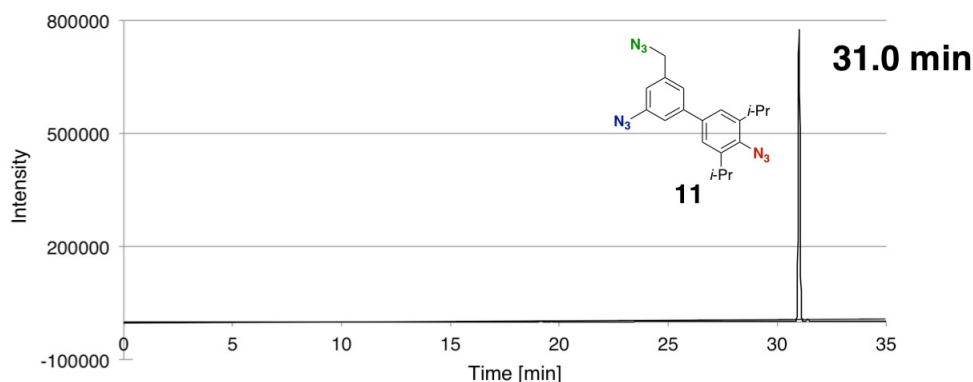
Synthesis of trifunctional probe **16d** (**11** + **13b** + **14** + **15c**)



Since NMR analysis of these compounds gave complex spectra, the purity of each product was confirmed by analytical reverse phase HPLC [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. $\times$ 250 mm); mobile phase: $\text{CH}_3\text{CN}:\text{H}_2\text{O}$ = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm].

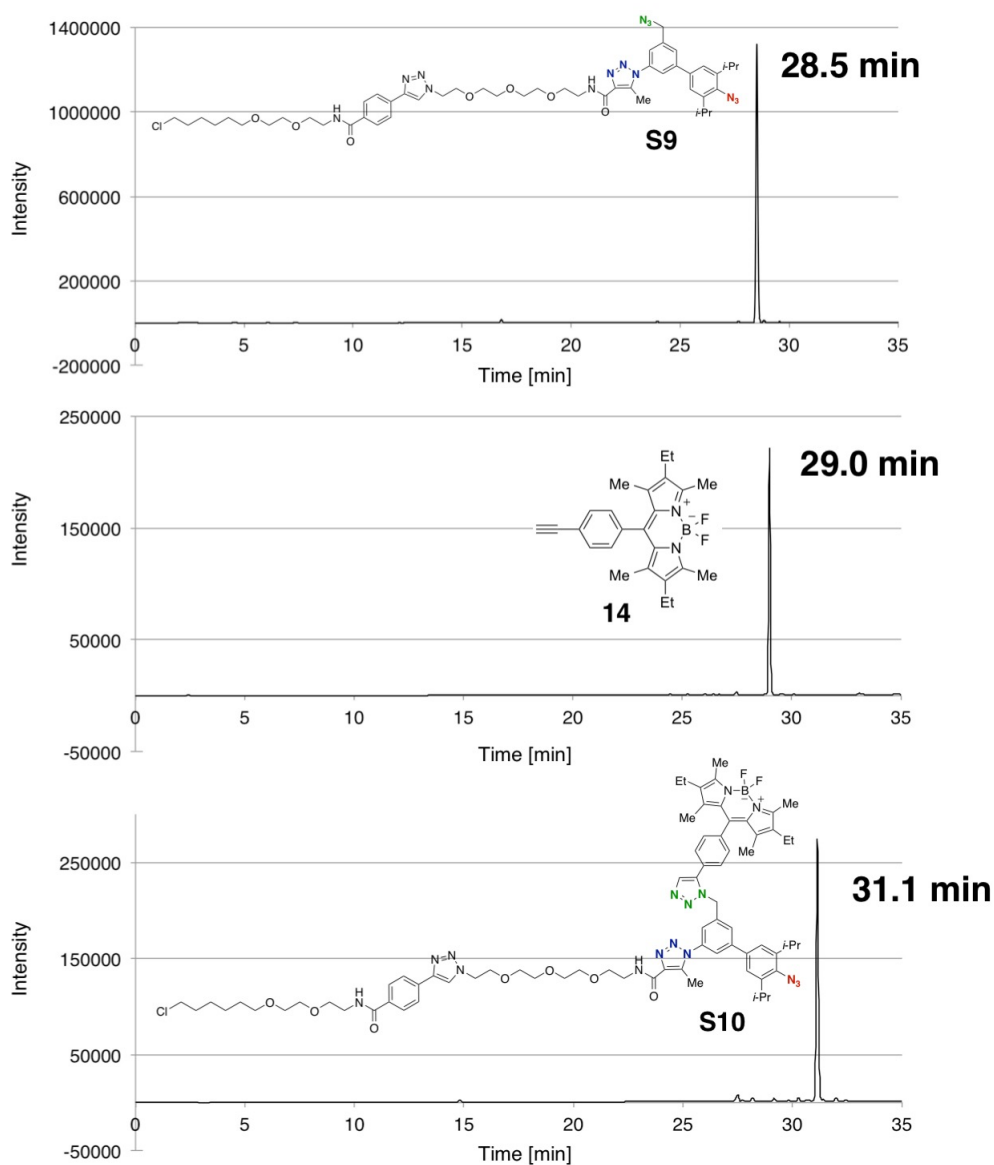
Base-catalyzed cycloaddition at the aromatic azido group of triazide 11 using β -ketoamide 13b

To a solution of 3',4'-diazido-5'-azidomethyl-3,5-diisopropyl-1,1'-biphenyl (**11**) (21.0 mg, 55.9 μ mol, 1.00 equiv) dissolved in DMF (1.0 mL) were added *N*-(2-(2-((6-chlorohexyl)oxy)ethoxy)ethyl)-4-(1-(13,15-dioxo-3,6,9-trioxa-12-azahexadecyl)-1*H*-1,2,3-triazol-4-yl)benzamide (**13b**) (44.0 mg, 67.3 μ mol, 1.20 equiv) and potassium carbonate (1.4 mg, 10 μ mol, 18 mol %) at room temperature. After stirring for 24 h at the same temperature, to the mixture was added water (10 mL). The mixture was extracted with EtOAc (20 mL $\times$ 3), and the combined organic extract was washed with water (10 mL $\times$ 3), brine (5 mL), dried (Na_2SO_4), and after filtration, the filtrate was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, $\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 50/1 to 30/1) to give diazide **S9** (48.3 mg, 47.7 μ mol, 85.4%) as a pale orange oil.



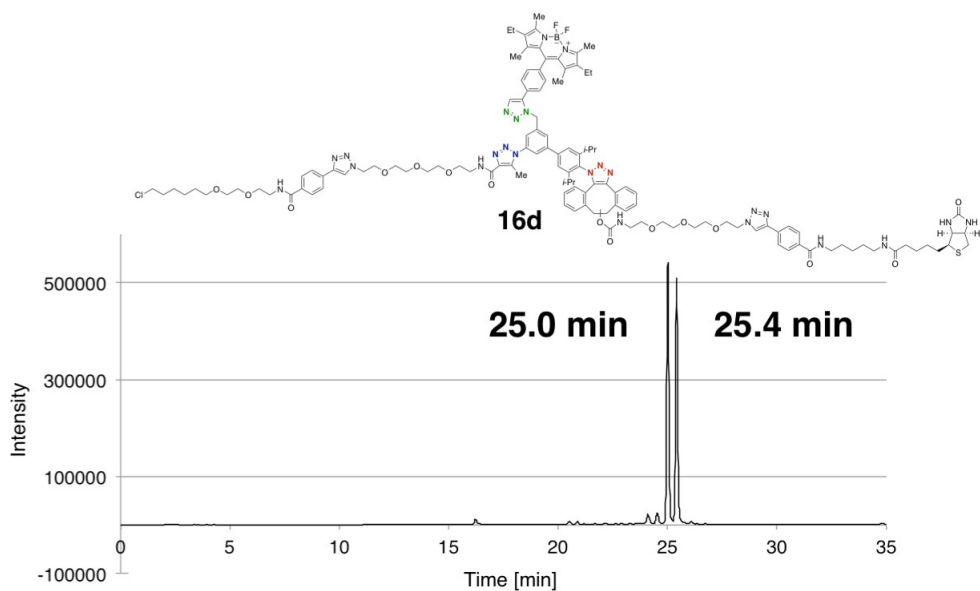
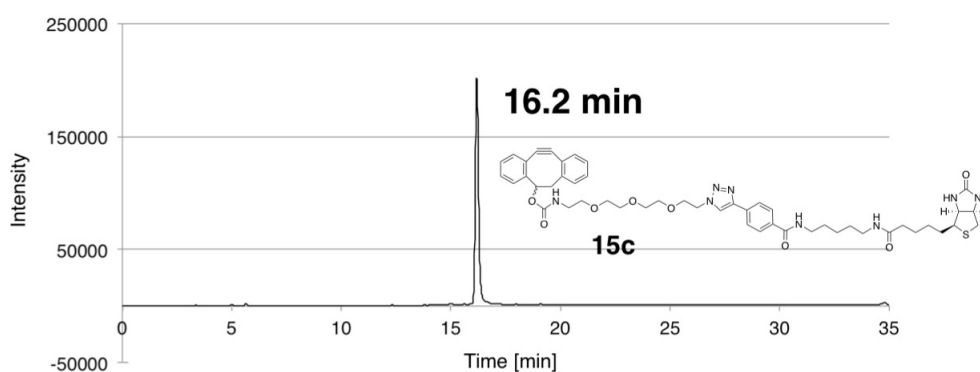
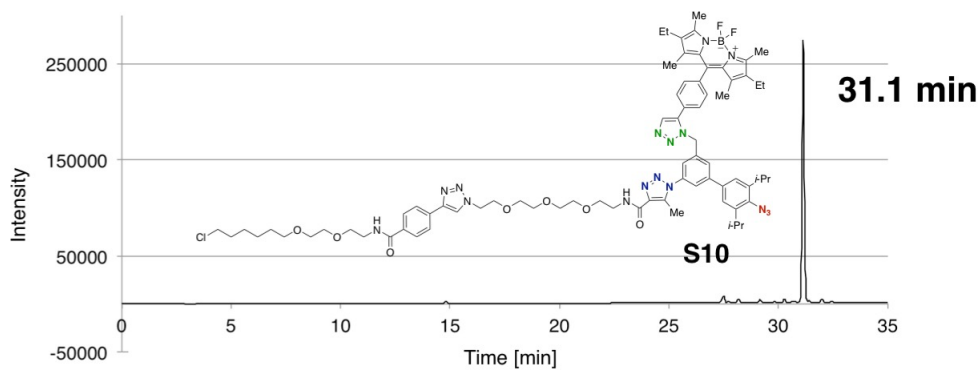
*Ru-catalyzed cycloaddition at the aliphatic azido group of diazide **S9** with terminal alkyne **14***

To a solution of diazide **S9** (31.6 mg, 31.2 μmol , 1.00 equiv) dissolved in toluene (0.30 mL) were added [3-ethyl-5-[(4-ethyl-3,5-dimethyl-2*H*-pyrrol-2-ylidene- κN)(4-ethynylphenyl)methyl]-2,4-dimethyl-1*H*-pyrrolato- κN]difluoroboron (**14**) (18.0 mg, 44.5 μmol , 1.43 equiv) and (pentamethylcyclopentadieny)bis-(triphenylphosphine)ruthenium(II) chloride (1.1 mg, 2.9 μmol , 9.3 mol %) at room temperature and the mixture was heated at 80 $^{\circ}\text{C}$ with stirring for 2 h. After cooling to room temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 5 g, CH_2Cl_2 only; then $\text{CH}_2\text{Cl}_2/\text{MeOH} = 10/1$) to give bistriazole **S10** (41.0 mg, 29.0 μmol , 92.7%) as a pink solid.



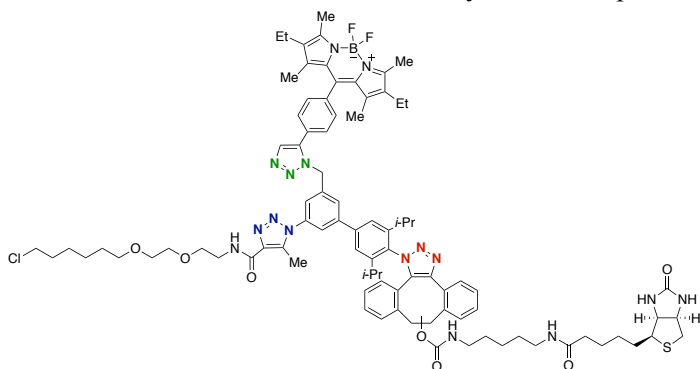
*Strain-promoted cycloaddition of azide **S10** with cyclooctyne **15c***

To a solution of azide **S10** (12.0 mg, 8.48 μmol , 1.1 equiv) dissolved in dichloromethane (0.50 mL) and methanol (0.50 mL) was added cyclooctyne **15c** (5.0 mg, 8.1 μmol , 1.0 equiv) at room temperature. After stirring for 2 h at the same temperature, the mixture was concentrated under reduced pressure. The residue was purified by flash column chromatography (silica-gel 2 g, CH_2Cl_2 only; then $\text{CH}_2\text{Cl}_2/\text{MeOH} = 20/1$ to 5/1) to give tris triazole **16d** (15.5 mg, 7.6 μmol , 94%) as a pink solid.

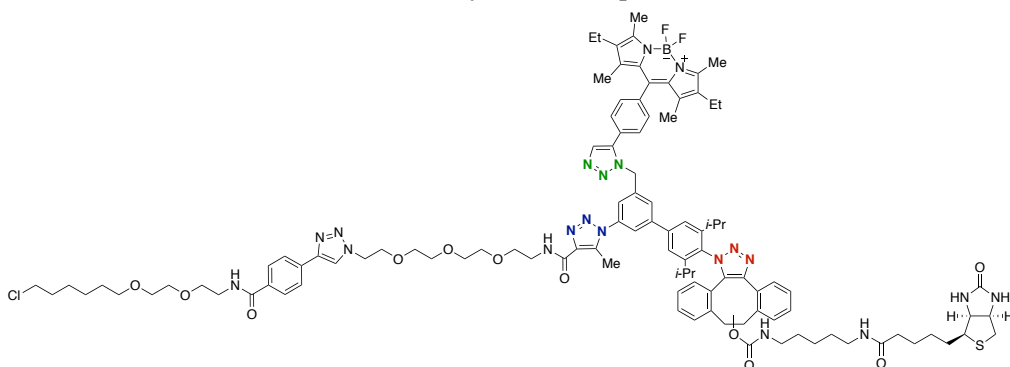


Similarly, trifunctional probe candidates **16a–c** and three kinds of bisfunctional tri-triazoles **S11–S13** for control experiment were prepared from triazide **11** according to the general procedures **A–C**.

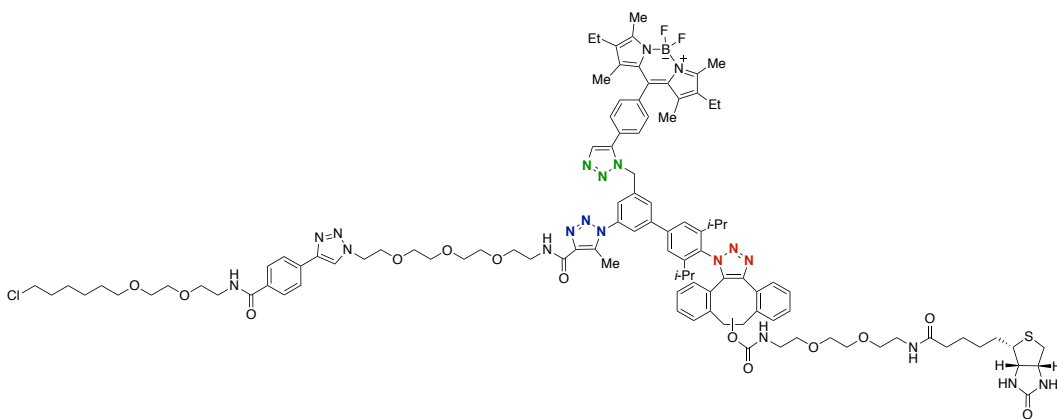
16a: 11 + 13a + 14 + 15a: 72% overall yield in 3 steps.



16b: 11 + 13b + 14 + 15a: 75% overall yield in 3 steps.



16c: 11 + 13b + 14 + 15b: 71% overall yield in 3 steps.



[illegible][illegible]

Biological Experiments

Production of recombinant GST-HaloTag protein in *E. coli*.

Escherichia coli strain Rosetta (DE3) pLysS cells (Millipore, Merck Chemicals Ltd., Nottingham, England) were transformed with pGEX6P-1-HaloTag vector,^{S11} and cultured in LB media containing 50 mg·L⁻¹ carbenicillin (Nacalai Tesque, Kyoto, Japan) and 34 mg·L⁻¹ chloramphenicol (Wako Pure Chemical Industries, Ltd., Osaka, Japan). Expression was induced by the addition of isopropyl β-D-thiogalactopyranoside (IPTG) (final concentration at 0.2 mM) (Nacalai Tesque), when the culture had reached an OD₆₀₀ of approximately 0.8. At the same time, the culture without IPTG was also prepared. After induction for 16 h at 37 °C, the cells were collected by centrifugation at 5000 g for 10 min, and suspended in phosphate buffered saline (PBS). The suspended cells were frozen in liquid N₂, and stored at -80 °C until use. After thawing on ice, Triton X-100 (final concentration at 1%) was added to the cell lysate, which were then incubated on ice for 20 min. MgCl₂ (final concentration at 2 mM) and DNase I (final concentration of approximately 10 μg·mL⁻¹) were added to the cell lysate, and incubation was continued at 4 °C for 20 min with gentle agitation. Cell debris and larger particles were removed by centrifugation at 12,000 g for 30 min, and the supernatant was then filtered through a 0.45-μm filter. The filtrated supernatants were frozen in liquid N₂, and stored at -80 °C until use for the following labeling experiments.

Chemical modification of the HaloTag protein in the crude extracts.

Into five hundred microlitter of the filtrated supernatants in a 1.5 mL-tube, five microlitter of the indicated compounds (10 mM stock in DMSO) were added, and immediately mix by vortex for 10 sec to be the final concentration of 100 μM. The solvent DMSO was used as a negative control. The mixtures were rotated gently in a dark room at room temperature for 16 h. Twenty-five microlitter of this reacted mixtures was diluted with equal volume of 2× SDS sample loading buffer (0.12 M Tris-HCl, pH 6.8, containing 3.4% SDS, 10% glycerol, and 20 mM DTT; Nacalai tesque), heated at 98 °C for 10 min.

SDS-PAGE analysis was carried out under reducing conditions using a 5–10% gradient separation gel (ATTO Corporation, Tokyo, Japan). The gels were directly visualized by laser-scanning with excitation by 488-nm wavelength of the blue laser followed by detecting emission light through 520-nm band-pass filter (520 BP 40) in a fluorescence imaging analyzer Typhoon 9410 (GE Healthcare), and then stained with Coomassie brilliant blue (CBB) EzStain AQua (ATTO Corporation). Images of the stained gels were taken by a flathead scanner GT-X970 (Seiko Epson Corporation, Japan).

The separated proteins in the gels were electrically transferred onto PVDF membranes in Mini Trans-Blot Cell (Bio-Rad Laboratories, Inc., Japan). The membranes were immersed in Blocking One solution (Nacalai Tesque), and then incubated with horseradish peroxidase-conjugated streptavidin (HRP-streptavidin) (Kirkegaard & Perry Laboratories, Meryland, USA) diluted in 1% Blocking One/Tris-based saline containing 0.1% Tween 20 (TBST) at 4 °C for 16 h. The membranes were extensively washed with TBST, and then reacted with ECL Western Blotting Detection Reagents (GE Healthcare). Luminescence signals were imaged on ChemiDox XRS Imaging System (Bio-Rad).

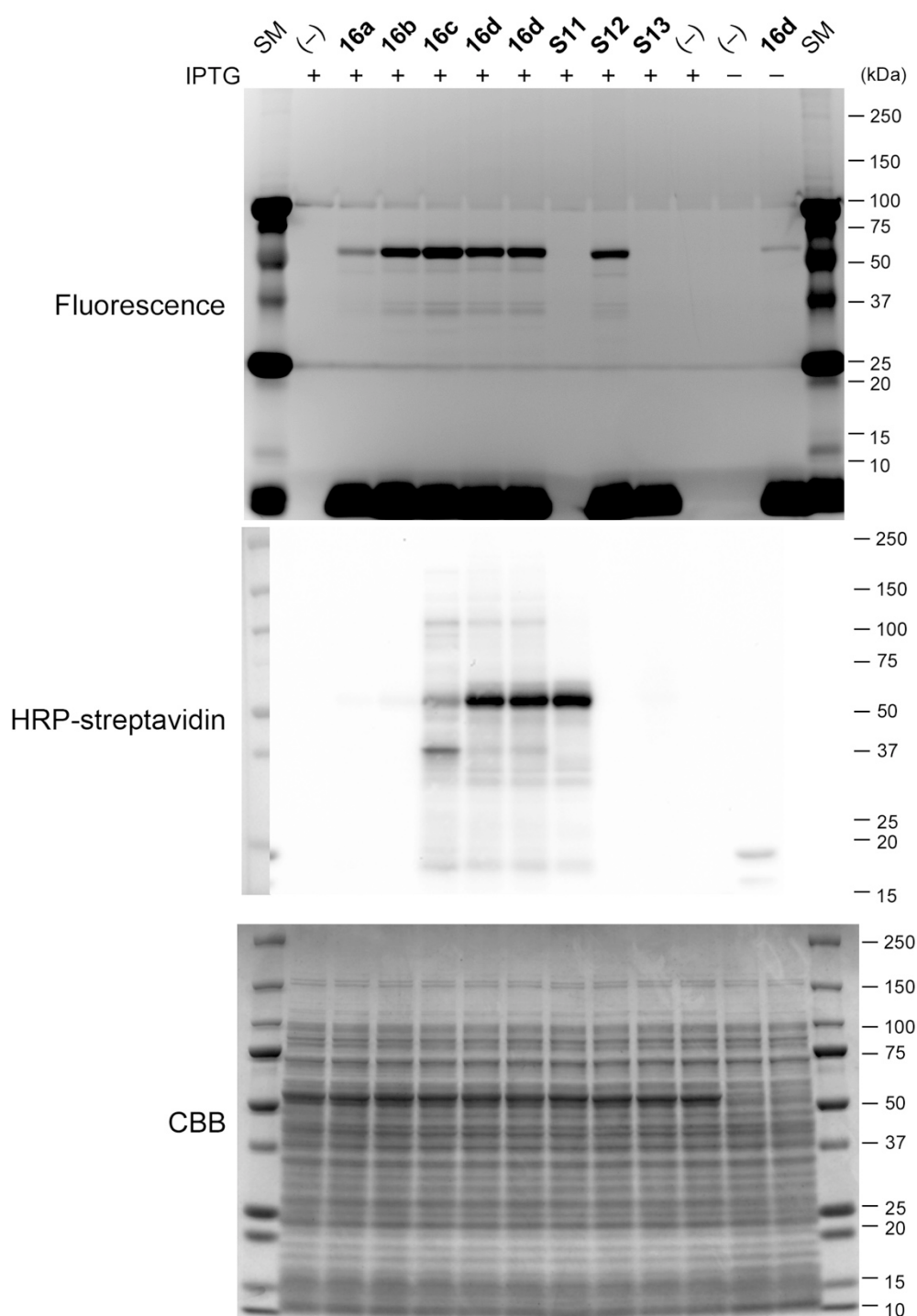


Figure S1. Chemical modification of the HaloTag protein by probe candidates **16a–d** and **S11–S13**.

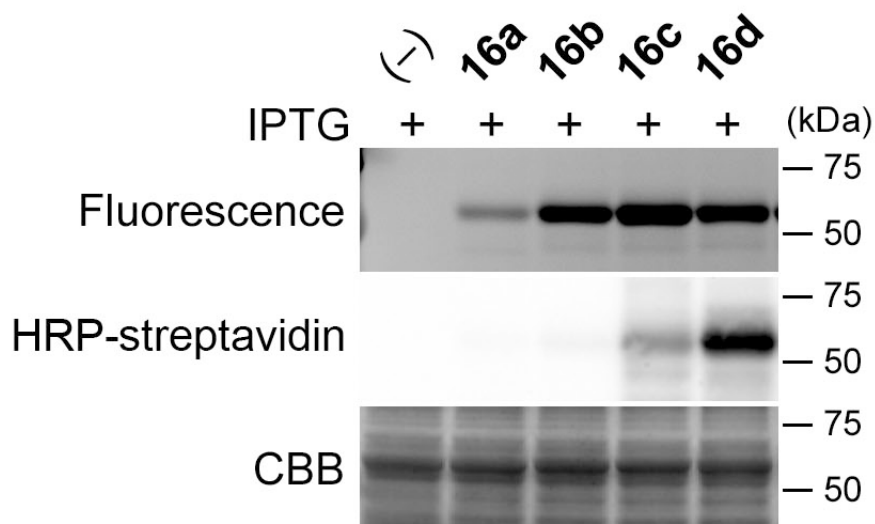


Figure S2. Chemical modification of the HaloTag protein by trifunctional probe candidates **16a**–**d** (focused Figure S1).

The results clearly show that each function of trifunctional probe **16d**, chemical modification of HaloTag protein (59 kDa) and detection of fluorescence and biotin, worked efficiently as expected. On the other hand, in the case of using **S11** without the HaloTag ligand moiety, neither fluorescence nor biotin was detected. When **S12** without the biotin moiety was used for the chemical modification, fluorescent labeling of HaloTag protein was achieved, but detection by HRP-streptavidin failed. Chemical modification by **S13** without the BODIPY moiety also resulted in a single-labeling; although modified HaloTag protein was detected by using HRP-streptavidin, fluorescence was not detected.

