Supporting Information

Palladium-mediated Synthesis of a Near-IR Fluorescent K+ Sensor

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