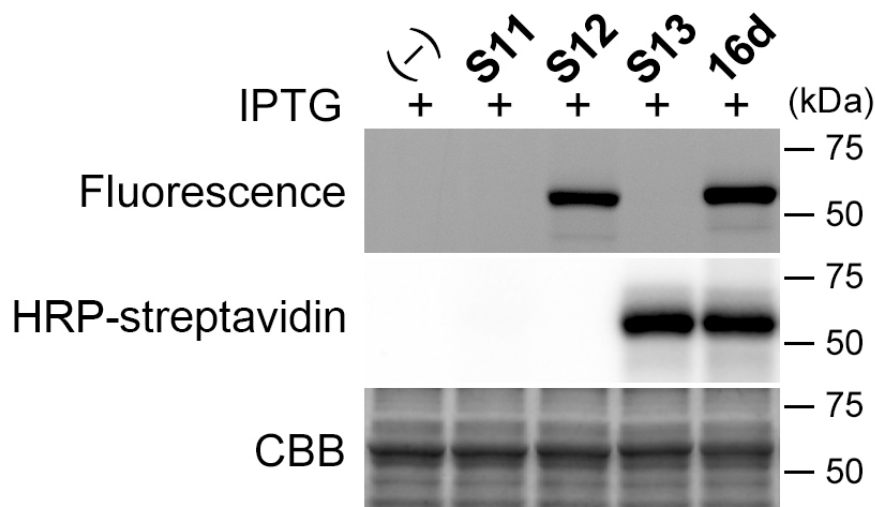
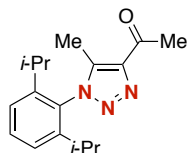


Figure S3. Chemical modification of the HaloTag protein by trifunctional probe candidate **16d** and bisfunctional tristriazoles **S11**–**S13** (focused Figure S1).

Characterization Data of New Compounds

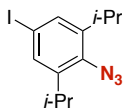
1-(2,6-Diisopropylphenyl)-8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazole (**3a**),^{S12} 1-phenyl-8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazole (**3b**),^{S12} 1-benzyl-8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazole (**3c**),^{S13} 1-(2,6-diisopropylphenyl)-4-phenyl-1*H*-[1,2,3]-triazole (**5a**),^{S14} 1,4-diphenyl-1*H*-[1,2,3]triazole (**5b**),^{S15} 1-benzyl-4-phenyl-1*H*-[1,2,3]triazole (**5c**),^{S16} 1-(2,6-diisopropylphenyl)-5-phenyl-1*H*-[1,2,3]triazole (**6a**),^{S12} 1,5-diphenyl-1*H*-[1,2,3]triazole (**6b**),^{S12} 1-benzyl-4-phenyl-1*H*-[1,2,3]triazole (**6c**),^{S17} 1-(1-phenyl-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**8b**),^{S18} and 1-(1-benzyl-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**8c**)^{S19} showed identical spectra with those reported in the literature.

1-(1-(2,6-Diisopropylphenyl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**8a**)



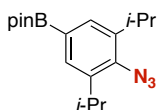
Colorless solid; Mp 115–118 °C; TLC R_f = 0.42 (*n*-hexane/EtOAc = 5/1); ¹H NMR (500 MHz, CDCl₃) δ 1.12 (d, 6H, J = 7.5 Hz), 1.13 (d, 6H, J = 7.5 Hz), 2.07 (tt, 2H, J = 7.5, 7.5 Hz), 2.35 (s, 3H), 2.80 (s, 3H), 7.33 (d, 2H, J = 7.5 Hz), 7.54 (t, 1H, J = 7.5 Hz); ¹³C NMR (126 MHz, CDCl₃) δ 9.63 (1C), 23.1 (2C), 24.6 (2C), 27.7 (1C), 28.5 (2C), 124.2 (2C), 130.5 (1C), 131.3 (1C), 139.0 (1C), 143.0 (1C), 146.2 (2C), 194.6 (1C); IR (KBr, cm⁻¹) 763, 952, 1275, 1369, 1477, 1556, 1685, 2967; HRMS (ESI⁺) m/z 308.1729 ([M+Na]⁺, C₁₇H₂₃N₃NaO⁺ requires 308.1733).

4-Iodo-2,6-diisopropylphenyl azide (**S1**)



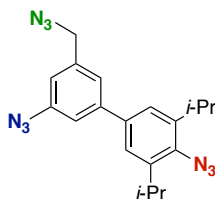
Pale brown oil; TLC R_f = 0.65 (*n*-hexane); ¹H NMR (400 MHz, CDCl₃) δ 1.24 (d, 12H, J = 6.8 Hz), 3.28 (sept, 2H, J = 6.8 Hz), 7.42 (s, 2H); ¹³C NMR (126 MHz, CDCl₃) δ 23.3 (4C), 28.7 (2C), 92.3 (1C), 133.2 (2C), 135.3 (1C), 145.5 (2C); IR (KBr, cm⁻¹) 865, 1071, 1234, 1332, 1438, 1565, 2102, 2124, 2930, 2964; Anal. calcd. for C₁₂H₁₆IN₃: C, 43.78; H, 4.90; N, 12.77%; Found: C, 43.61; H, 5.20; N, 12.53%.

2-(4-Azido-3,5-diisopropylphenyl)-4,4,5,5-tetramethyl-1,3,2-dioxaborolane (**10**)



Pale brown oil; TLC R_f = 0.53 (*n*-hexane/EtOAc = 10/1); ¹H NMR (400 MHz, CDCl₃) δ 1.29 (d, 12H, J = 6.8 Hz), 1.34 (s, 12H), 3.35 (sept, 2H, J = 6.8 Hz), 7.57 (s, 2H); ¹³C NMR (126 MHz, CDCl₃) δ 23.5 (4C), 24.8 (4C), 28.8 (2C), 83.8 (2C), 130.6 (2C), 138.0 (1C), 142.2 (2C). The signal for the carbon which is attached to the boron atom was not observed; IR (KBr, cm⁻¹) 692, 852, 966, 1146, 1378, 1463, 1601, 2117, 2933, 2967; Anal. calcd. for C₁₈H₂₈BN₃O₂: C, 65.66; H, 8.57; N, 12.76%; Found: C, 65.67; H, 8.86; N, 12.83%.

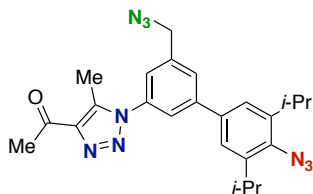
3',4-Diazido-5'-azidomethyl-3,5-diisopropyl-1,1'-biphenyl (**11**)



Pale brown oil; TLC R_f = 0.14 (*n*-hexane); ¹H NMR (400 MHz, CDCl₃) δ 1.32 (d, 12H), 3.41 (sept, 2H, J = 6.8 Hz), 4.42 (s, 2H), 6.98 (dd, 1H, J = 1.6, 1.6 Hz), 7.13 (dd, 1H, J = 1.6, 1.6 Hz), 7.23 (s, 1H), 7.28 (s, 2H);

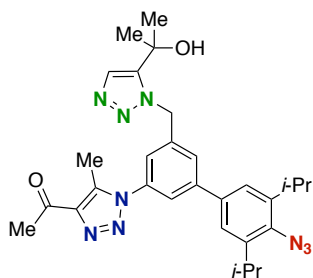
^{13}C NMR (126 MHz, CDCl_3) δ 23.5 (4C), 28.9 (2C), 54.3 (1C), 117.1 (1C), 117.7 (1C), 122.9 (2C), 123.4 (1C), 135.4 (1C), 137.9 (1C), 138.2 (1C), 141.1 (1C), 143.7 (1C), 143.7 (2C); IR (KBr, cm^{-1}) 852, 1292, 1335, 1439, 1593, 2108, 2871, 2966; Anal. calcd. for $\text{C}_{19}\text{H}_{21}\text{N}_9$: C, 60.78; H, 5.64; N, 33.58%; Found: C, 61.02; H, 5.94; N, 33.42%.

1-(1-(4'-Azido-5-azidomethyl-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**S2**)



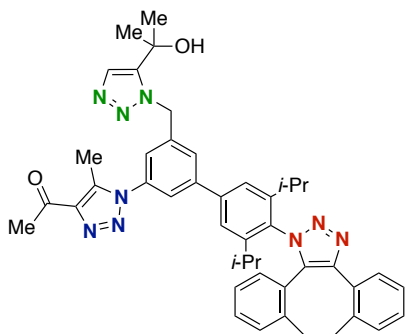
Pale yellow solid; Mp 116–118 °C; TLC R_f = 0.40 (*n*-hexane/EtOAc = 3/1); ^1H NMR (400 MHz, CDCl_3) δ 1.33 (d, 12H, J = 6.8 Hz), 2.65 (s, 3H), 2.78 (s, 3H), 3.43 (sept, 2H, J = 6.8 Hz), 4.57 (s, 2H), 7.32 (s, 2H), 7.39 (dd, 1H, J = 1.2, 1.2 Hz), 7.58 (dd, 1H, J = 1.2, 1.2 Hz), 7.65 (dd, 1H, J = 1.2, 1.2 Hz); ^{13}C NMR (126 MHz, CDCl_3) δ 10.3 (1C), 23.5 (4C), 27.9 (1C), 28.9 (2C), 54.0 (1C), 123.0 (2C), 123.1 (1C), 123.8 (1C), 128.1 (1C), 135.8 (1C), 136.2 (1C), 137.3 (1C), 137.6 (1C), 138.1 (1C), 143.79 (1C), 143.84 (1C), 144.0 (2C), 194.4 (1C); IR (KBr, cm^{-1}) 866, 953, 1076, 1337, 1443, 1557, 1597, 1682, 2101, 2965, 3368; Anal. calcd. for $\text{C}_{24}\text{H}_{27}\text{N}_9\text{O}$: C, 63.00; H, 5.95; N, 27.55%; Found: C, 62.96; H, 5.68; N, 27.33%.

1-(1-(4'-Azido-5-((5-(2-hydroxypropan-2-yl)-1*H*-1,2,3-triazol-1-yl)methyl)-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**S3**)



Pale yellow solid; Mp 78 °C (dec.); TLC R_f = 0.47 (EtOAc); ^1H NMR (400 MHz, CDCl_3) δ 1.31 (d, 12H, J = 6.8 Hz), 1.63 (s, 6H), 2.55 (s, 3H), 2.65–2.69 (br, 1H), 2.73 (s, 3H), 3.41 (sept, 2H, J = 6.8 Hz), 5.99 (s, 2H), 7.18 (s, 1H), 7.28 (s, 2H), 7.45 (s, 1H), 7.53 (s, 1H), 7.65 (s, 1H); ^{13}C NMR (126 MHz, CDCl_3) δ 10.2 (1C), 23.5 (4C), 27.8 (1C), 28.9 (2C), 30.9 (2C), 52.1 (1C), 68.0 (1C), 122.7 (1C), 122.9 (2C), 123.4 (1C), 127.8 (1C), 130.9 (1C), 135.7 (1C), 135.9 (1C), 137.2 (1C), 137.5 (1C), 138.9 (1C), 143.1 (1C), 143.5 (2C), 143.6 (1C), 143.9 (1C), 194.2 (1C); IR (KBr, cm^{-1}) 732, 1175, 1464, 1558, 1683, 2123, 2966, 3322; Anal. calcd. for $\text{C}_{29}\text{H}_{35}\text{N}_9\text{O}_2$: C, 64.31; H, 6.51; N, 23.27%; Found: C, 64.09; H, 6.78; N, 23.08%.

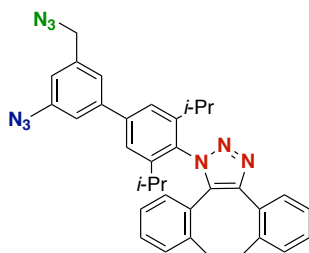
1-(1-(4'-(8,9-dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazol-1-yl)-5-((5-(2-hydroxypropan-2-yl)-1*H*-1,2,3-triazol-1-yl)methyl)-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**12a**)



Colorless solid; Mp 145 °C (dec); TLC R_f = 0.53 (EtOAc); ^1H NMR (400 MHz, CDCl_3) δ 0.67–1.27 (br,

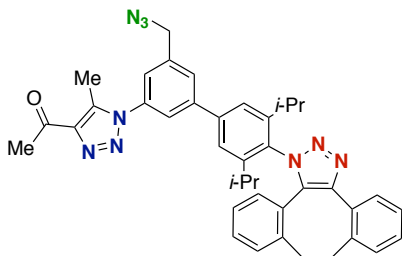
12H), 1.57 (s, 6H), 2.31–2.45 (br, 2H), 2.49 (s, 3H), 2.66 (s, 3H), 2.78 (s, 1H), 2.99–3.14 (br, 2H), 3.24–3.39 (br, 2H), 5.92 (s, 2H), 6.65 (dd, 1H, $J = 0.8, 7.6$ Hz), 6.90 (ddd, 1H, $J = 0.8, 7.6, 7.6$ Hz), 7.12–7.20 (m, 5H), 7.24–7.29 (m, 3H), 7.39 (s, 1H), 7.52 (dd, 1H, $J = 1.6, 1.6$ Hz), 7.59–7.61 (m, 1H), 7.65 (s, 1H); ^{13}C NMR (126 MHz, CDCl_3) δ 10.2 (1C), 22.5 (2C), 25.4 (2C), 27.9 (1C), 28.9 (2C), 30.9 (2C), 32.7 (1C), 36.8 (1C), 52.0 (1C), 68.0 (1C), 122.9 (2C), 123.2 (1C), 123.8 (1C), 125.7 (1C), 126.0 (1C), 126.2 (1C), 128.0 (1C), 128.1 (1C+1C, two signals overlapped), 128.7 (1C), 129.4 (1C), 129.7(1C), 129.9 (1C), 130.91 (1C), 130.94 (1C), 132.3 (1C), 132.5 (1C), 135.1 (1C), 136.0 (1C), 137.4 (1C), 137.6 (1C), 139.1 (1C), 141.0 (1C), 141.4 (1C), 143.1 (2C), 143.7 (1C), 146.2 (1C), 147.3 (1C), 194.2 (1C); IR (KBr, cm^{-1}) 731, 910, 1096, 1368, 1557, 1682, 2965, 3300; HRMS (ESI^+) m/z 746.3901 ($[\text{M}+\text{H}]^+$, $\text{C}_{45}\text{H}_{48}\text{N}_9\text{O}_2^+$ requires 746.3925).

1-(3'-Azido-5'-azidomethyl-3,5-diisopropyl-1,1'-biphenyl-4-yl)-8,9-dihydro-1H-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazole (**S4**)



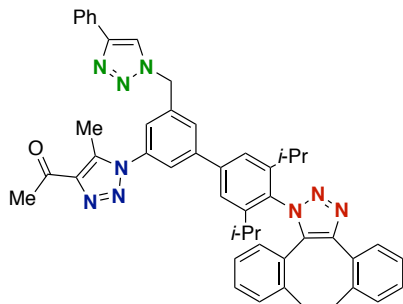
Colorless solid; Mp 155 °C (dec.); TLC $R_f = 0.60$ (n -hexane/EtOAc = 3/1); ^1H NMR (400 MHz, CDCl_3) δ 0.78–1.34 (br, 12H), 2.39–2.53 (br, 2H), 3.12–3.21 (br, 2H), 3.35–3.51 (br, 2H), 4.44 (s, 2H), 6.74 (dd, 1H, $J = 1.2, 8.0$ Hz), 6.96 (ddd, 1H, $J = 1.2, 7.6, 7.6$ Hz), 7.02 (s, 1H), 7.16–7.35 (m, 9H), 7.68–7.71 (m, 1H); ^{13}C NMR (126 MHz, CDCl_3) δ 22.6 (1C), 25.5 (2C), 28.9 (2C), 32.7 (2C), 36.9 (1C), 54.3 (1C), 117.6 (1C), 117.9 (1C), 122.9 (2C), 123.5 (1C), 125.8 (1C), 126.0 (1C+1C, two signals overlapped), 126.1 (1C), 128.1 (1C), 128.8 (1C), 129.6 (2C), 129.9 (1C), 130.9 (1C), 131.9 (1C), 132.6 (1C), 135.1 (1C), 137.4 (1C), 138.0 (1C), 141.2 (1C), 141.5 (1C), 141.9 (1C), 143.3 (1C), 146.2 (1C), 147.0 (br, 1C); IR (KBr, cm^{-1}) 732, 1274, 1354, 1454, 1597, 2108, 2871, 2931, 2965; Anal. calcd. for $\text{C}_{35}\text{H}_{33}\text{N}_9$: C, 72.52; H, 5.74; N, 21.75%; Found: C, 72.27; H, 6.03; N, 21.45%.

1-(5-Azidomethyl-4'-(8,9-dihydro-1H-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazol-1-yl)-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-5-methyl-1H-1,2,3-triazol-4-yl)ethanone (**S5**)



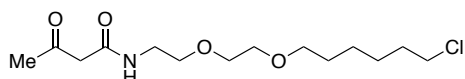
Colorless solid; Mp 218 °C (dec.); TLC $R_f = 0.45$ (n -hexane/EtOAc = 2/1); ^1H NMR (400 MHz, CDCl_3) δ 0.84–1.36 (br, 12H), 2.42–2.55 (br, 2H), 2.67 (s, 3H), 2.78 (s, 3H), 3.09–3.23 (br, 2H), 3.33–3.46 (br, 2H), 4.58 (s, 2H), 6.73 (dd, 1H, $J = 1.2, 7.6$ Hz), 6.96 (ddd, 1H, $J = 1.2, 7.6, 7.6$ Hz), 7.20–7.42 (m, 8H), 7.64 (dd, 1H, $J = 1.2, 1.2$ Hz), 7.68–7.71 (m, 2H); ^{13}C NMR (126 MHz, CDCl_3) δ 10.3 (1C), 22.5 (2C), 25.4 (2C), 27.9 (1C), 28.9 (2C), 32.7 (1C), 36.8 (1C), 54.0 (1C), 123.0 (2C), 123.5 (1C), 124.1 (1C), 125.8 (1C), 126.0 (1C), 126.1 (1C), 128.1 (1C), 128.3 (1C), 128.7 (1C), 129.5 (1C), 129.6 (1C), 129.9 (1C), 130.9 (2C), 132.4 (1C), 132.6 (1C), 135.1 (1C), 136.2 (1C), 137.4 (1C), 137.6 (1C), 138.2 (1C), 141.0 (1C), 141.5 (1C), 143.5 (1C), 143.8 (1C), 146.2 (1C), 147.4 (br, 1C), 194.3 (1C); IR (KBr, cm^{-1}) 733, 908, 1292, 1341, 1439, 1593, 2110, 2928, 2963; HRMS (ESI^+) m/z 662.3353 ($[\text{M}+\text{H}]^+$, $\text{C}_{40}\text{H}_{40}\text{N}_9\text{O}^+$ requires 662.3350).

1-(1-(4'-(8,9-Dihydro-1*H*-dibenzo[3,4:7,8]cycloocta[1,2-*d*][1,2,3]triazol-1-yl)-3',5'-diisopropyl-5-((4-phenyl-1*H*-1,2,3-triazol-1-yl)methyl)-1,1'-biphenyl)-3-yl-5-methyl-1*H*-1,2,3-triazol-4-yl)ethanone (**12b**)



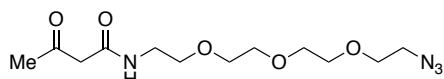
Colorless solid; Mp 144–146 °C; TLC R_f = 0.22 (*n*-hexane/EtOAc = 1/1); ^1H NMR (400 MHz, CDCl_3) δ 0.85–1.34 (br, 12H), 2.41–2.53 (br, 2H), 2.61 (s, 3H), 2.76 (s, 3H), 3.11–3.23 (br, 2H), 3.36–3.45 (br, 2H), 5.78 (s, 2H), 6.72 (dd, 1H, J = 0.8, 7.6 Hz), 6.96 (ddd, 1H, J = 1.2, 7.6, 7.6 Hz), 7.19–7.44 (m, 11H), 7.65–7.72 (m, 3H), 7.81–7.84 (m, 3H); ^{13}C NMR (126 MHz, CDCl_3) δ 10.3 (1C), 22.5 (2C), 25.5 (2C), 27.9 (1C), 28.9 (2C), 32.7 (1C), 36.8 (1C), 53.4 (1C), 119.7 (1C), 123.0 (2C), 123.4 (1C), 124.6 (1C), 125.69 (1C), 125.73 (2C), 126.0 (1C+1C, two signals overlapped), 126.1 (1C), 128.1 (1C), 128.2 (1C), 128.5 (1C), 128.7 (1C), 128.9 (2C), 129.4 (1C), 129.7 (1C), 129.9 (1C), 130.0 (1C), 130.9 (1C), 132.50 (1C), 132.55 (1C), 135.1 (1C), 136.5 (1C), 137.4 (1C), 137.48 (1C), 137.50 (1C), 140.7 (1C), 141.5 (1C), 143.8 (2C), 146.2 (1C), 147.4 (1C), 148.7 (1C), 194.2 (1C); IR (KBr, cm^{-1}) 763, 1074, 1366, 1464, 1557, 1682, 2962, 3054; HRMS (ESI^+) m/z 764.3792 ($[\text{M}+\text{H}]^+$, $\text{C}_{48}\text{H}_{46}\text{N}_9\text{O}^+$ requires 764.3820).

N-(2-(2-((6-Chlorohexyl)oxy)ethoxy)ethyl)-3-oxobutanamide (**13a**)



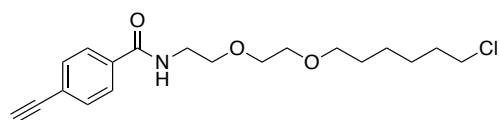
Pale yellow oil; TLC R_f = 0.25 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 10/1); ^1H NMR (400 MHz, CDCl_3) δ 1.41–1.48 (m, 4H), 1.58–1.65 (m, 2H), 1.75 (m, 2H), 2.27 (s, 3H), 3.40 (s, 2H), 3.46–3.63 (m, 12H), 7.06 (br s, 1H); ^{13}C NMR (126 MHz, CDCl_3) δ 25.4 (1C), 26.7 (1C), 29.4 (1C), 30.9 (1C), 32.5 (1C), 39.3 (1C), 45.0 (1C), 50.0 (1C), 69.6 (1C), 70.0 (1C), 70.4 (1C), 71.3 (1C), 165.5 (1C), 204.0 (1C); IR (KBr, cm^{-1}) 1119, 1358, 1547, 1649, 1719, 2862, 2936, 3308; HRMS (ESI^+) m/z 308.1626 ($[\text{M}+\text{H}]^+$, $\text{C}_{14}\text{H}_{27}^{35}\text{ClNO}_4^+$ requires 308.1623).

N-(2-(2-(2-(2-Azidoethoxy)ethoxy)ethoxy)ethyl)-3-oxobutanamide (**S6**)



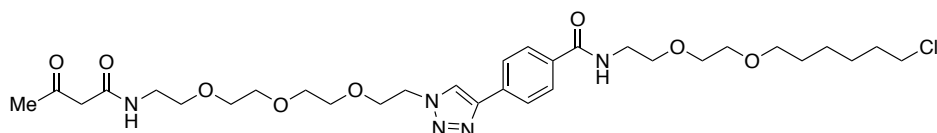
Pale yellow oil; TLC R_f = 0.70 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 10/1); ^1H NMR (400 MHz, CDCl_3) δ 2.28 (s, 3H), 3.32–3.34 (m, 4H), 3.39–3.42 (m, 2H), 3.50–3.52 (m, 2H), 3.56–3.63 (m, 10H), 7.15 (br s, 1H); ^{13}C NMR (126 MHz, CDCl_3) δ 30.7 (1C), 39.2 (1C), 50.1 (1C), 50.6 (1C), 69.5 (1C), 69.9 (1C), 70.2 (1C), 70.49 (1C), 70.52 (1C), 70.6 (1C), 165.6 (1C), 203.9 (1C); IR (KBr, cm^{-1}) 1118, 1288, 1535, 1649, 1716, 2108, 2870, 3066, 3305; HRMS (ESI^-) m/z 301.1510 ($[\text{M}-\text{H}]^-$, $\text{C}_{12}\text{H}_{21}\text{N}_4\text{O}_5^-$ requires 301.1517).

N-(2-(2-((6-Chlorohexyl)oxy)ethoxy)ethyl)-4-ethynylbenzamide (**S7**)



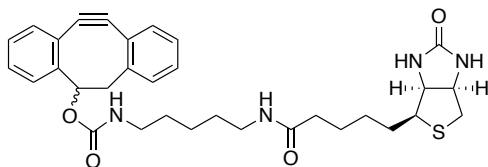
Colorless solid; TLC R_f = 0.27 (*n*-hexane/EtOAc = 1/1); ^1H NMR (400 MHz, CDCl_3) δ 1.29–1.45 (m, 4H), 1.53–1.59 (m, 2H), 1.70–1.76 (m, 2H), 3.21 (s, 1H), 3.44–3.68 (m, 12H), 6.82 (br s, 1H), 7.53 (d, 2H, J = 8.0 Hz), 7.75 (d, 2H, J = 8.0 Hz); ^{13}C NMR (126 MHz, CDCl_3) δ 25.2 (1C), 26.5 (1C), 29.3 (1C), 32.3 (1C), 39.6 (1C), 44.9 (1C), 69.4 (1C), 69.8 (1C), 70.1 (1C), 71.1 (1C), 79.4 (1C), 82.6 (1C), 125.1 (1C), 126.9 (2C), 132.0 (2C), 134.4 (1C), 166.5 (1C); IR (KBr, cm^{-1}) 768, 855, 1113, 1303, 1538, 1642, 2862, 2937, 3290; HRMS (ESI^+) m/z 374.1490 ($[\text{M}+\text{Na}]^+$, $\text{C}_{19}\text{H}_{26}^{35}\text{ClNNaO}_3^+$ requires 374.1493).

N-(2-(2-(((6-Chlorohexyl)oxy)ethoxy)ethyl)-4-(1-(13,15-dioxo-3,6,9-trioxa-12-azahexadecyl)-1*H*-1,2,3-triazol-4-yl)benzamide (**13b**)



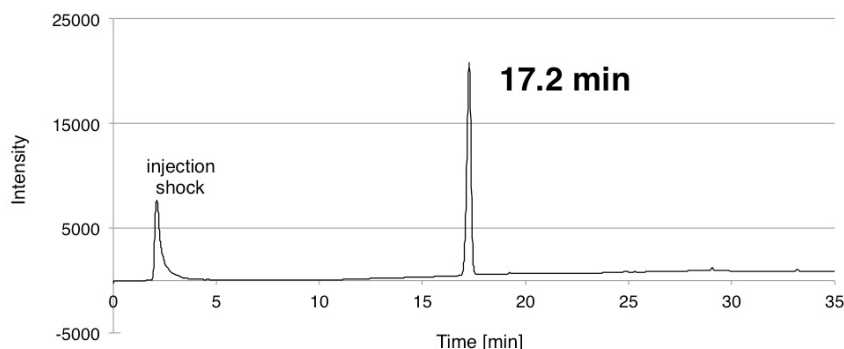
Colorless solid; Mp 79–81 °C; TLC R_f = 0.38 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 10/1); ^1H NMR (400 MHz, CDCl_3) δ 1.32–1.44 (m, 4H), 1.56–1.58 (m, 2H), 1.69–1.76 (m, 2H), 2.24 (s, 3H), 3.39–3.52 (m, 10H), 3.56–3.70 (m, 16H), 3.94 (t, 2H, J = 4.8 Hz), 4.62 (t, 2H, J = 4.8 Hz), 6.80–6.85 (br, 1H), 7.14–7.22 (br, 1H), 7.85 (d, 2H, J = 8.4 Hz), 7.92 (d, 2H, J = 8.4 Hz), 8.09 (s, 1H); ^{13}C NMR (126 MHz, CDCl_3) δ 25.4 (1C), 26.7 (1C), 29.4 (1C), 30.9 (1C), 32.5 (1C), 39.2 (1C), 39.7 (1C), 45.0 (1C), 50.0 (1C), 50.4 (1C), 69.4 (1C), 69.6 (1C), 69.7 (1C), 70.0 (1C), 70.2 (1C), 70.3 (1C), 70.5 (1C), 70.5 (1C), 70.6 (1C), 71.3 (1C), 121.7 (1C), 125.6 (2C), 127.7 (2C), 133.7 (1C), 133.9 (1C), 146.7 (1C), 165.7 (1C), 167.0 (1C), 204.2 (1C); IR (KBr, cm^{-1}) 1145, 1302, 1356, 1535, 1647, 1719, 2864, 2928, 3308; Anal. calcd. for $\text{C}_{31}\text{H}_{48}^{35}\text{ClN}_5\text{O}_8$: C, 56.91; H, 7.40; N, 10.71%; Found: C, 57.06; H, 7.69; N, 10.45%.

Cyclooctyne **15a** bearing a biotin moiety conjugated with a pentamethylene linker

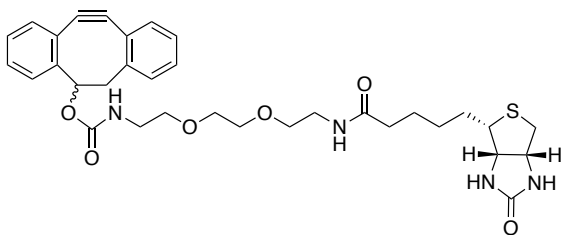


Colorless solid; Mp 112–115 °C; TLC R_f = 0.25 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 10/1); HPLC analysis: the diastereomers did not separate and observed as a single peak with R_t = 17.2 min [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. $\times$ 250 mm); mobile phase: $\text{CH}_3\text{CN}:\text{H}_2\text{O}$ = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm]; Cyclooctyne **15a** is a mixture of diastereomers and conformational isomers were observed by NMR analysis. For the major isomer: ^1H NMR (400 MHz, CDCl_3) δ 1.19–1.64 (m, 13H), 2.08–2.17 (m, 2H), 2.63–2.67 (m, 1H), 2.83–2.91 (m, 2H), 3.08–3.18 (m, 5H), 4.21–4.30 (br, 1H), 4.39–4.46 (br, 1H), 5.47 (s, 1H), 7.25–7.37 (m, 7H), 7.48–7.50 (m, 1H); ^{13}C NMR (126 MHz, CD_3OD) δ 25.2 (1C), 27.1 (1C), 29.6 (1C), 29.9 (1C), 30.1 (1C), 30.7 (1C), 36.9 (1C), 40.4 (1C), 41.2 (1C), 41.8 (1C), 47.3 (1C), 57.1 (1C), 61.7 (1C), 63.5 (1C), 78.0 (1C), 111.1 (1C), 114.0 (1C), 122.6 (1C), 125.07 (1C), 125.13 (1C), 127.1 (1C), 127.3 (1C), 128.4 (1C), 128.5 (1C), 129.4 (1C), 129.5 (1C), 131.2 (1C), 152.6 (1C), 153.9 (1C), 158.2 (1C), 166.2 (1C), 176.1 (1C); IR (KBr, cm^{-1}) 757, 1023, 1262, 1450, 1532, 1695, 1650, 2927, 3290; HRMS (ESI $^+$) m/z 575.2678 ($[\text{M}+\text{H}]^+$, $\text{C}_{32}\text{H}_{39}\text{N}_4\text{O}_4\text{S}^+$ requires 575.2687).

HPLC chart:

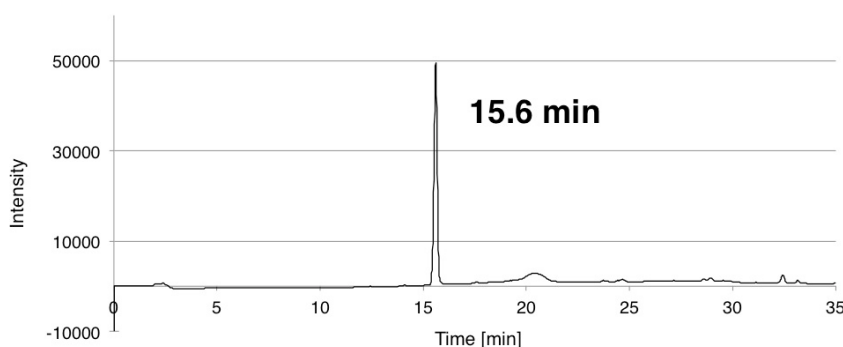


Cyclooctyne **15b** bearing a biotin moiety conjugated with a 3,6-dioxaoctamethylene (PEO₂) linker

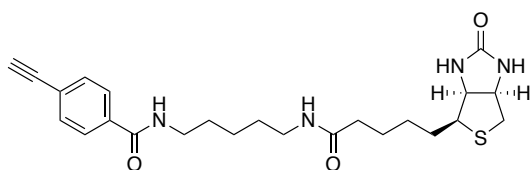


Colorless solid; Mp 80 °C (dec.); TLC R_f = 0.38 (CH₂Cl₂/MeOH = 10/1); HPLC analysis: the diastereomers did not separate and observed as a single peak with R_t = 15.6 min [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. × 250 mm); mobile phase: CH₃CN:H₂O = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm]; Cyclooctyne **15b** is a mixture of diastereomers and conformational isomers were observed by NMR analysis. For the major isomer: ¹H NMR (400 MHz, CD₃OD) δ 1.38–1.73 (m, 6H), 2.18 (t, 2H, J = 7.0 Hz), 2.66–2.69 (m, 1H), 2.81–2.90 (m, 2H), 3.14–3.80 (m, 14H), 4.22–4.24 (m, 1H), 4.43–4.45 (m, 1H), 5.42 (s, 1H), 7.32–7.58 (m, 8H); ¹³C NMR (126 MHz, CD₃OD) δ 26.9 (1C), 29.6 (1C), 29.8 (1C), 36.8 (1C), 40.4 (1C), 41.1 (1C), 41.9 (1C), 47.2 (1C), 57.1 (1C), 61.7 (1C), 63.4 (1C), 70.7 (1C), 71.0 (1C), 71.4 (1C), 71.44 (1C+1C, two signals overlapped), 78.1 (1C), 111.1 (1C), 113.9 (1C), 122.5 (1C), 125.1 (1C), 127.0 (1C), 127.3 (1C), 128.4 (1C), 128.5 (1C), 129.4 (1C), 129.4 (1C), 131.2 (1C), 152.6 (1C), 153.8 (1C), 158.1 (1C), 166.2 (1C), 176.2 (1C); IR (KBr, cm⁻¹) 763, 1038, 1139, 1264, 1428, 1694, 2472, 2928, 3268; HRMS (ESI⁺) m/z 643.2548 ([M+Na]⁺, C₃₃H₄₀N₄NaO₆S⁺ requires 643.2561).

HPLC chart:

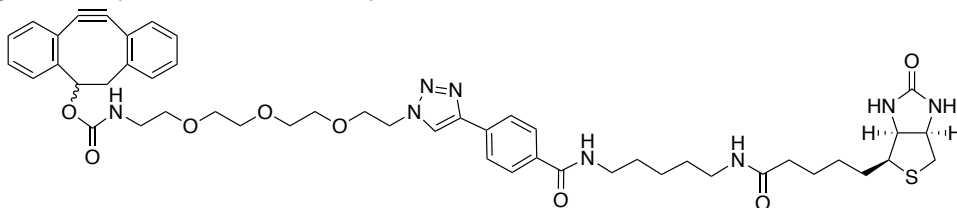


4-Ethynyl-*N*-(5-(5-((3*a*S,4*S*,6*a**R*)-2-oxohexahydro-1*H*-thieno[3,4-*d*]imidazol-4-yl)pentanamido)pentyl)benzamide (**S8**)



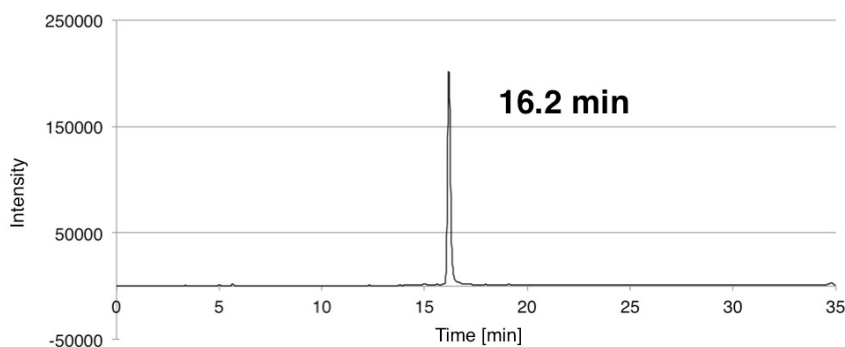
Colorless solid; Mp 170–172 °C; TLC R_f = 0.29 (CH₂Cl₂/MeOH = 10/1). ¹H NMR (400 MHz, CD₃OD) δ 1.40–1.44 (m, 4H), 1.54–1.78 (m, 8H), 2.18 (t, 2H, J = 7.6 Hz), 2.70 (d, 1H, J = 12.4 Hz), 2.92 (dd, 1H, J = 4.8, 12.4 Hz), 3.12–3.20 (m, 3H), 3.38 (t, 2H, J = 7.2 Hz), 3.67 (s, 1H), 4.28–4.31 (m, 1H), 4.47–4.50 (m, 1H), 7.53–7.58 (AA'BB', 2H), 7.78–7.82 (AA'BB', 2H); ¹³C NMR (126 MHz, CD₃OD) δ 25.4 (1C), 27.0 (1C), 29.6 (1C), 29.9 (1C), 30.2 (1C+1C, two signals overlapped), 36.9 (1C), 40.3 (1C), 41.0 (1C), 41.1 (1C), 57.1 (1C), 61.7 (1C), 63.5 (1C), 81.2 (1C), 83.7 (1C), 127.0 (1C), 128.5 (2C), 133.2 (2C), 135.9 (1C), 166.2 (1C), 169.4 (1C), 176.1 (1C); IR (KBr, cm⁻¹) 1323, 1465, 1546, 1638, 1696, 2930, 3290; HRMS (ESI⁺) m/z 457.2288 ([M+H]⁺, C₂₄H₃₃N₄O₃S⁺ requires 457.2268).

Cyclooctyne **15c** bearing a biotin moiety conjugated with 3,6,9-trioxaundecamethylene (PEO₃) and pentamethylene linkers linked by a 1,2,3-triazole skeleton

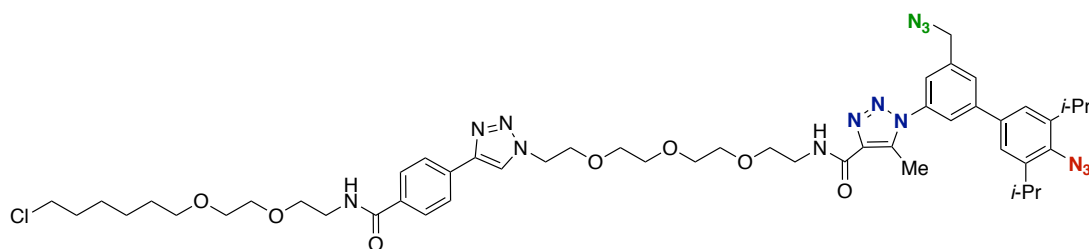


Colorless solid; Mp 131 °C (dec.); TLC R_f = 0.48 (CH₂Cl₂/MeOH = 5/1); HPLC analysis: the diastereomers did not separate and observed as a single peak with R_t = 16.2 min [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. × 250 mm); mobile phase: CH₃CN:H₂O = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm]; Cyclooctyne **15c** is a mixture of diastereomers and conformational isomers. For the major isomer: ¹H NMR (400 MHz, CD₃OD) δ 1.37–1.68 (m, 12H), 2.15–2.18 (m, 2H), 2.66–2.88 (m, 3H), 3.16–3.62 (m, 18H), 3.90–3.92 (m, 2H), 4.24–4.27 (m, 1H), 4.43–4.47 (m, 1H), 4.60–4.62 (m, 2H), 5.39–5.43 (m, 1H), 7.26–7.37 (m, 7H), 7.52–7.53 (m, 1H), 7.84–7.95 (m, 4H), 8.43 (s, 1H); IR (KBr, cm⁻¹) 768, 1105, 1258, 1450, 1635, 1696, 2475, 2928; HRMS (ESI⁺) m/z 943.4130 ([M+Na]⁺, C₄₉H₆₀N₈NaO₈S⁺ requires 943.4147).

HPLC chart:

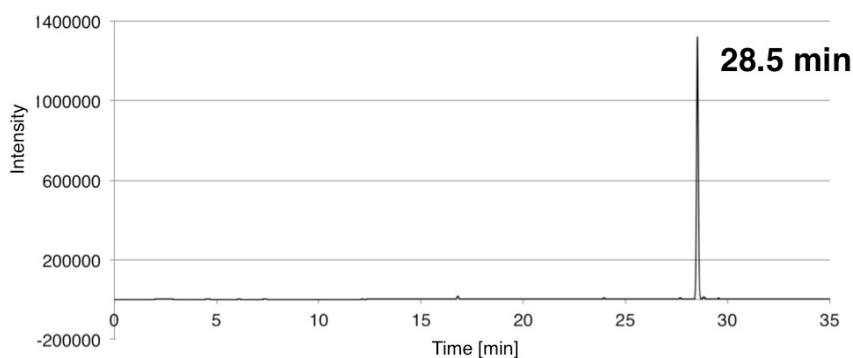


1-(4'-Azido-5-azidomethyl-3',5'-diisopropyl-1,1'-biphenyl-3-yl)-N-(2-(2-(2-(2-(4-(4-((2-(2-((6-chlorohexyl)oxy)ethoxy)ethyl)carbamoyl)phenyl)-1*H*-1,2,3-triazol-1-yl)ethoxy)ethoxy)ethoxy)ethyl)-5-methyl-1*H*-1,2,3-triazole-4-carboxamide (**S9**)

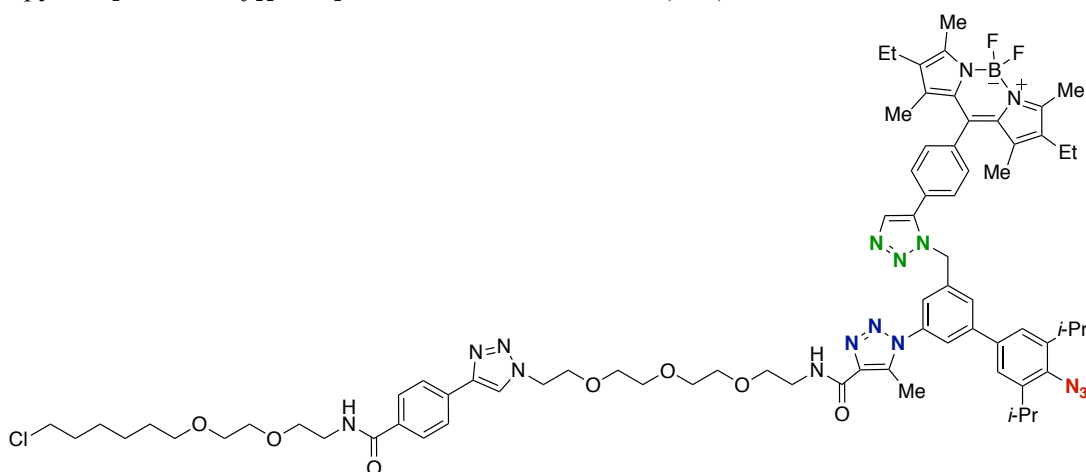


Pale orange oil; TLC R_f = 0.62 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 10/1); HPLC analysis: R_t = 28.5 min [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. $\times$ 250 mm); mobile phase: $\text{CH}_3\text{CN}:\text{H}_2\text{O}$ = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm]; ^1H NMR (400 MHz, CDCl_3) δ 1.25–1.45 (m, 16H), 1.53–1.62 (m, 2H), 1.69–1.77 (m, 2H), 2.64 (s, 3H), 3.44–3.50 (m, 6H), 3.59–3.69 (m, 20H), 3.95 (t, 2H, J = 4.8 Hz), 4.56 (s, 2H), 4.62 (t, 2H, J = 4.8 Hz), 6.82 (s, 1H), 7.32 (s, 2H), 7.37 (s, 1H), 7.57–7.64 (m, 3H), 7.84 (d, 2H, J = 8.4 Hz), 7.89 (d, 2H, J = 8.4 Hz), 8.10 (s, 1H); ^{13}C NMR (126 MHz, CDCl_3) δ 9.8 (1C), 23.5 (4C), 25.4 (1C), 26.6 (1C), 28.9 (2C), 29.4 (1C), 32.5 (1C), 38.7 (1C), 39.7 (1C), 45.0 (1C), 50.4 (1C), 54.0 (1C), 69.4 (1C), 69.7 (1C), 69.8 (1C), 70.0 (1C), 70.2 (1C), 70.4 (1C), 70.53 (1C), 70.55 (1C), 70.61 (1C), 71.3 (1C), 121.7 (1C), 123.0 (2C), 123.2 (1C), 123.7 (1C), 125.5 (2C), 127.6 (2C), 128.0 (1C), 133.77 (1C), 133.78 (1C), 135.7 (1C), 136.4 (1C), 136.8 (1C), 137.3 (1C), 138.1 (1C), 138.6 (1C), 143.7 (1C), 144.0 (2C), 146.6 (1C), 161.2 (1C), 167.0 (1C); IR (KBr, cm^{-1}) 862, 1111, 1287, 1460, 1518, 1659, 2102, 2932; HRMS (ESI^+) m/z 1033.4853 ($[\text{M}+\text{Na}]^+$, $\text{C}_{50}\text{H}_{67}^{35}\text{ClN}_{14}\text{NaO}_7^+$ requires 1033.4898).

HPLC chart:

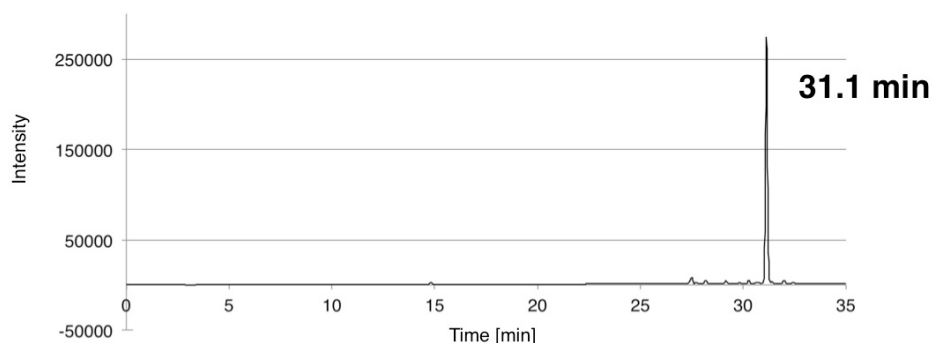


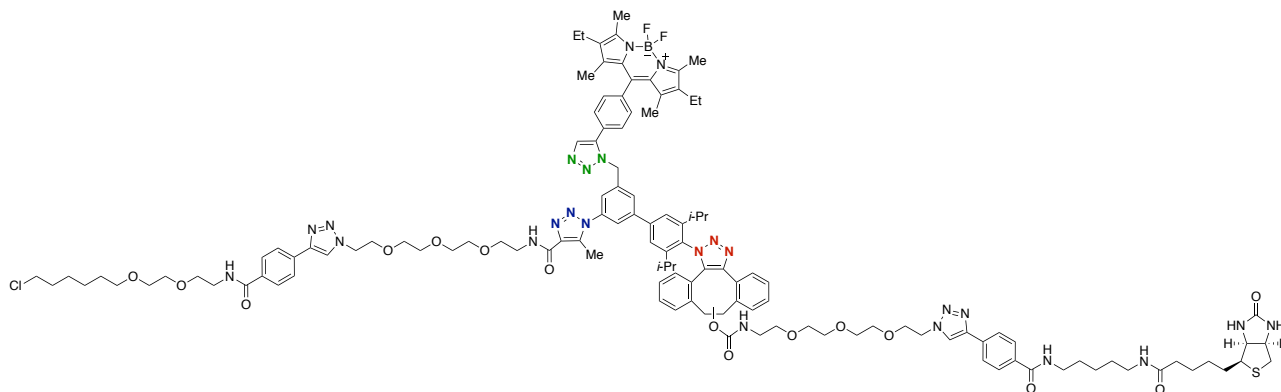
10-(4-(1-((4'-Azido-5-(4-((2-(2-(2-(2-(4-(4-((2-(2-((6-chlorohexyl)oxy)ethoxy)ethyl)carbamoyl)phenyl)-1*H*-1,2,3-triazol-1-yl)ethoxy)ethoxy)ethoxy)ethyl)carbamoyl)-5-methyl-1*H*-1,2,3-triazol-1-yl)-3',5'-diisopropyl-1,1'-biphenyl-3-yl)methyl)-1*H*-1,2,3-triazol-5-yl)phenyl)-2,8-diethyl-5,5-difluoro-1,3,7,9-tetramethyl-5*H*-dipyrrolo[1,2-*c*:2',1'-*f*][1,3,2]diazaborinin-4-ium-5-uide (**S10**)



Pink solid; Mp 62 °C (dec.); TLC R_f = 0.41 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 10/1); HPLC analysis: R_t = 31.1 min [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. $\times$ 250 mm); mobile phase: $\text{CH}_3\text{CN}:\text{H}_2\text{O}$ = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm]; ^1H NMR (400 MHz, CDCl_3) δ 0.92 (t, 6H, J = 7.2 Hz), 1.17–1.45 (m, 22H), 1.54–1.63 (m, 2H), 1.68–1.77 (m, 2H), 2.23 (q, 4H, J = 7.6 Hz), 2.52 (s, 6H), 2.56 (s, 3H), 3.37–3.50 (m, 6H), 3.58–3.68 (m, 20H), 3.92 (t, 2H, J = 4.8 Hz), 4.59 (t, 2H, J = 4.8 Hz), 5.80 (s, 2H), 6.82 (br s, 1H), 7.12 (s, 1H), 7.24 (s, 2H), 7.40–7.43 (m, 3H), 7.50–7.54 (m, 4H), 7.84–7.90 (m, 5H), 8.09 (s, 1H); ^{13}C NMR (126 MHz, CDCl_3) δ 9.7 (1C), 11.7 (2C), 12.5 (1C), 14.5 (2C), 17.0 (2C), 23.4 (4C), 25.4 (1C), 26.6 (2C), 28.9 (2C), 29.4 (1C), 29.7 (1C), 32.4 (1C), 38.7 (1C), 39.7 (1C), 45.0 (1C), 50.4 (1C), 51.3 (1C), 69.4 (1C), 69.7 (1C), 69.8 (1C), 70.0 (1C), 70.2 (1C), 70.4 (1C), 70.5 (1C), 70.6 (1C), 71.3 (1C), 121.7 (1C), 122.2 (1C), 122.8 (2C), 123.9 (1C), 125.5 (2C), 126.8 (2C), 127.1 (2C), 127.6 (2C), 129.2 (2C), 129.5 (2C), 130.4 (1C), 133.1 (1C), 133.75 (2C), 133.79 (1C+1C, two signals overlapped), 135.9 (1C), 136.6 (1C), 136.7 (1C), 136.9 (1C), 137.5 (1C), 137.6 (2C+1C, two signals overlapped), 137.7 (1C), 138.0 (1C), 138.6 (1C), 143.9 (1C), 144.1 (2C), 146.6 (1C), 154.4 (1C), 161.0 (1C), 167.0 (1C); FL (MeOH) λ_{max} = 538 nm; IR (KBr, cm^{-1}) 980, 1145, 1194, 1321, 1476, 1541, 1655, 2122, 2926; HRMS (ESI $^+$) m/z 1437.7143 ($[\text{M}+\text{Na}]^+$, $\text{C}_{75}\text{H}_{94}^{11}\text{B}^{35}\text{ClF}_2\text{N}_{16}\text{NaO}_7^+$ requires 1437.7133).

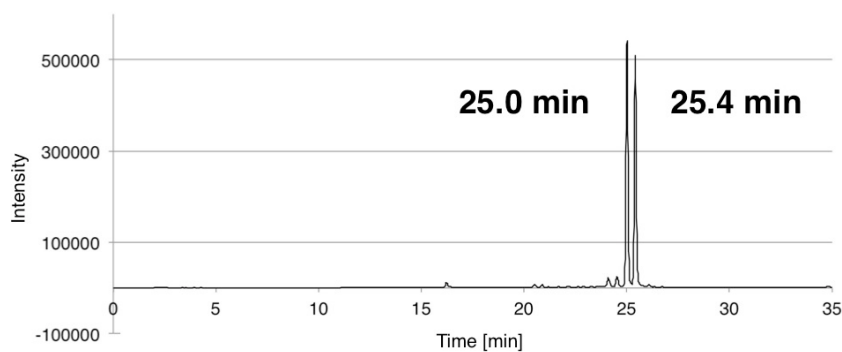
HPLC chart:

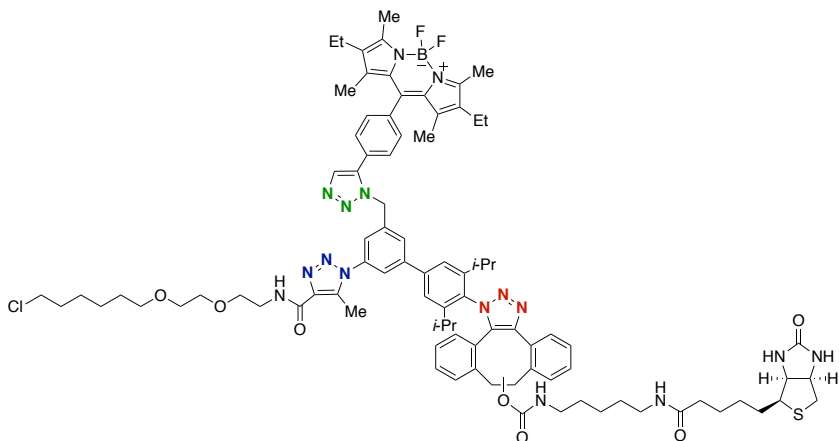


Trifunctional probe **16d**

Pink solid; Mp 104 °C (dec.); TLC R_f = 0.38 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 10/1); HPLC analysis: the diastereomers did not separate and two peaks for regioisomers were observed at R_t = 25.0 min (51%) and 25.4 min (49%) [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. $\times$ 250 mm); mobile phase: $\text{CH}_3\text{CN}:\text{H}_2\text{O}$ = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm]; FL (MeOH) λ_{max} = 538 nm; IR (KBr, cm^{-1}) 979, 1193, 1471, 1544, 1652, 1703, 2928, 3303; HRMS (ESI^+) m/z 2359.1422 ($[\text{M}+\text{Na}]^+$, $\text{C}_{124}\text{H}_{154}^{11}\text{B}^{35}\text{ClF}_2\text{N}_{24}\text{NaO}_{15}\text{S}^+$ requires 2359.1494).

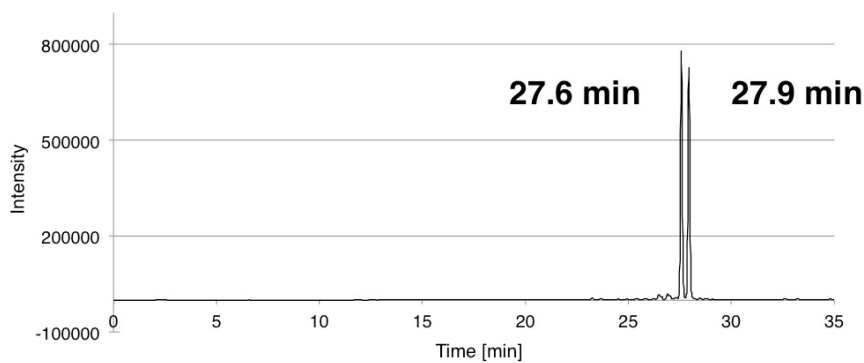
HPLC chart:



Trifunctional probe **16a**

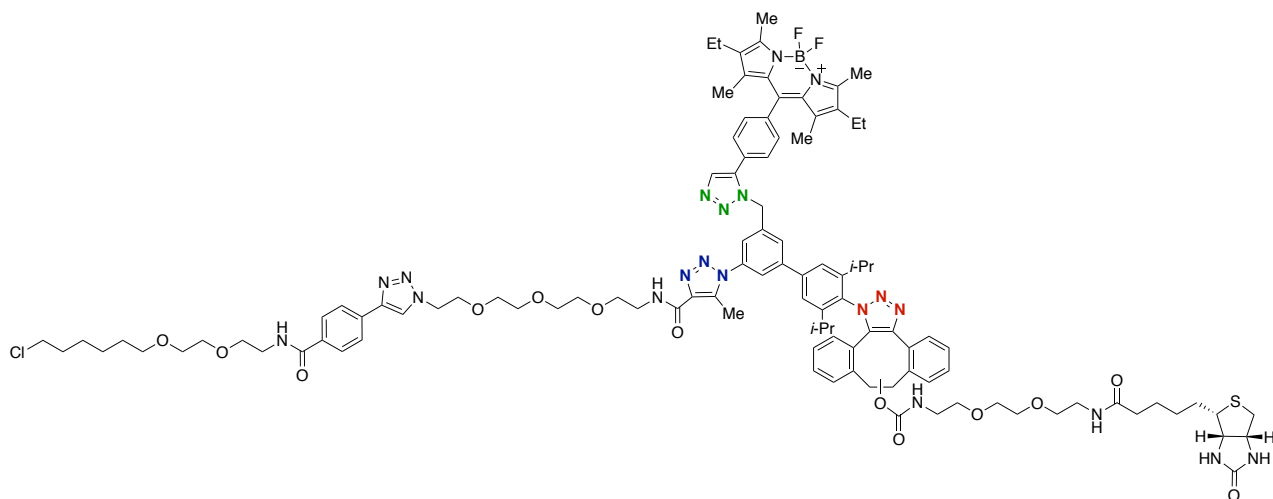
Pink solid; Mp 110 °C (dec.); TLC R_f = 0.10 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 20/1); HPLC analysis: the diastereomers did not separate and two peaks for regioisomers were observed at R_t = 27.6 min (51%) and 27.9 min (49%) [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. $\times$ 250 mm); mobile phase: $\text{CH}_3\text{CN}:\text{H}_2\text{O}$ = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm]; FL (MeOH) λ_{max} = 538 nm; IR (KBr, cm^{-1}) 737, 980, 1192, 1265, 1477, 1543, 1659, 1704, 2928, 3292; HRMS (ESI⁺) m/z 1665.8106 ($[\text{M}+\text{Na}]^+$, $\text{C}_{90}\text{H}_{110}^{11}\text{B}^{35}\text{ClF}_2\text{N}_{16}\text{NaO}_7\text{S}^+$ requires 1665.8106).

HPLC chart:



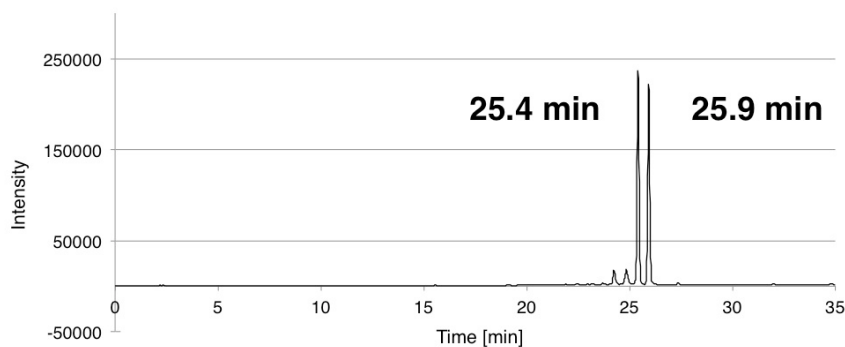
CCCCCCCCOCCOCCNC(=O)c1ccc(cc1)/N=N/c2cc(C#N)cc(C#N)c2NCCOCCOCCOCCOCCNC(=O)c3nc(C)c4nc(C)c5cc(C#N)cc5c4n3c6ccc(cc6)-c7cc(C#N)cc8c7c9c10c11c12c13c14c15c16c17c18c19c20c21c22c23c24c25c26c27c28c29c30c31c32c33c34c35c36c37c38c39c40c41c42c43c44c45c46c47c48c49c50c51c52c53c54c55c56c57c58c59c60c61c62c63c64c65c66c67c68c69c70c71c72c73c74c75c76c77c78c79c80c81c82c83c84c85c86c87c88c89c90c91c92c93c94c95c96c97c98c99c100c101c102c103c104c105c106c107c108c109c110c111c112c113c114c115c116c117c118c119c120c121c122c123c124c125c126c127c128c129c130c131c132c133c134c135c136c137c138c139c140c141c142c143c144c145c146c147c148c149c150c151c152c153c154c155c156c157c158c159c160c161c162c163c164c165c166c167c168c169c170c171c172c173c174c175c176c177c178c179c180c181c182c183c184c185c186c187c188c189c190c191c192c193c194c195c196c197c198c199c200c201c202c203c204c205c206c207c208c209c210c211c212c213c214c215c216c217c218c219c220c221c222c223c224c225c226c227c228c229c230c231c232c233c234c235c236c237c238c239c240c241c242c243c244c245c246c247c248c249c250c251c252c253c254c255c256c257c258c259c260c261c262c263c264c265c266c267c268c269c270c271c272c273c274c275c276c277c278c279c280c281c282c283c284c285c286c287c288c289c290c291c292c293c294c295c296c297c298c299c300c301c302c303c304c305c306c307c308c309c310c311c312c313c314c315c316c317c318c319c320c321c322c323c324c325c326c327c328c329c330c331c332c333c334c335c336c337c338c339c340c341c342c343c344c345c346c347c348c349c350c351c352c353c354c355c356c357c358c359c360c361c362c363c364c365c366c367c368c369c370c371c372c373c374c375c376c377c378c379c380c381c382c383c384c385c386c387c388c389c390c391c392c393c394c395c396c397c398c399c400c401c402c403c404c405c406c407c408c409c410c411c412c413c414c415c416c417c418c419c420c421c422c423c424c425c426c427c428c429c430c431c432c433c434c435c436c437c438c439c440c441c442c443c444c445c446c447c448c449c450c451c452c453c454c455c456c457c458c459c460c461c462c463c464c465c466c467c468c469c470c471c472c473c474c475c476c477c478c479c480c481c482c483c484c485c486c487c488c489c490c491c492c493c494c495c496c497c498c499c500c501c502c503c504c505c506c507c508c509c510c511c512c513c514c515c516c517c518c519c520c521c522c523c524c525c526c527c528c529c530c531c532c533c534c535c536c537c538c539c540c541c542c543c544c545c546c547c548c549c550c551c552c553c554c555c556c557c558c559c560c561c562c563c564c565c566c567c568c569c570c571c572c573c574c575c576c577c578c579c580c581c582c583c584c585c586c587c588c589c590c591c592c593c594c595c596c597c598c599c600c601c602c603c604c605c606c607c608c609c610c611c612c613c614c615c616c617c618c619c620c621c622c623c624c625c626c627c628c629c630c631c632c633c634c635c636c637c638c639c640c641c642c643c644c645c646c647c648c649c650c651c652c653c654c655c656c657c658c659c660c661c662c663c664c665c666c667c668c669c670c671c672c673c674c675c676c677c678c679c680c681c682c683c684c685c686c687c688c689c690c691c692c693c694c695c696c697c698c699c700c701c702c703c704c705c706c707c708c709c710c711c712c713c714c715c716c717c718c719c720c721c722c723c724c725c726c727c728c729c730c731c732c733c734c735c736c737c738c739c740c741c742c743c744c745c746c747c748c749c750c751c752c753c754c755c756c757c758c759c760c761c762c763c764c765c766c767c768c769c770c771c772c773c774c775c776c777c778c779c780c781c782c783c784c785c786c787c788c789c790c791c792c793c794c795c796c797c798c799c800c801c802c803c804c805c806c807c808c809c810c811c812c813c814c815c816c817c818c819c820c821c822c823c824c825c826c827c828c829c830c831c832c833c834c835c836c837c838c839c840c841c842c843c844c845c846c847c848c849c850c851c852c853c854c855c856c857c858c859c860c861c862c863c864c865c866c867c868c869c870c871c872c873c874c875c876c877c878c879c880c881c882c883c884c885c886c887c888c889c890c891c892c893c894c895c896c897c898c899c900c901c902c903c904c905c906c907c908c909c910c911c912c913c914c915c916c917c918c919c920c921c922c923c924c925c926c927c928c929c930c931c932c933c934c935c936c937c938c939c940c941c942c943c944c945c946c947c948c949c950c951c952c953c954c955c956c957c958c959c960c961c962c963c964c965c966c967c968c969c970c971c972c973c974c975c976c977c978c979c980c981c982c983c984c985c986c987c988c989c990c991c992c993c994c995c996c997c998c999

The chromatogram displays Intensity on the y-axis (ranging from -20,000 to 160,000) and Time [min] on the x-axis (ranging from 0 to 35). Two prominent peaks are observed: one at 25.9 min and another at 26.4 min. The peak at 25.9 min is slightly taller than the peak at 26.4 min.

Trifunctional probe **16c**

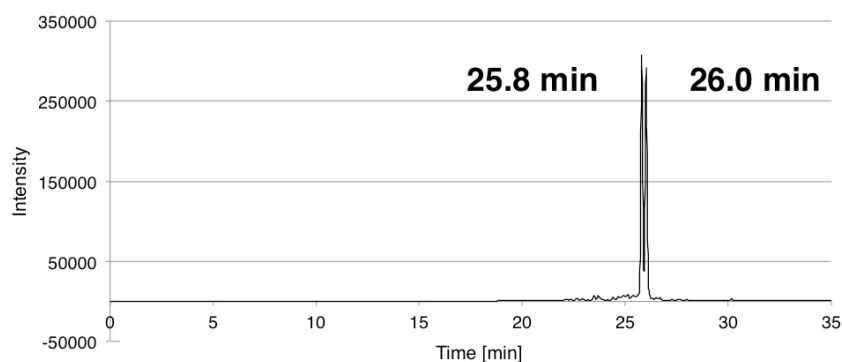
Pink solid; Mp 108 °C (dec.); TLC R_f = 0.24 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 10/1); HPLC analysis: the diastereomers did not separate and two peaks for regioisomers were observed at R_t = 25.4 min (52%) and 25.9 min (48%) [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. $\times$ 250 mm); mobile phase: $\text{CH}_3\text{CN}:\text{H}_2\text{O}$ = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm]; FL (MeOH) λ_{max} = 538 nm; IR (KBr, cm^{-1}) 742, 1076, 1084, 1117, 1195, 1460, 1543, 1649, 1693, 1693, 2929; HRMS (ESI^+) m/z 2058.9778 ($[\text{M}+\text{Na}]^+$, $\text{C}_{108}\text{H}_{134}^{11}\text{B}^{35}\text{ClF}_2\text{N}_{20}\text{NaO}_{13}\text{S}^+$ requires 2058.9835).

HPLC chart:



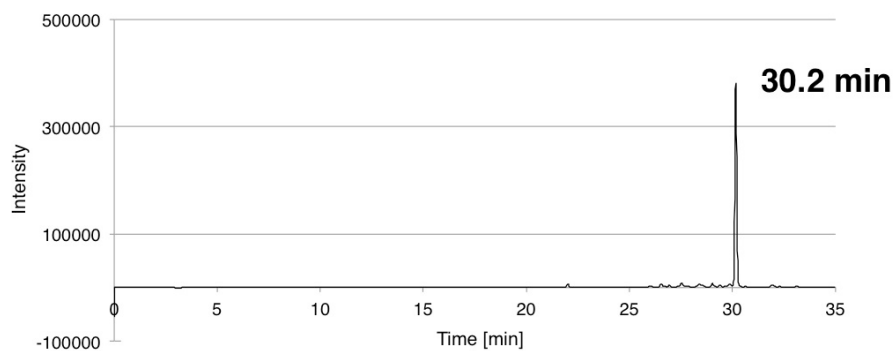
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HPLC chart:

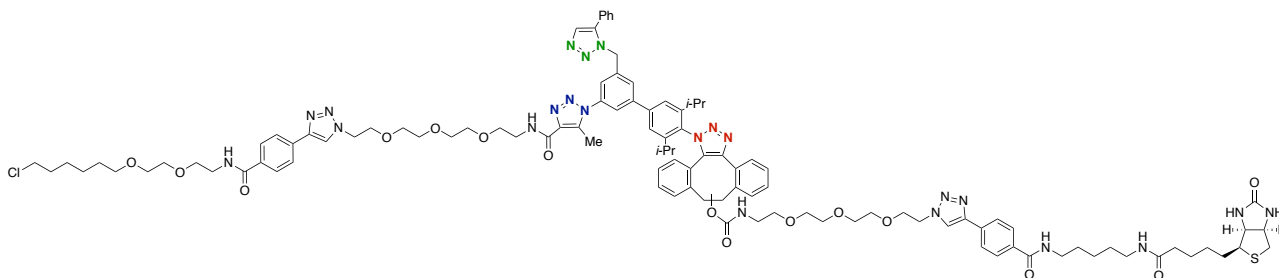


Chemical structure of compound 10, showing a complex molecule with a long alkyl chain, a linker with multiple ether groups, and a complex aromatic system including a triazole ring, a pyrazole ring, and a fluorinated boron atom.

HPLC chart:

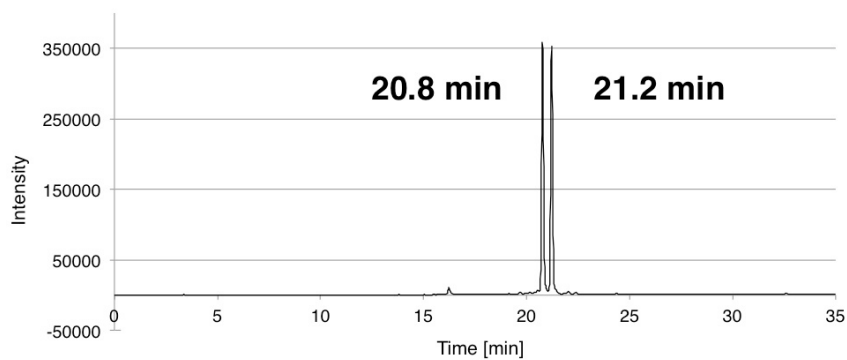


Bisfunctional triazole **S13**



Colorless solid; TLC R_f = 0.58 ($\text{CH}_2\text{Cl}_2/\text{MeOH}$ = 6/1); HPLC analysis: the diastereomers did not separate and two peaks for regioisomers were observed at R_t = 20.8 min (50%) and 21.2 min (50%) [column: Shiseido CAPCELL PAK MG II (4.6 mm i.d. $\times$ 250 mm); mobile phase: $\text{CH}_3\text{CN}:\text{H}_2\text{O}$ = 40:60 (0–5 min), linear gradient from 40:60 to 99:1 (5–25 min), 99:1 (25–35 min); flow rate: 1.00 mL/min; detection: UV at 254 nm]; HRMS (ESI⁺) m/z 2055.9575 ($[\text{M}+\text{Na}]^+$, $\text{C}_{107}\text{H}_{133}^{35}\text{ClN}_{22}\text{NaO}_{15}\text{S}^+$ requires 2055.9622).

HPLC chart:

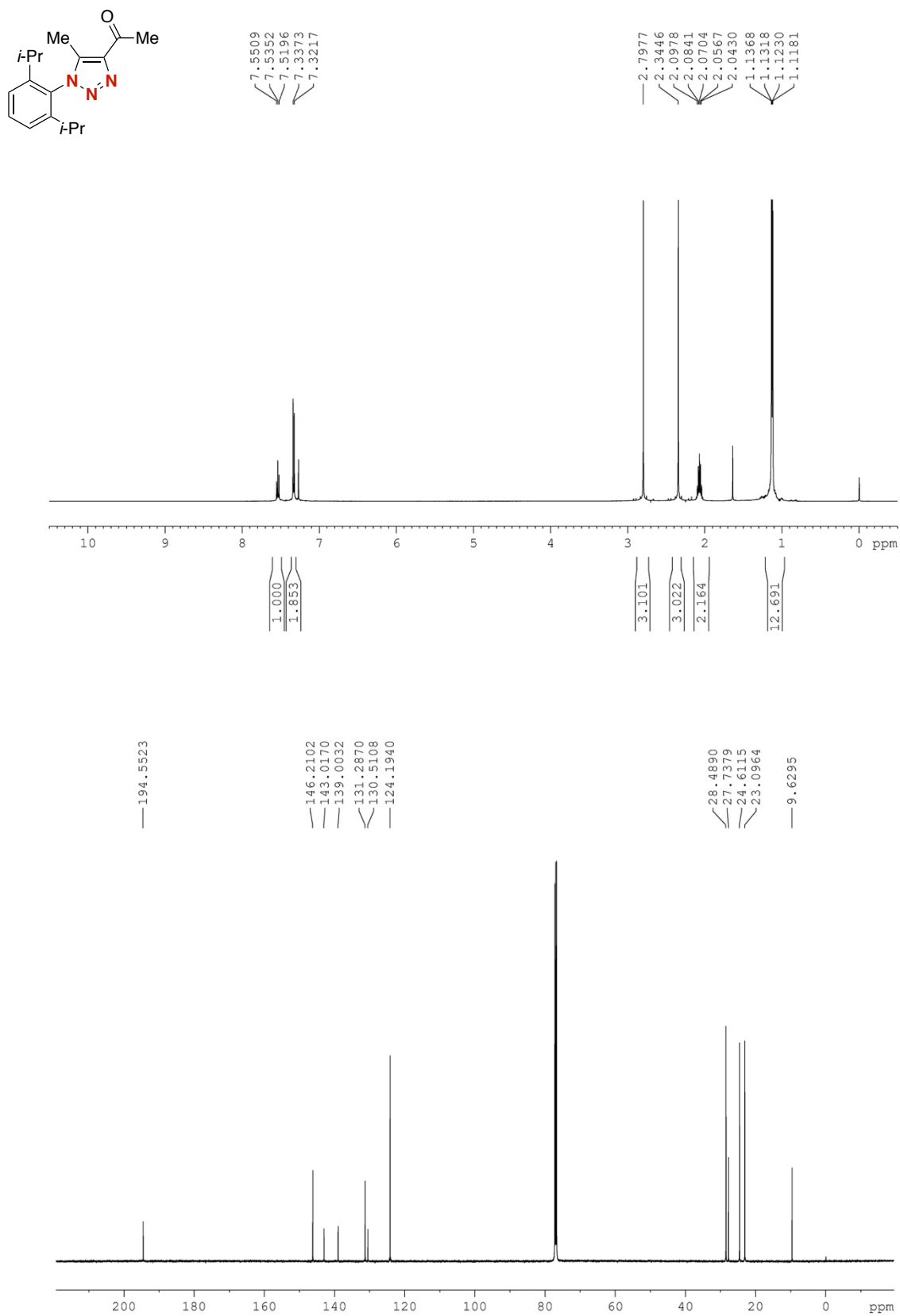


References for Supporting Information

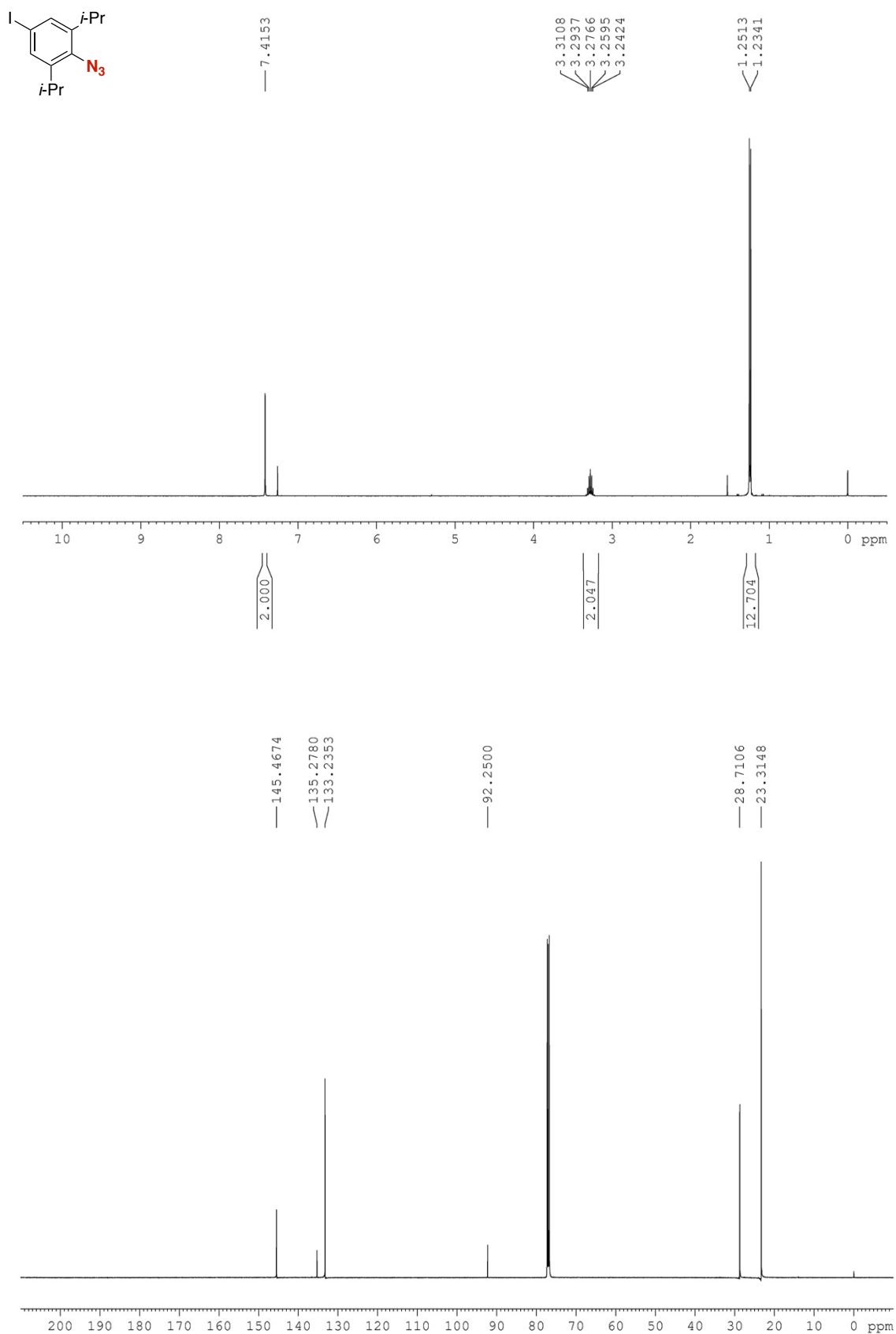
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^1H and ^{13}C NMR Spectra of Compounds

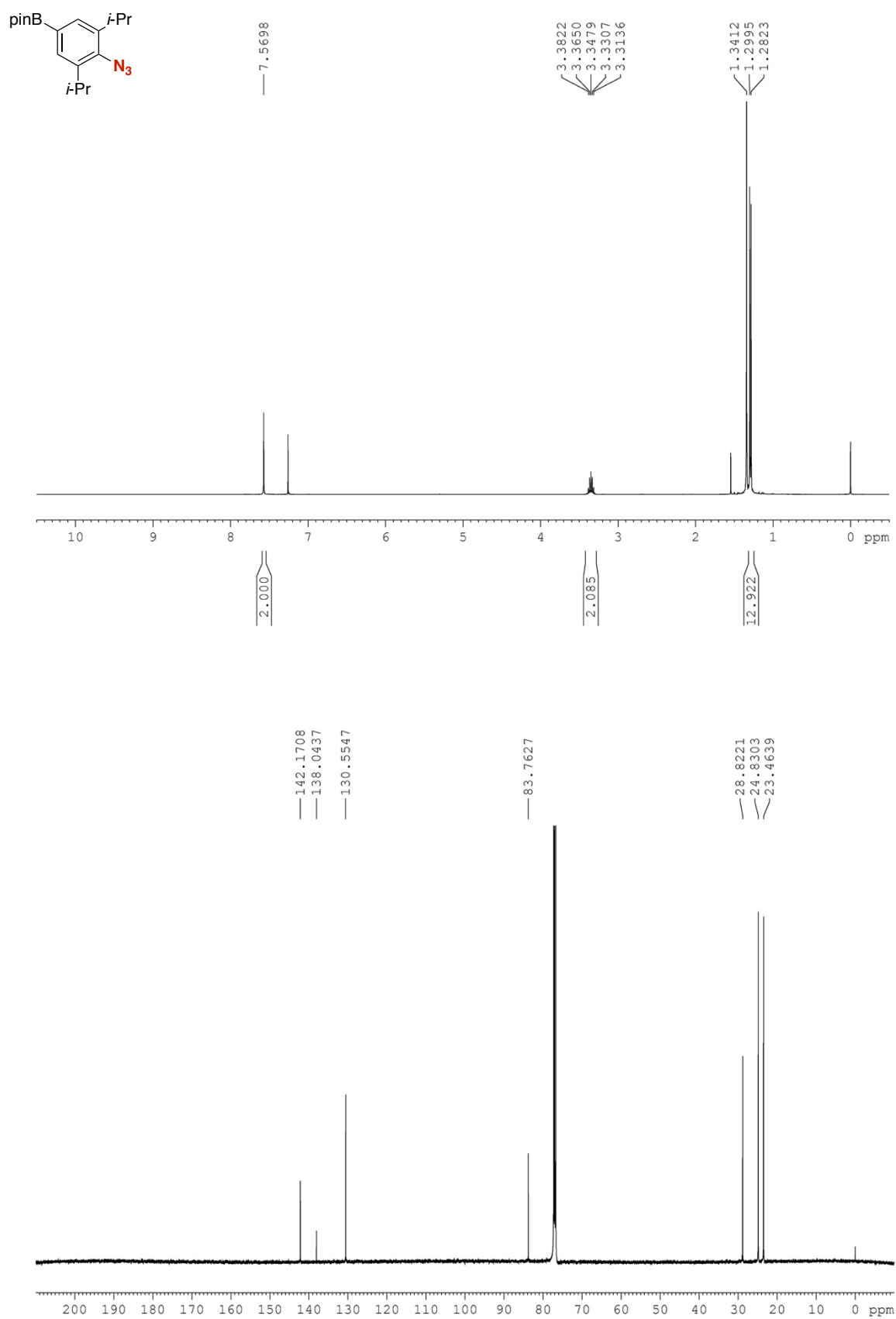
^1H NMR (500 MHz) and ^{13}C NMR (126 MHz) spectra of **8a** (CDCl_3)



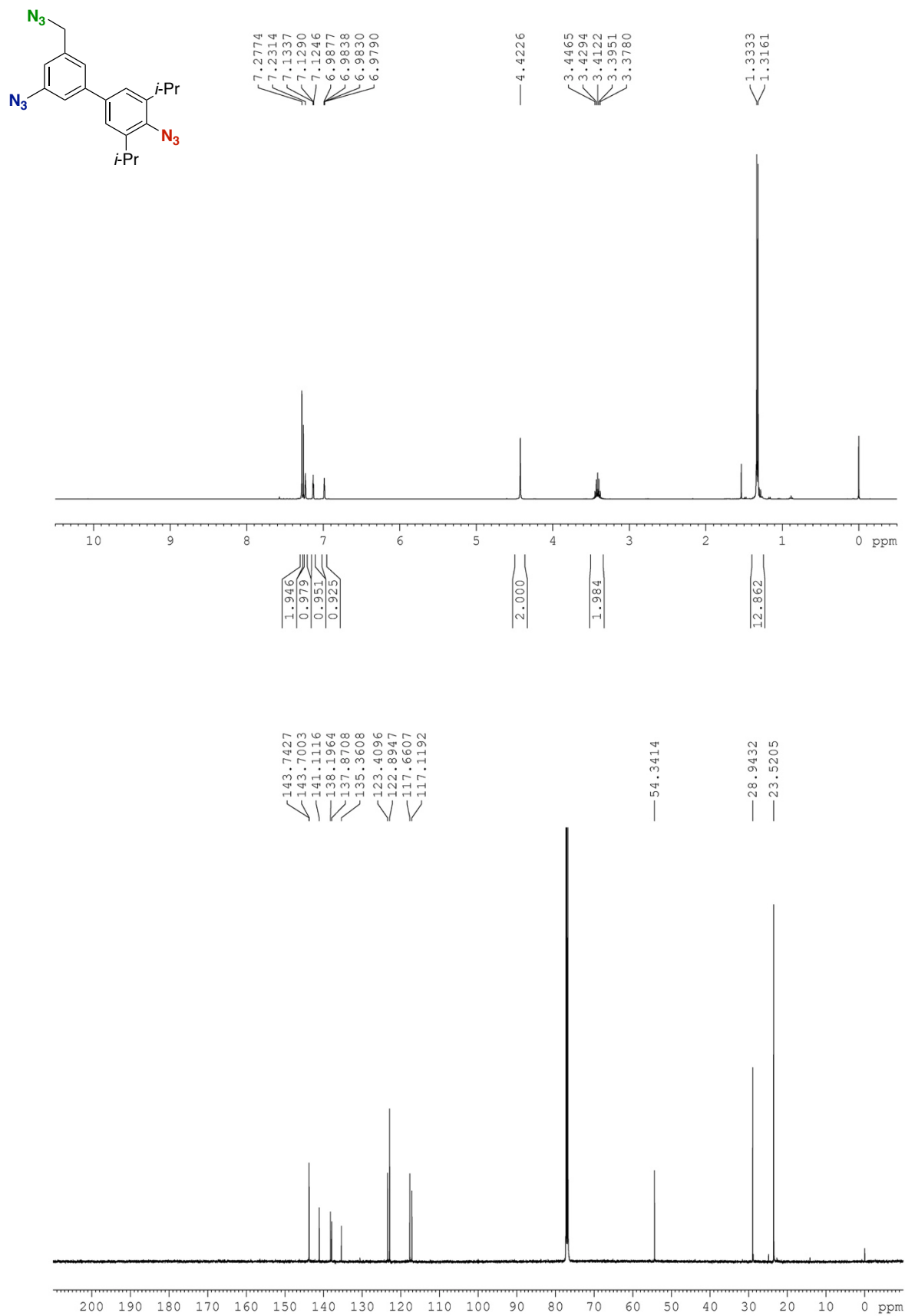
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **S1** (CDCl_3)



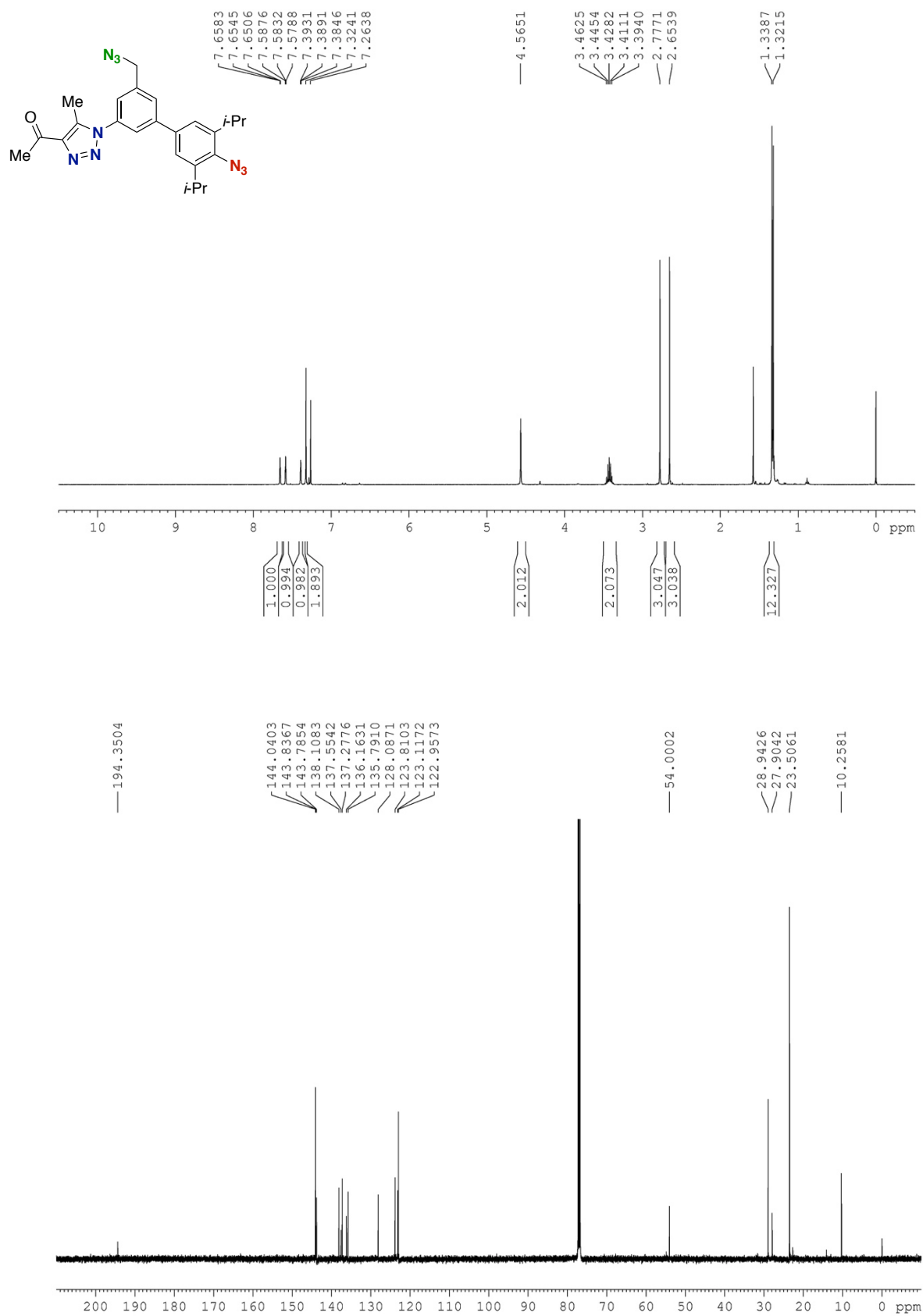
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **10** (CDCl_3)



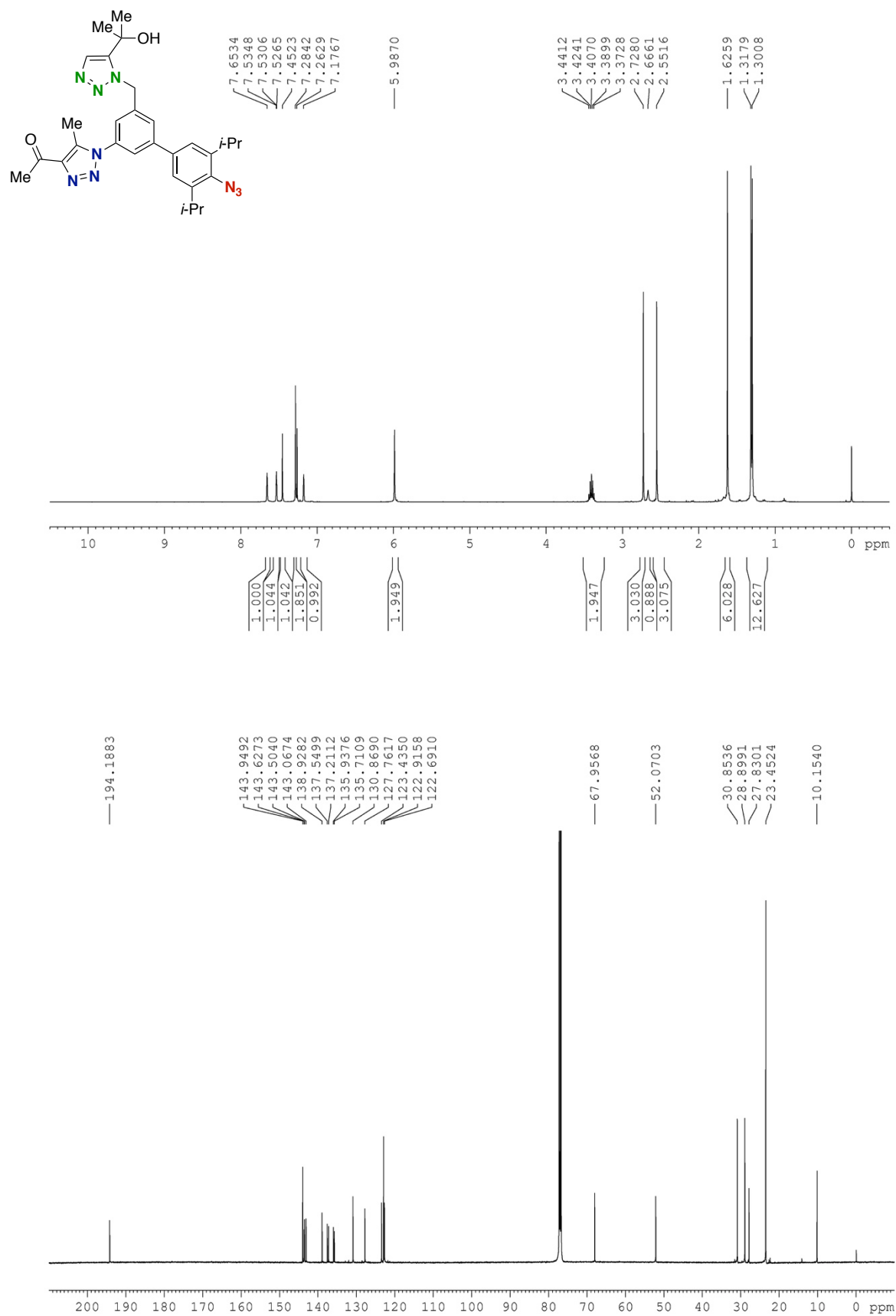
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **11** (CDCl_3)



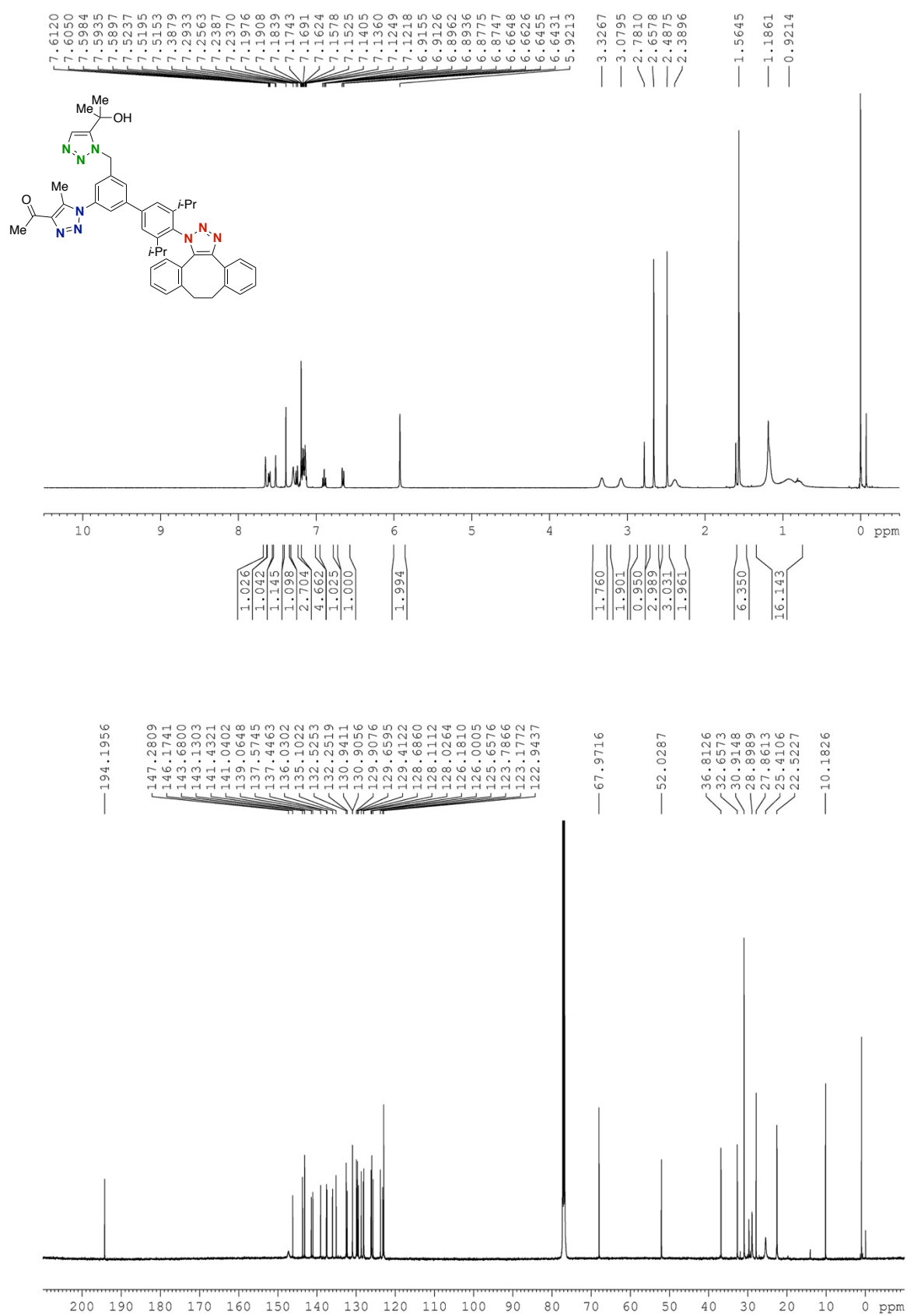
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **S2** (CDCl_3)



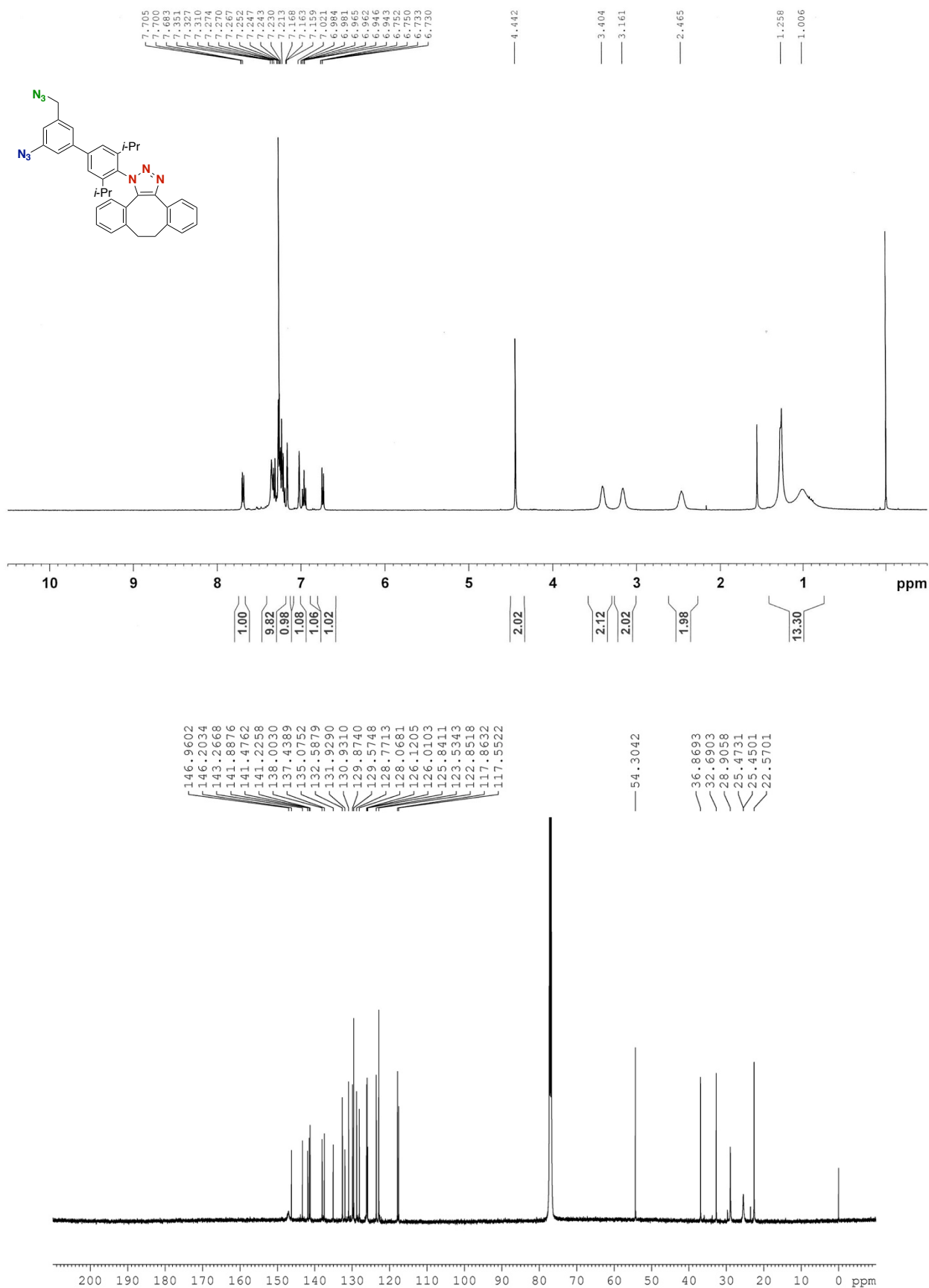
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **S3** (CDCl_3)



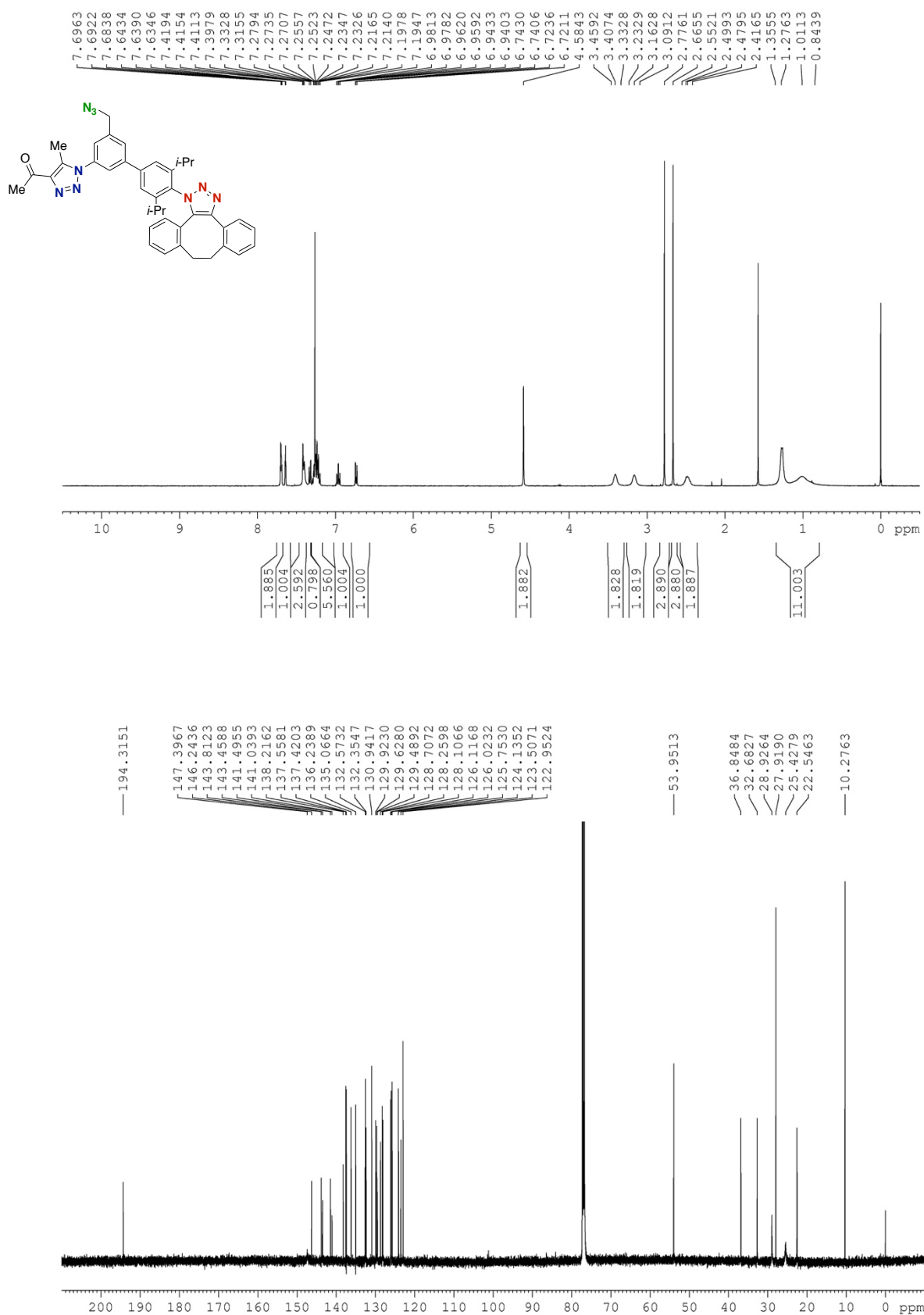
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **12a** (CDCl_3)



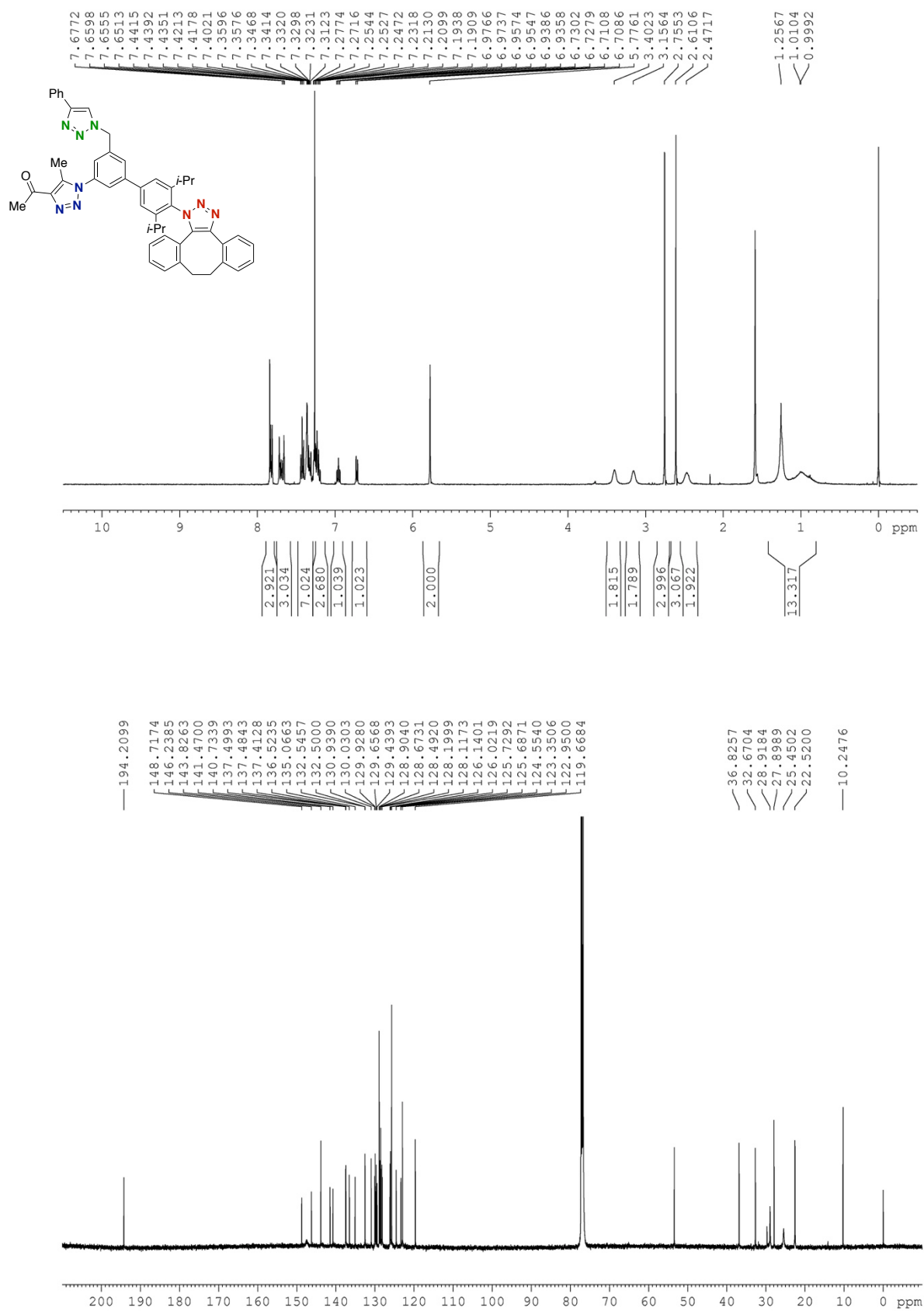
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **S4** (CDCl_3)



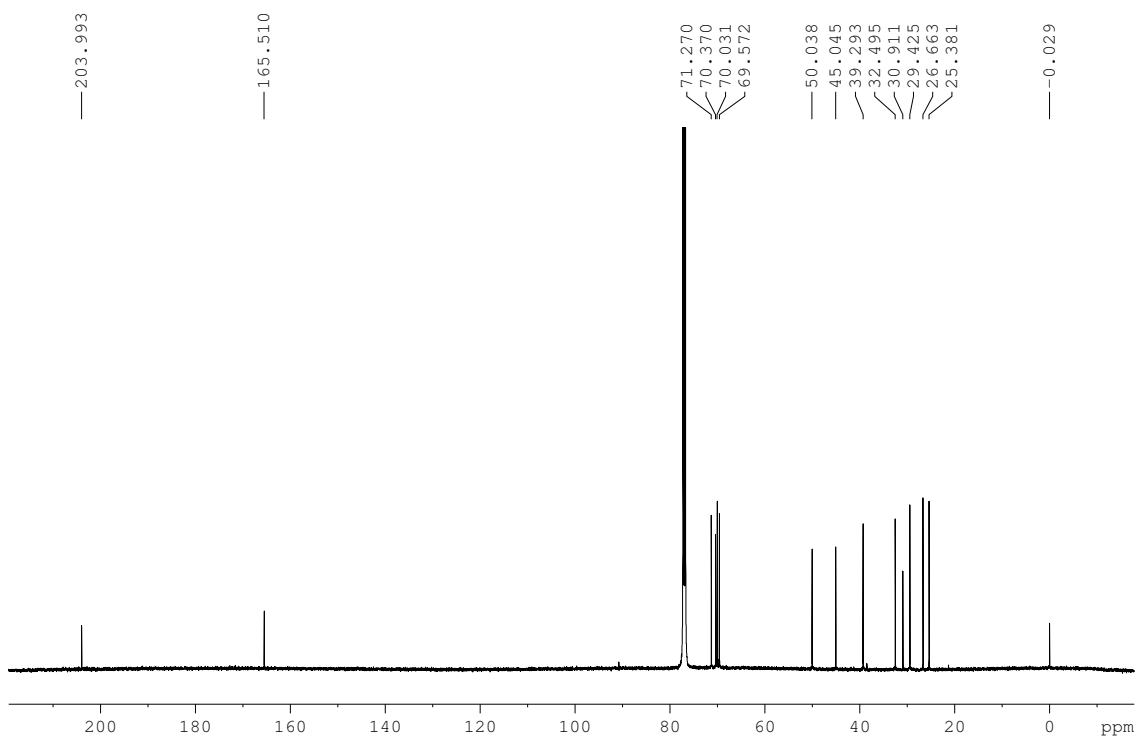
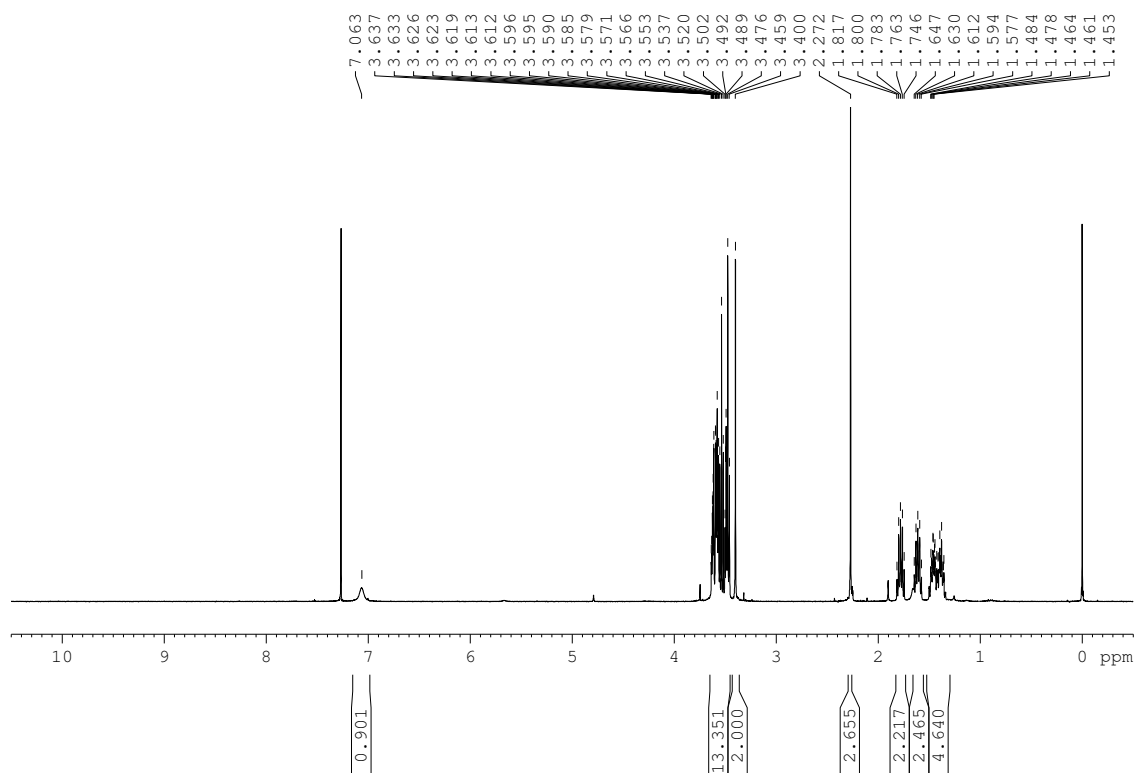
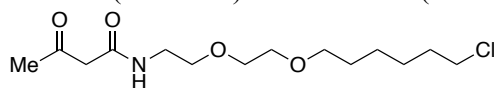
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **S5** (CDCl_3)



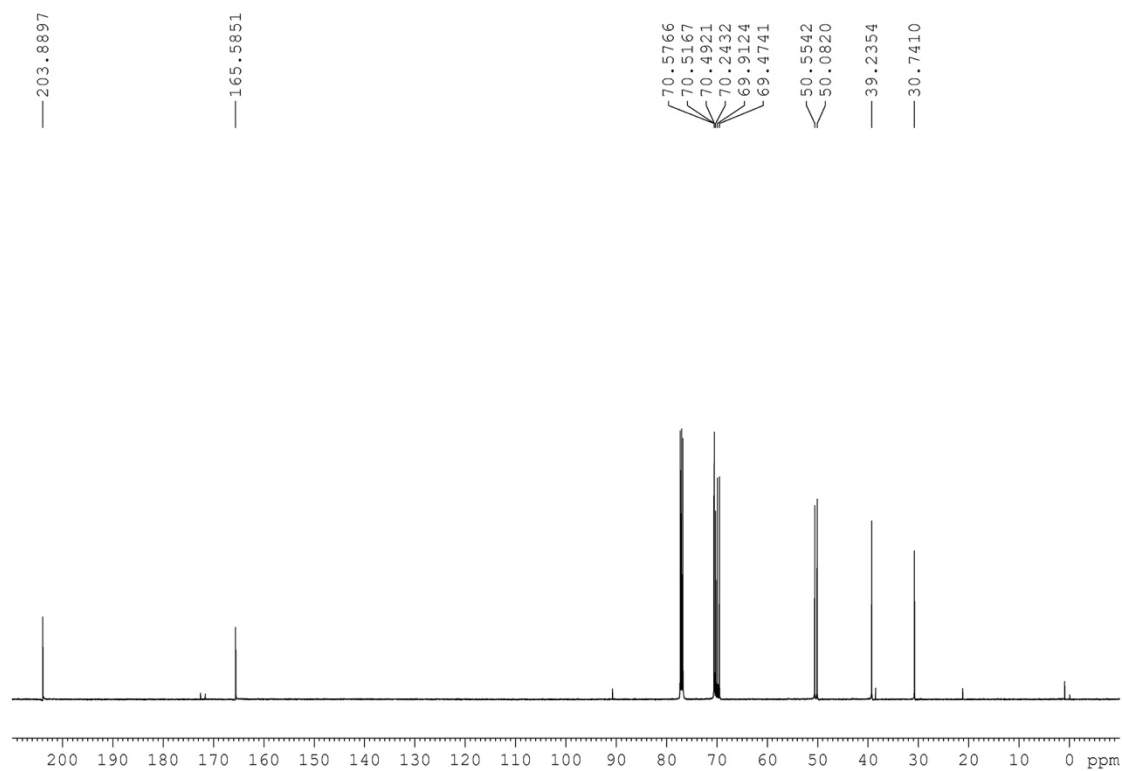
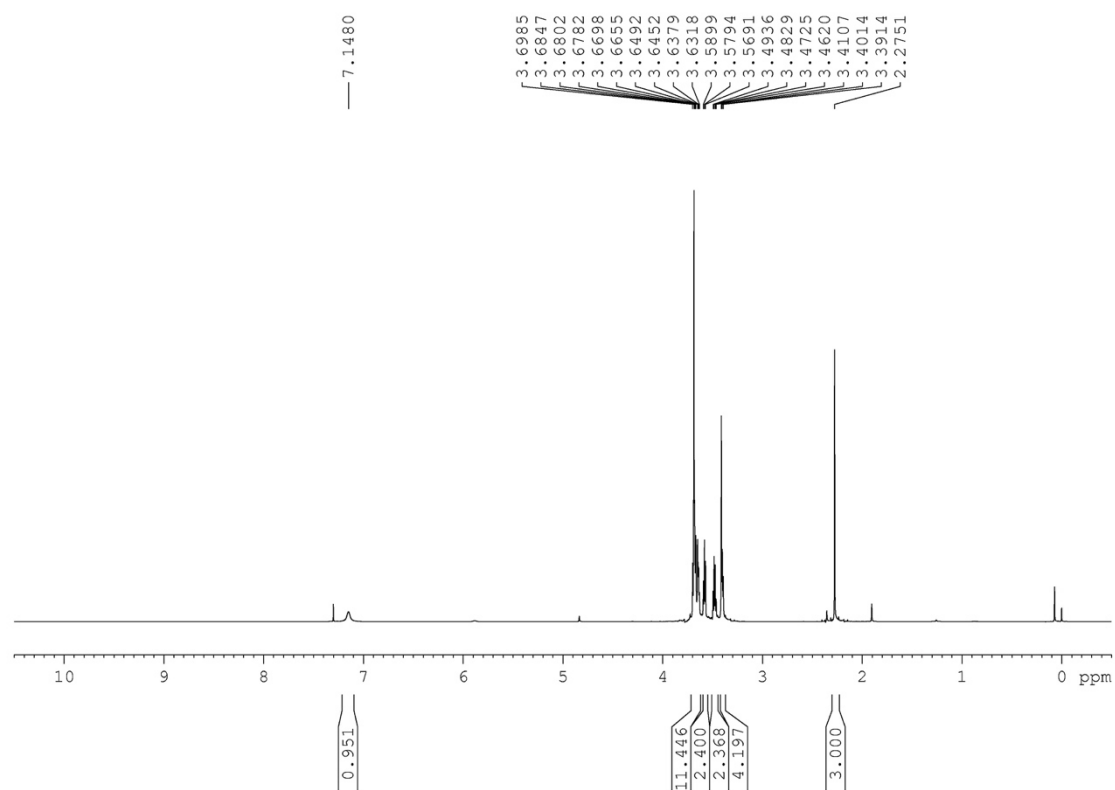
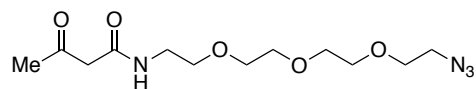
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **12b** (CDCl_3)



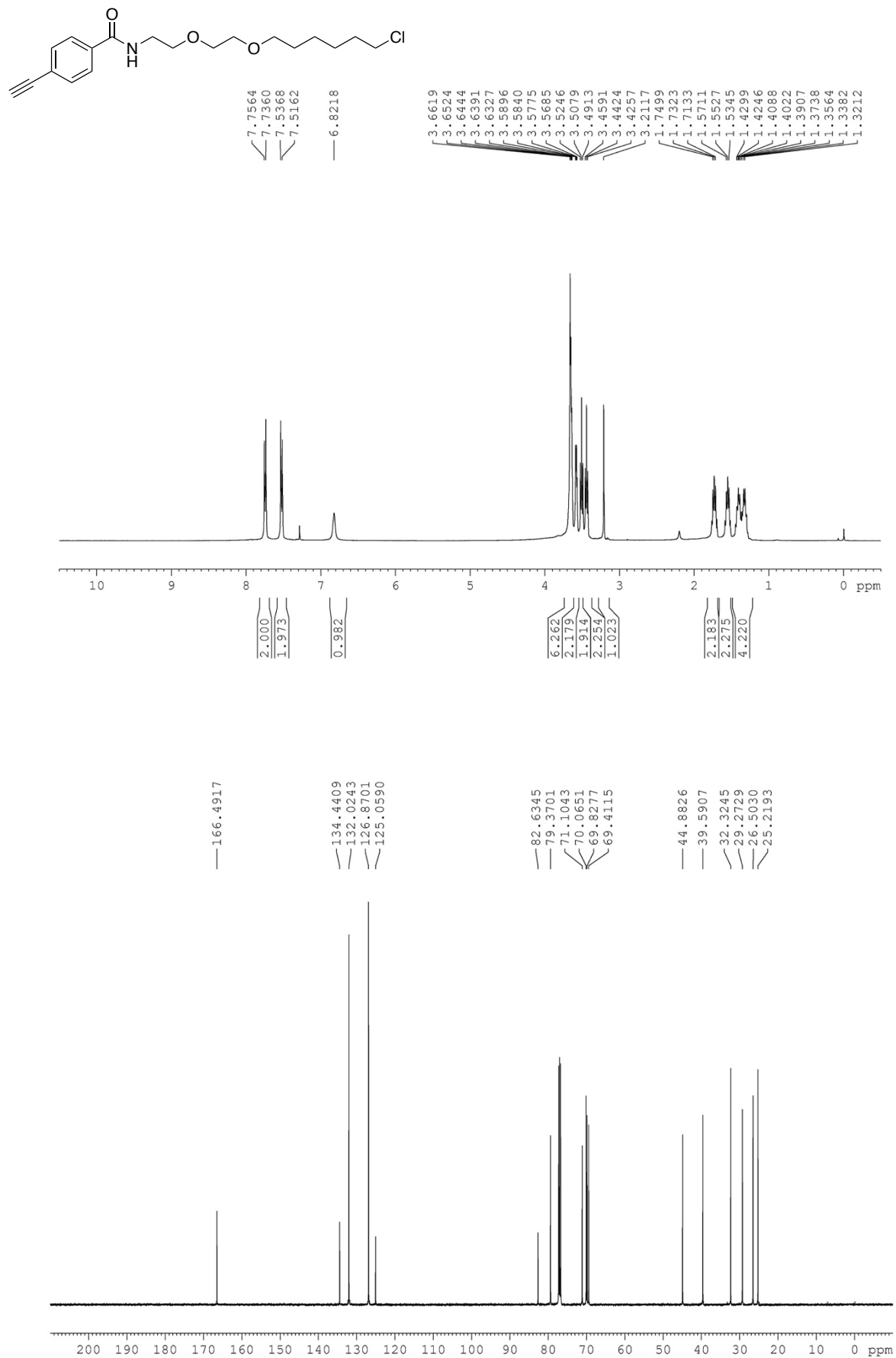
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **13a** (CDCl_3)



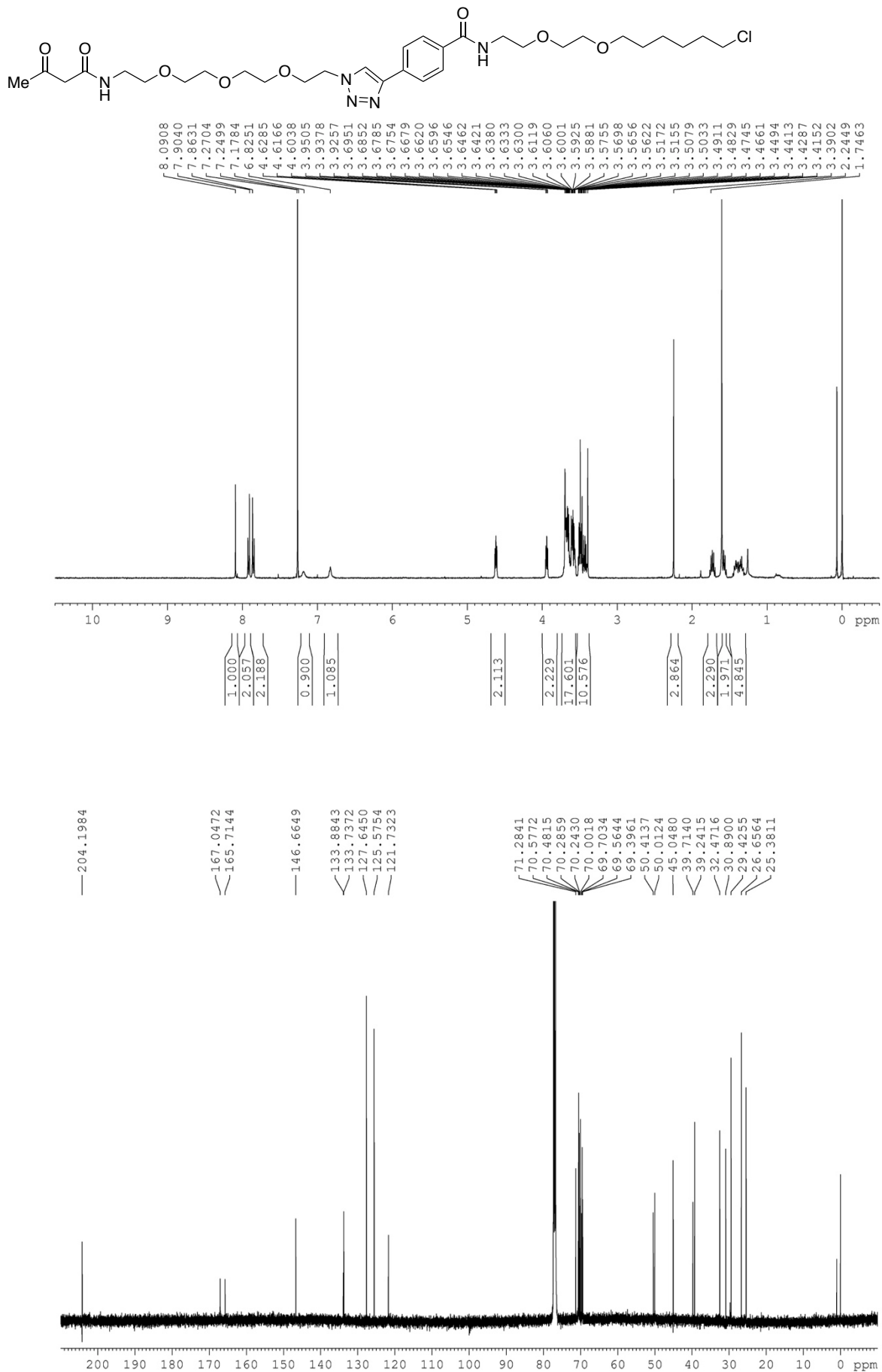
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **S6** (CDCl_3)



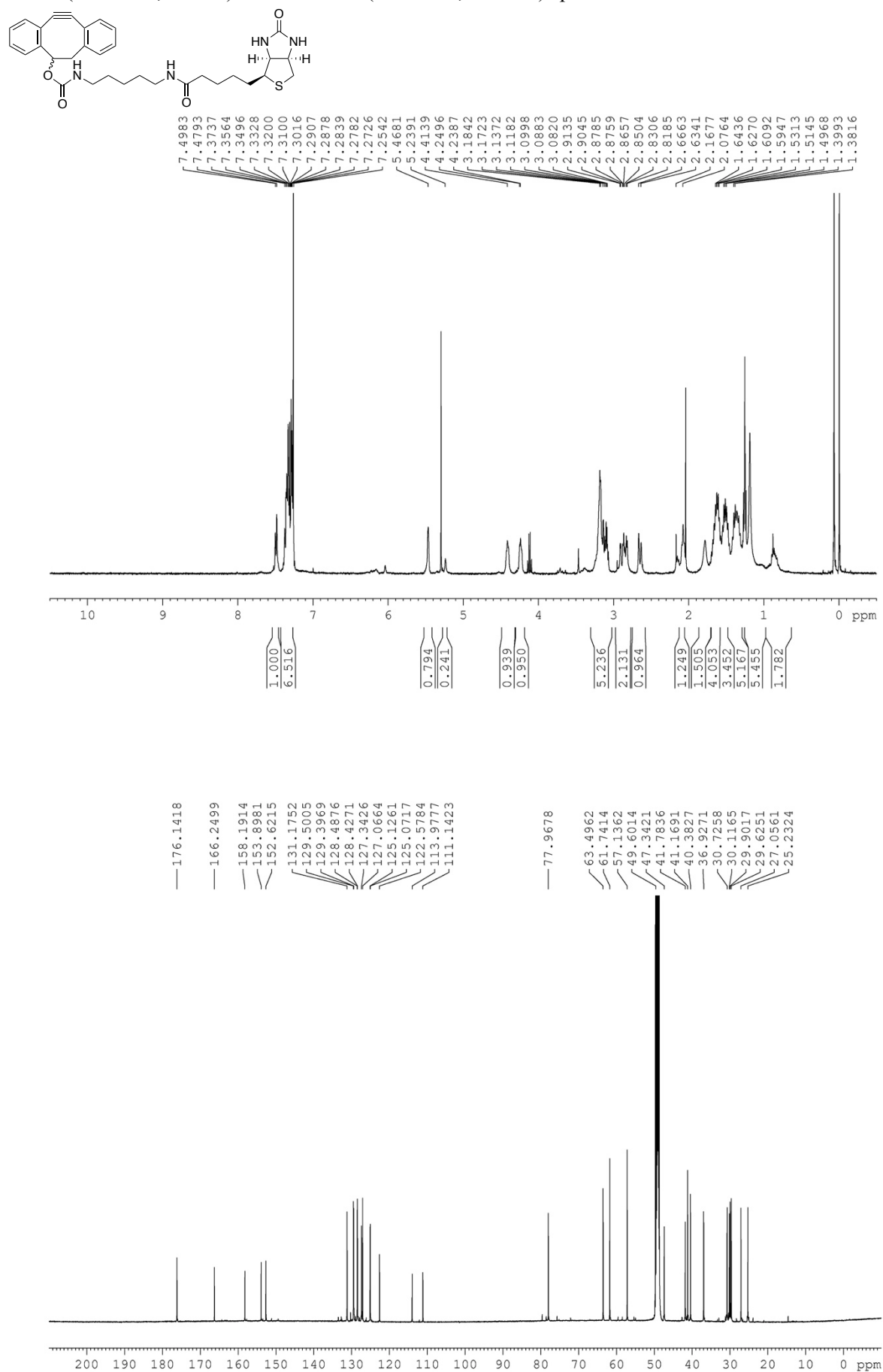
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **S7** (CDCl_3)



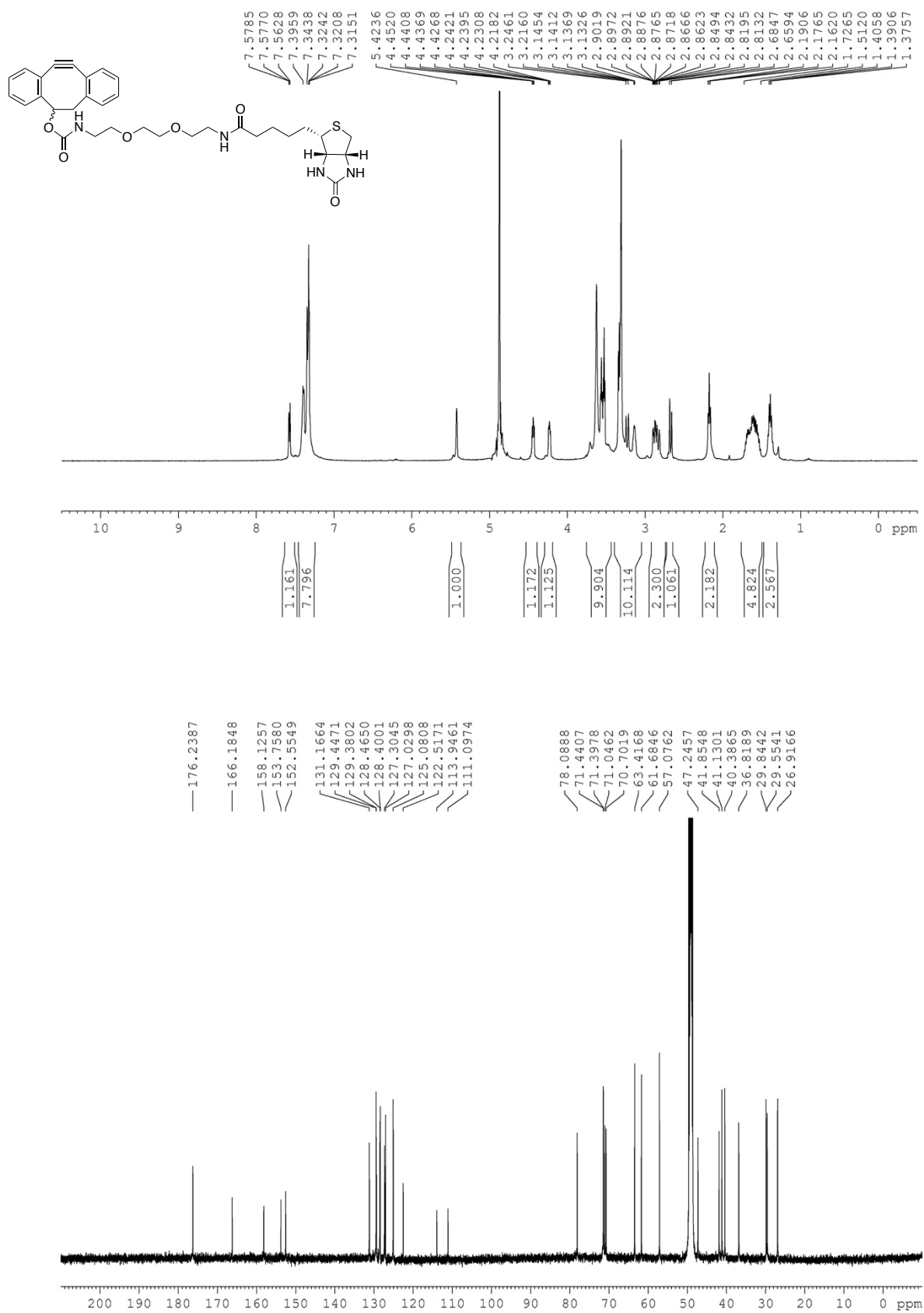
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **13b** (CDCl_3)

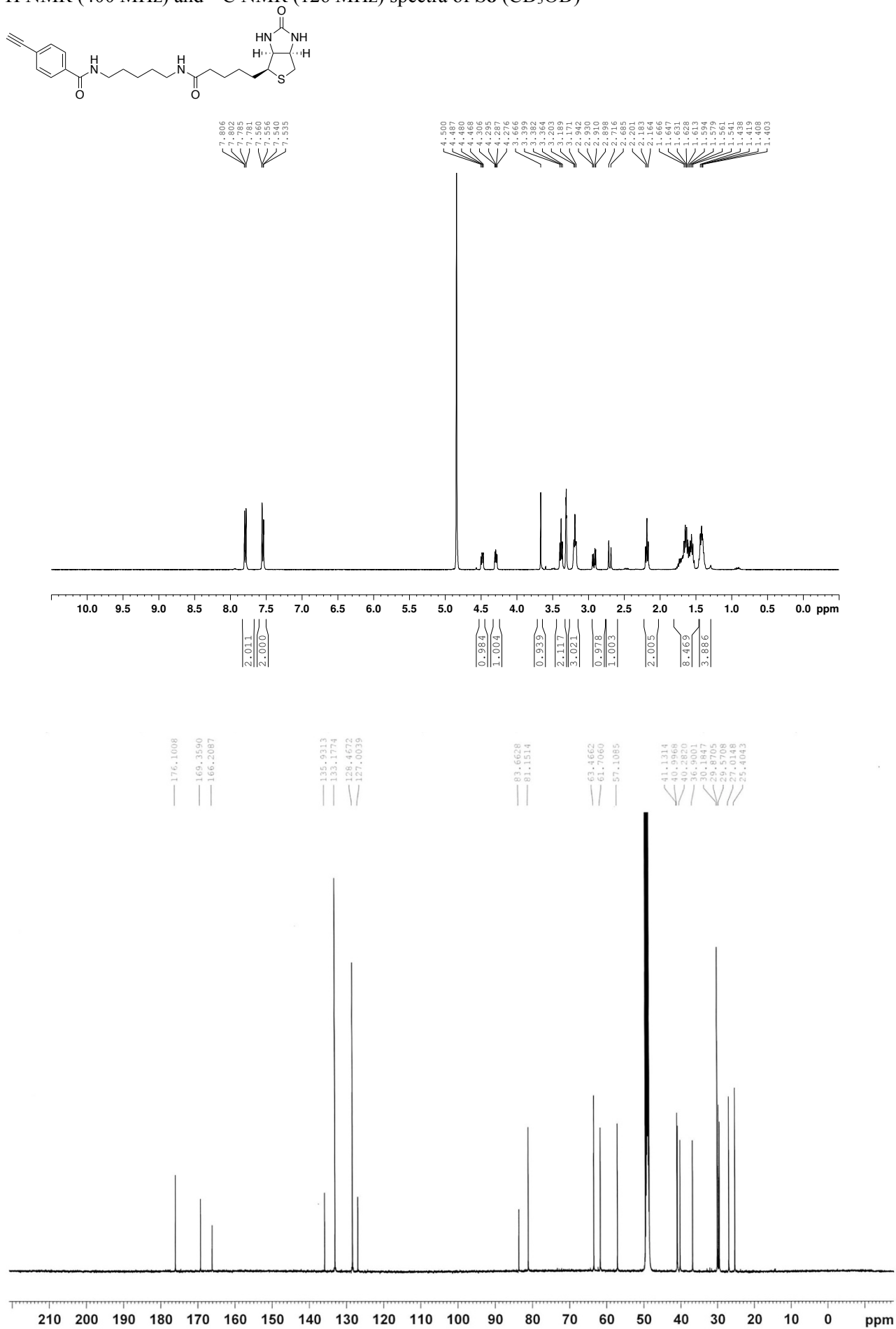


^1H NMR (400 MHz, CDCl_3) and ^{13}C NMR (126 MHz, CD_3OD) spectra of **15a**

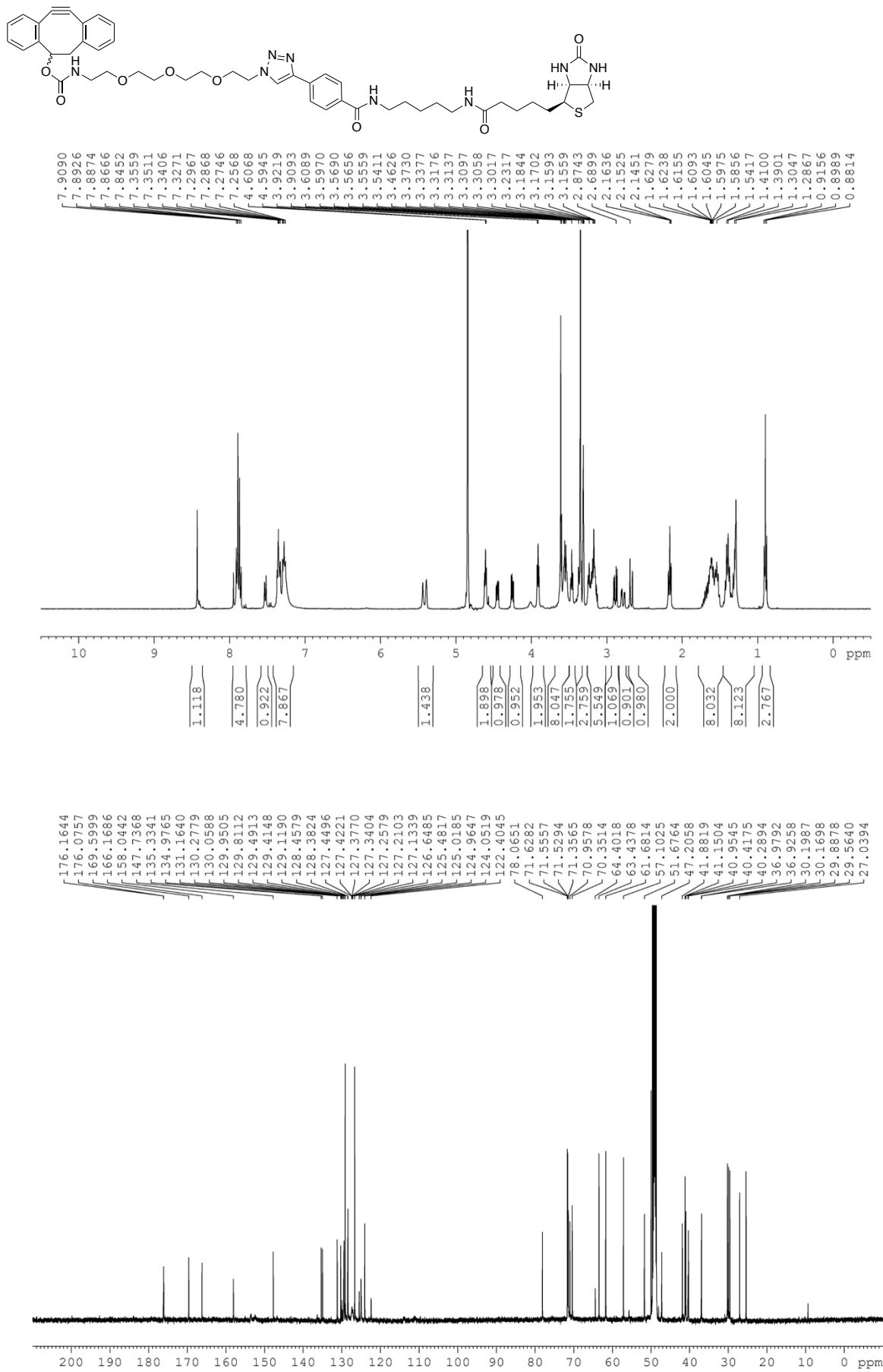


^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **15b** (CD_3OD)

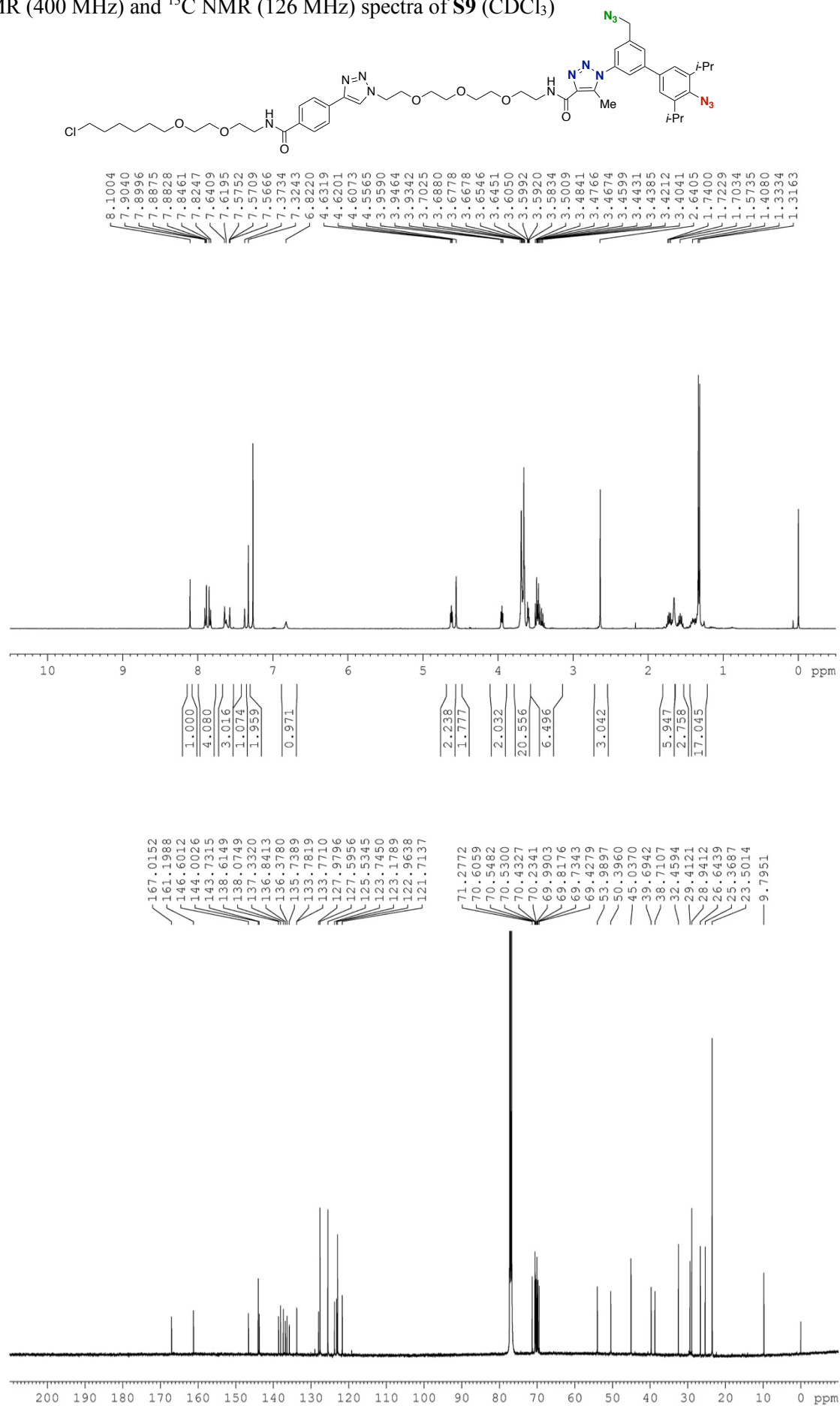


¹H NMR (400 MHz) and ¹³C NMR (126 MHz) spectra of **S8** (CD₃OD)

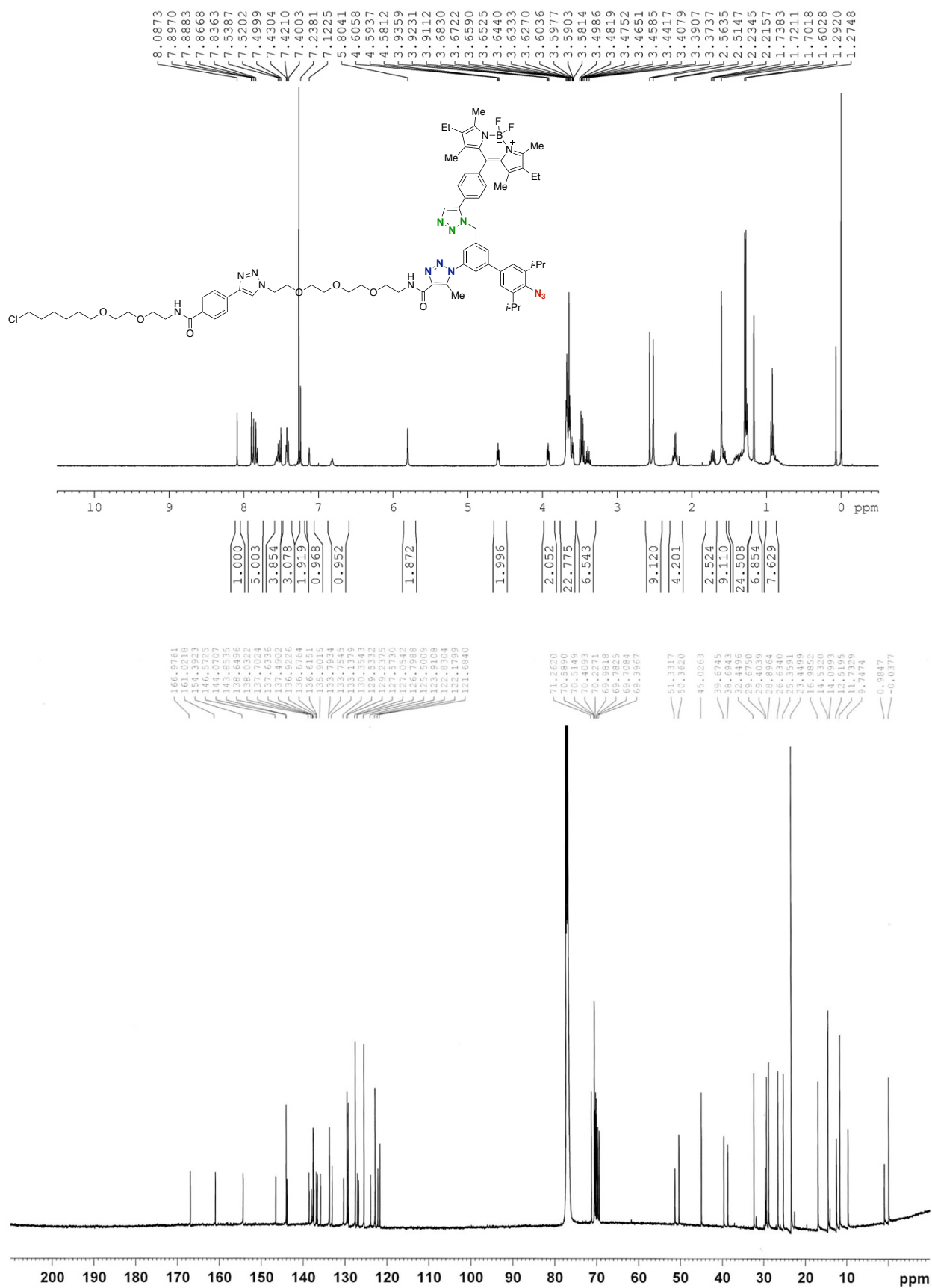
¹H NMR (400 MHz) and ¹³C NMR (126 MHz) spectra of **15c** (CD₃OD)



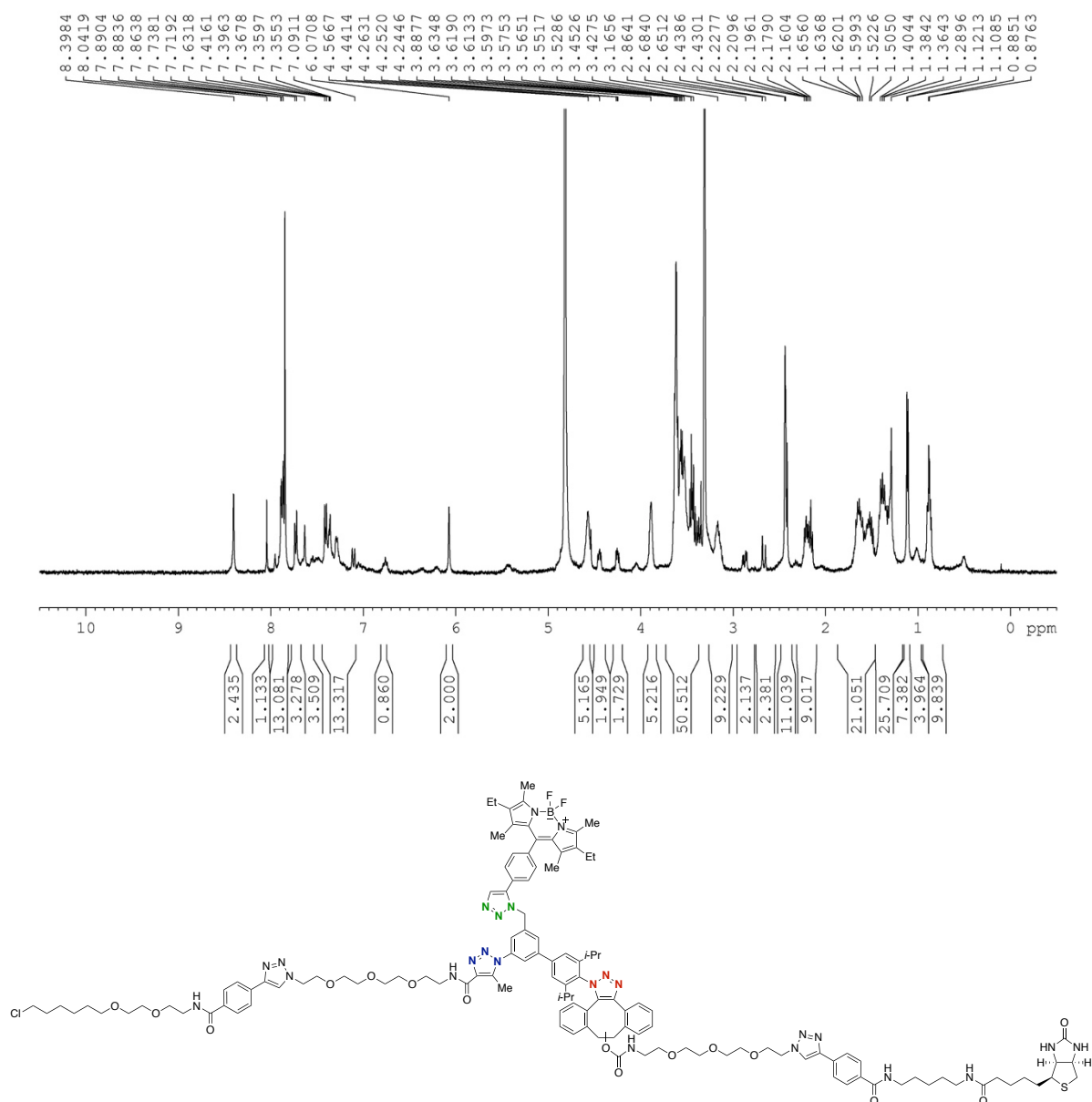
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **S9** (CDCl_3)



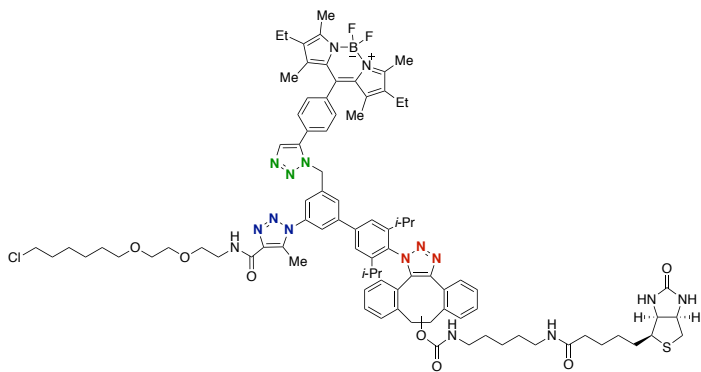
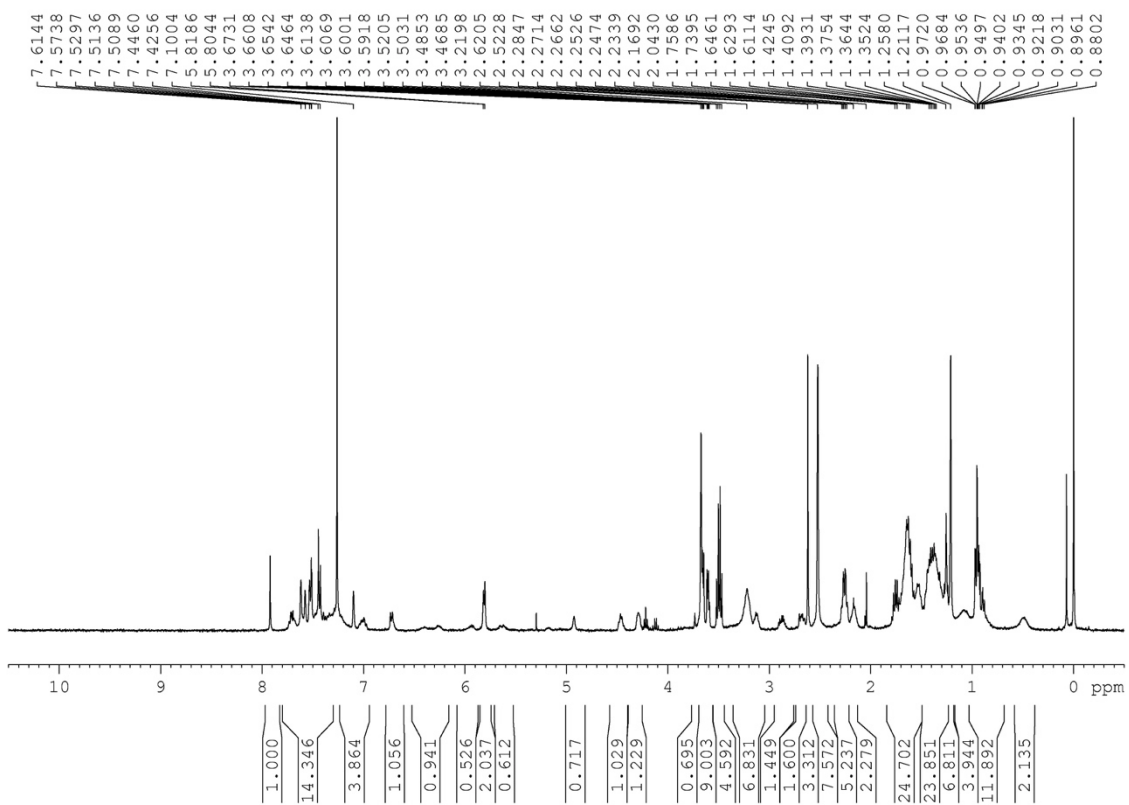
^1H NMR (400 MHz) and ^{13}C NMR (126 MHz) spectra of **S10** (CDCl_3)



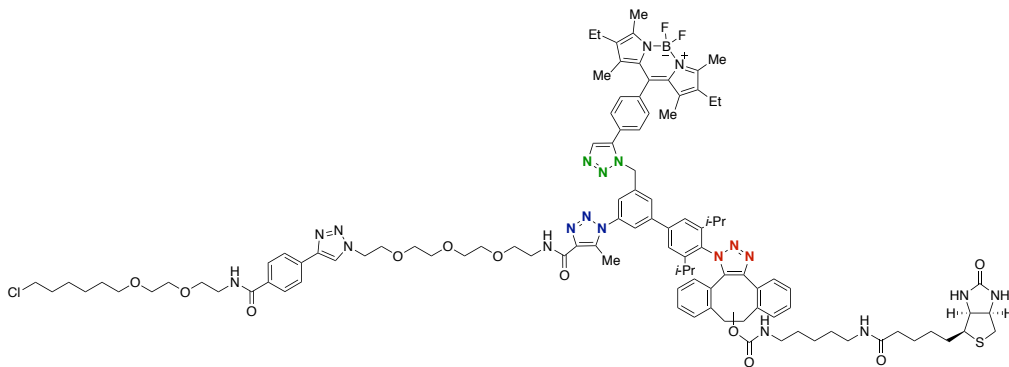
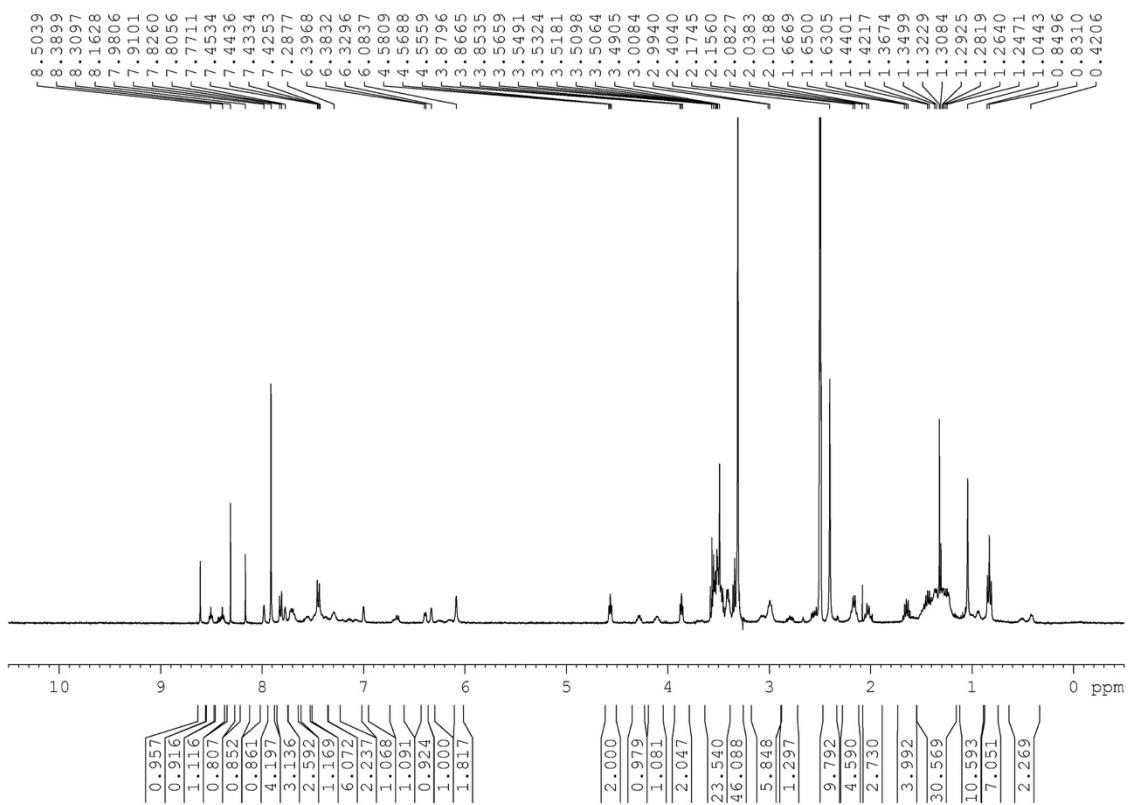
^1H NMR (400 MHz) spectrum of **16d** (CD_3OD)

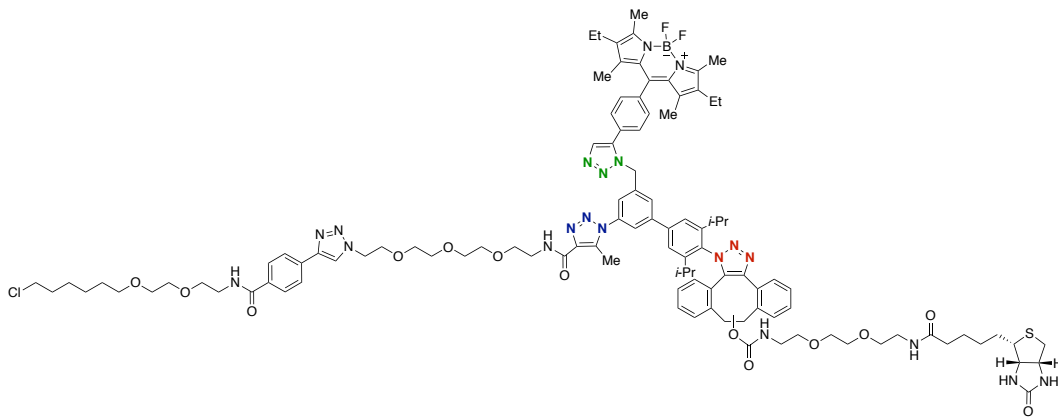
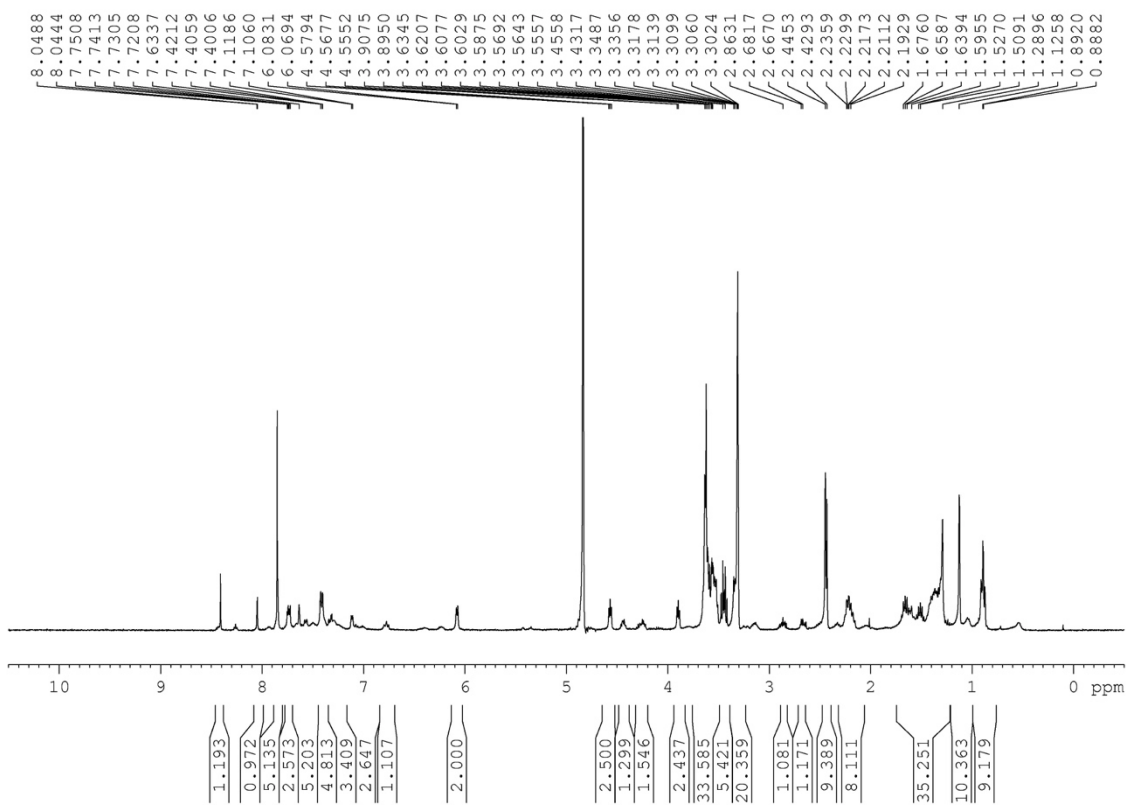


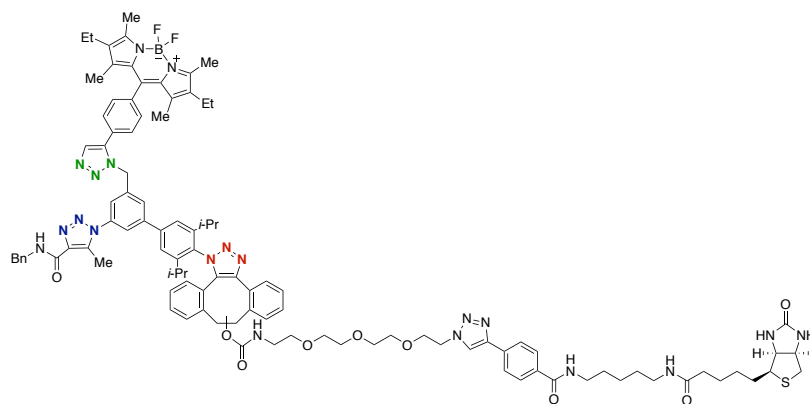
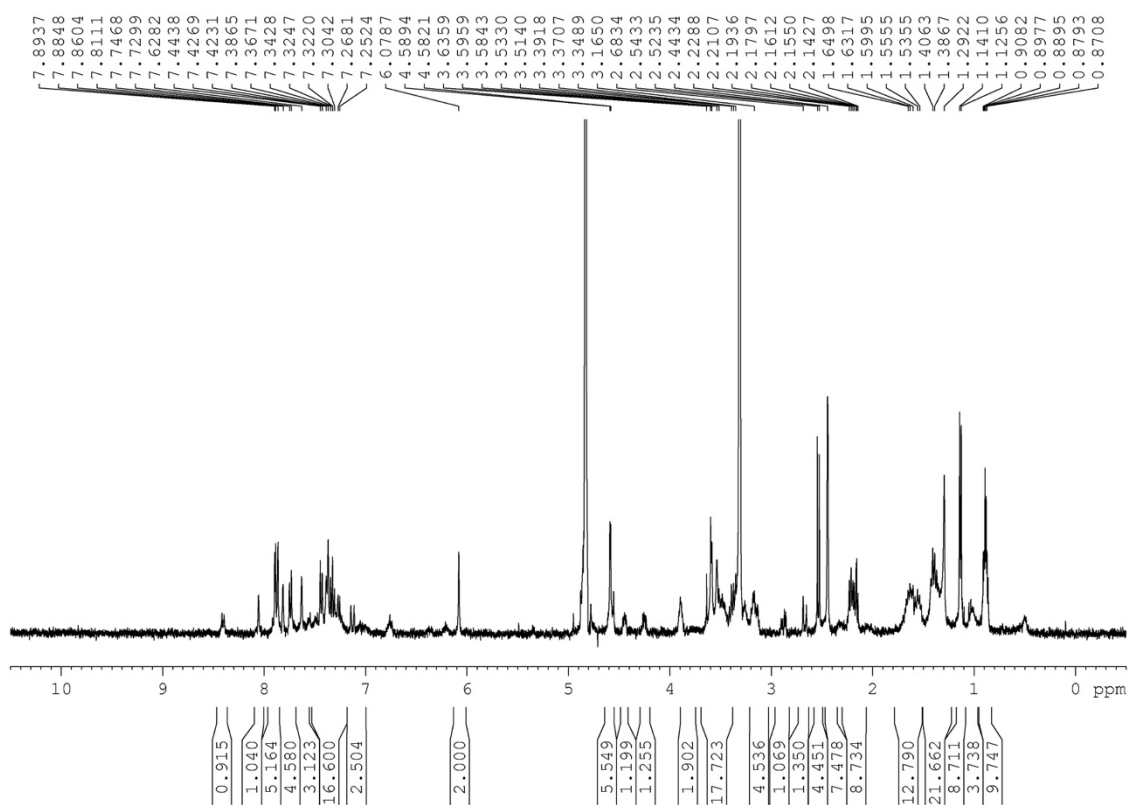
¹H NMR (400 MHz) spectrum of **16a** (DMSO-*d*₆)

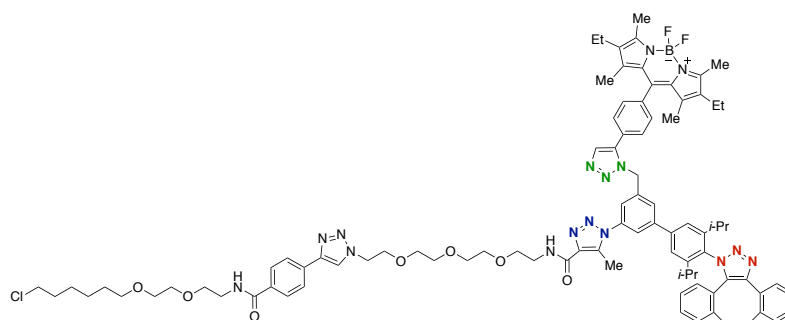
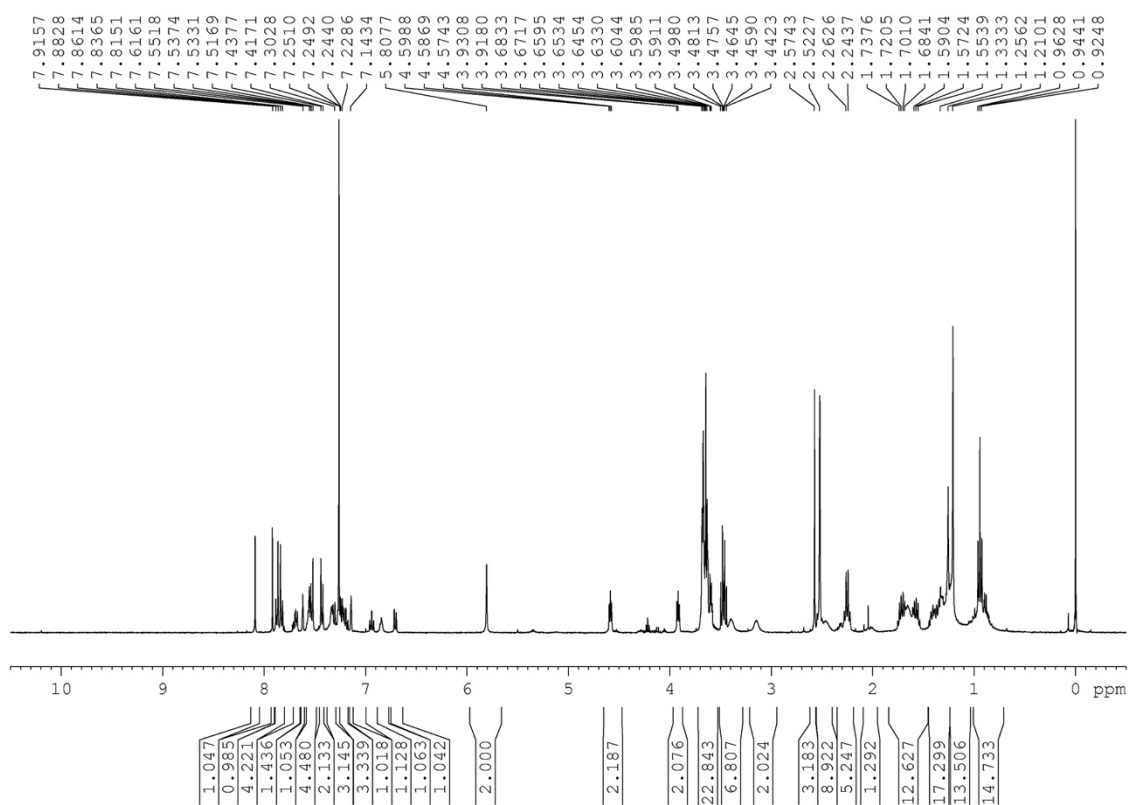


¹H NMR (400 MHz) spectrum of **16b** (DMSO-*d*₆)



¹H NMR (400 MHz) spectrum of **16c** (CD₃OD)

¹H NMR (400 MHz) spectrum of **S11** (CD₃OD)

¹H NMR (400 MHz) spectrum of **S12** (CDCl₃)

CCCCCCCCOCCOCCNC(=O)c1ccc(cc1)/N=N/c2ccc(cc2)NCCOCCOCCOCCOCCNC(=O)c3c(C)c([N+]=[N-])n3Cc4ccc(cc4)C5=CC(=CC=C5C6=CC(=CC=C6)N7C(=O)NCCOCCOCCOCCOCCNC(=O)c8ccc(cc8)/N=N/c9ccc(cc9)NC(=O)NCCCCNC(=O)CCCCC1=NC2=C(N1)NC(=O)N2C=S2)C5C(C(C)C)=C(C(C)C)N7C(=O)c3ccc(cc3)C4=CC(=CC=C4)N5C(=O)NCCOCCOCCOCCOCCNC(=O)c6ccc(cc6)/N=N/c7ccc(cc7)NCCOCCOCCOCCOCCNC(=O)c8ccc(cc8)C9=CC(=CC=C9)N10C(=O)NCCOCCOCCOCCOCCNC(=O)c11ccc(cc11)/N=N/c12ccc(cc12)NCCOCCOCCOCCOCCNC(=O)c13ccc(cc13)C14=CC(=CC=C14)N15C(=O)NCCOCCOCCOCCOCCNC(=O)c16ccc(cc16)/N=N/c17ccc(cc17)NCCOCCOCCOCCOCCNC(=O)c18ccc(cc18)C19=CC(=CC=C19)N20C(=O)NCCOCCOCCOCCOCCNC(=O)c21ccc(cc21)/N=N/c22ccc(cc22)NCCOCCOCCOCCOCCNC(=O)c23ccc(cc23)C24=CC(=CC=C24)N25C(=O)NCCOCCOCCOCCOCCNC(=O)c26ccc(cc26)/N=N/c27ccc(cc27)NCCOCCOCCOCCOCCNC(=O)c28ccc(cc28)C29=CC(=CC=C29)N30C(=O)NCCOCCOCCOCCOCCNC(=O)c31ccc(cc31)/N=N/c32ccc(cc32)NCCOCCOCCOCCOCCNC(=O)c33ccc(cc33)C34=CC(=CC=C34)N35C(=O)NCCOCCOCCOCCOCCNC(=O)c36ccc(cc36)/N=N/c37ccc(cc37)NCCOCCOCCOCCOCCNC(=O)c38ccc(cc38)C39=CC(=CC=C39)N40C(=O)NCCOCCOCCOCCOCCNC(=O)c41ccc(cc41)/N=N/c42ccc(cc42)NCCOCCOCCOCCOCCNC(=O)c43ccc(cc43)C44=CC(=CC=C44)N45C(=O)NCCOCCOCCOCCOCCNC(=O)c46ccc(cc46)/N=N/c47ccc(cc47)NCCOCCOCCOCCOCCNC(=O)c48ccc(cc48)C49=CC(=CC=C49)N50C(=O)NCCOCCOCCOCCOCCNC(=O)c51ccc(cc51)/N=N/c52ccc(cc52)NCCOCCOCCOCCOCCNC(=O)c53ccc(cc53)C54=CC(=CC=C54)N55C(=O)NCCOCCOCCOCCOCCNC(=O)c56ccc(cc56)/N=N/c57ccc(cc57)NCCOCCOCCOCCOCCNC(=O)c58ccc(cc58)C59=CC(=CC=C59)N60C(=O)NCCOCCOCCOCCOCCNC(=O)c61ccc(cc61)/N=N/c62ccc(cc62)NCCOCCOCCOCCOCCNC(=O)c63ccc(cc63)C64=CC(=CC=C64)N65C(=O)NCCOCCOCCOCCOCCNC(=O)c66ccc(cc66)/N=N/c67ccc(cc67)NCCOCCOCCOCCOCCNC(=O)c68ccc(cc68)C69=CC(=CC=C69)N70C(=O)NCCOCCOCCOCCOCCNC(=O)c71ccc(cc71)/N=N/c72ccc(cc72)NCCOCCOCCOCCOCCNC(=O)c73ccc(cc73)C74=CC(=CC=C74)N75C(=O)NCCOCCOCCOCCOCCNC(=O)c76ccc(cc76)/N=N/c77ccc(cc77)NCCOCCOCCOCCOCCNC(=O)c78ccc(cc78)C79=CC(=CC=C79)N80C(=O)NCCOCCOCCOCCOCCNC(=O)c81ccc(cc81)/N=N/c82ccc(cc82)NCCOCCOCCOCCOCCNC(=O)c83ccc(cc83)C84=CC(=CC=C84)N85C(=O)NCCOCCOCCOCCOCCNC(=O)c86ccc(cc86)/N=N/c87ccc(cc87)NCCOCCOCCOCCOCCNC(=O)c88ccc(cc88)C89=CC(=CC=C89)N90C(=O)NCCOCCOCCOCCOCCNC(=O)c91ccc(cc91)/N=N/c92ccc(cc92)NCCOCCOCCOCCOCCNC(=O)c93ccc(cc93)C94=CC(=CC=C94)N95C(=O)NCCOCCOCCOCCOCCNC(=O)c96ccc(cc96)/N=N/c97ccc(cc97)NCCOCCOCCOCCOCCNC(=O)c98ccc(cc98)C99=CC(=CC=C99)N100C(=O)NCCOCCOCCOCCOCCNC(=O)c101ccc(cc101)/N=N/c102ccc(cc102)NCCOCCOCCOCCOCCNC(=O)c103ccc(cc103)C104=CC(=CC=C104)N105C(=O)NCCOCCOCCOCCOCCNC(=O)c106ccc(cc106)/N=N/c107ccc(cc107)NCCOCCOCCOCCOCCNC(=O)c108ccc(cc108)C109=CC(=CC=C109)N110C(=O)NCCOCCOCCOCCOCCNC(=O)c111ccc(cc111)/N=N/c112ccc(cc112)NCCOCCOCCOCCOCCNC(=O)c113ccc(cc113)C114=CC(=CC=C114)N115C(=O)NCCOCCOCCOCCOCCNC(=O)c116ccc(cc116)/N=N/c117ccc(cc117)NCCOCCOCCOCCOCCNC(=O)c118ccc(cc118)C119=CC(=CC=C119)N120C(=O)NCCOCCOCCOCCOCCNC(=O)c121ccc(cc121)/N=N/c122ccc(cc122)NCCOCCOCCOCCOCCNC(=O)c123ccc(cc123)C124=CC(=CC=C124)N125C(=O)NCCOCCOCCOCCOCCNC(=O)c126ccc(cc126)/N=N/c127ccc(cc127)NCCOCCOCCOCCOCCNC(=O)c128ccc(cc128)C129=CC(=CC=C129)N130C(=O)NCCOCCOCCOCCOCCNC(=O)c131ccc(cc131)/N=N/c132ccc(cc132)NCCOCCOCCOCCOCCNC(=O)c133ccc(cc133)C134=CC(=CC=C134)N135C(=O)NCCOCCOCCOCCOCCNC(=O)c136ccc(cc136)/N=N/c137ccc(cc137)NCCOCCOCCOCCOCCNC(=O)c138ccc(cc138)C139=CC(=CC=C139)N140C(=O)NCCOCCOCCOCCOCCNC(=O)c141ccc(cc141)/N=N/c142ccc(cc142)NCCOCCOCCOCCOCCNC(=O)c143ccc(cc143)C144=CC(=CC=C144)N145C(=O)NCCOCCOCCOCCOCCNC(=O)c146ccc(cc146)/N=N/c147ccc(cc147)NCCOCCOCCOCCOCCNC(=O)c148ccc(cc148)C149=CC(=CC=C149)N150C(=O)NCCOCCOCCOCCOCCNC(=O)c151ccc(cc151)/N=N/c152ccc(cc152)NCCOCCOCCOCCOCCNC(=O)c153ccc(cc153)C154=CC(=CC=C154)N155C(=O)NCCOCCOCCOCCOCCNC(=O)c156ccc(cc156)/N=N/c157ccc(cc157)NCCOCCOCCOCCOCCNC(=O)c158ccc(cc158)C159=CC(=CC=C159)N160C(=O)NCCOCCOCCOCCOCCNC(=O)c161ccc(cc161)/N=N/c162ccc(cc162)NCCOCCOCCOCCOCCNC(=O)c163ccc(cc163)C164=CC(=CC=C164)N165C(=O)NCCOCCOCCOCCOCCNC(=O)c166ccc(cc166)/N=N/c167ccc(cc167)NCCOCCOCCOCCOCCNC(=O)c168ccc(cc168)C169=CC(=CC=C169)N170C(=O)NCCOCCOCCOCCOCCNC(=O)c171ccc(cc171)/N=N/c172ccc(cc172)NCCOCCOCCOCCOCCNC(=O)c173ccc(cc173)C174=CC(=CC=C174)N175C(=O)NCCOCCOCCOCCOCCNC(=O)c176ccc(cc176)/N=N/c177ccc(cc177)NCCOCCOCCOCCOCCNC(=O)c178ccc(cc178)C179=CC(=CC=C179)N180C(=O)NCCOCCOCCOCCOCCNC(=O)c181ccc(cc181)/N=N/c182ccc(cc182)NCCOCCOCCOCCOCCNC(=O)c183ccc(cc183)C184=CC(=CC=C184)N185C(=O)NCCOCCOCCOCCOCCNC(=O)c186ccc(cc186)/N=N/c187ccc(cc187)NCCOCCOCCOCCOCCNC(=O)c188ccc(cc188)C189=CC(=CC=C189)N190C(=O)NCCOCCOCCOCCOCCNC(=O)c191ccc(cc191)/N=N/c192ccc(cc192)NCCOCCOCCOCCOCCNC(=O)c193ccc(cc193)C194=CC(=CC=C194)N195C(=O)NCCOCCOCCOCCOCCNC(=O)c196ccc(cc196)/N=N/c197ccc(cc197)NCCOCCOCCOCCOCCNC(=O)c198ccc(cc198)C199=CC(=CC=C199)N200C(=O)NCCOCCOCCOCCOCCNC(=O)c201ccc(cc201)/N=N/c202ccc(cc202)NCCOCCOCCOCCOCCNC(=O)c203ccc(cc203)C204=CC(=CC=C204)N205C(=O)NCCOCCOCCOCCOCCNC(=O)c206ccc(cc206)/N=N/c207ccc(cc207)NCCOCCOCCOCCOCCNC(=O)c208ccc(cc208)C209=CC(=CC=C209)N210C(=O)NCCOCCOCCOCCOCCNC(=O)c211ccc(cc211)/N=N/c212ccc(cc212)NCCOCCOCCOCCOCCNC(=O)c213ccc(cc213)C214=CC(=CC=C214)N215C(=O)NCCOCCOCCOCCOCCNC(=O)c216ccc(cc216)/N=N/c217ccc(cc217)NCCOCCOCCOCCOCCNC(=O)c218ccc(cc218)C219=CC(=CC=C219)N220C(=O)NCCOCCOCCOCCOCCNC(=O)c221ccc(cc221)/N=N/c222ccc(cc222)NCCOCCOCCOCCOCCNC(=O)c223ccc(cc223)C224=CC(=CC=C224)N225C(=O)NCCOCCOCCOCCOCCNC(=O)c226ccc(cc226)/N=N/c227ccc(cc227)NCCOCCOCCOCCOCCNC(=O)c228ccc(cc228)C229=CC(=CC=C229)N230C(=O)NCCOCCOCCOCCOCCNC(=O)c231ccc(cc231)/N=N/c232ccc(cc232)NCCOCCOCCOCCOCCNC(=O)c233ccc(cc233)C234=CC(=CC=C234)N235C(=O)NCCOCCOCCOCCOCCNC(=O)c236ccc(cc236)/N=N/c237ccc(cc237)NCCOCCOCCOCCOCCNC(=O)c238ccc(cc238)C239=CC(=CC=C239)N240C(=O)NCCOCCOCCOCCOCCNC(=O)c241ccc(cc241)/N=N/c242ccc(cc242)NCCOCCOCCOCCOCCNC(=O)c243ccc(cc243)C244=CC(=CC=C244)N245C(=O)NCCOCCOCCOCCOCCNC(=O)c246ccc(cc246)/N=N/c247ccc(cc247)NCCOCCOCCOCCOCCNC(=O)c248ccc(cc248)C249=CC(=CC=C249)N250C(=O)NCCOCCOCCOCCOCCNC(=O)c2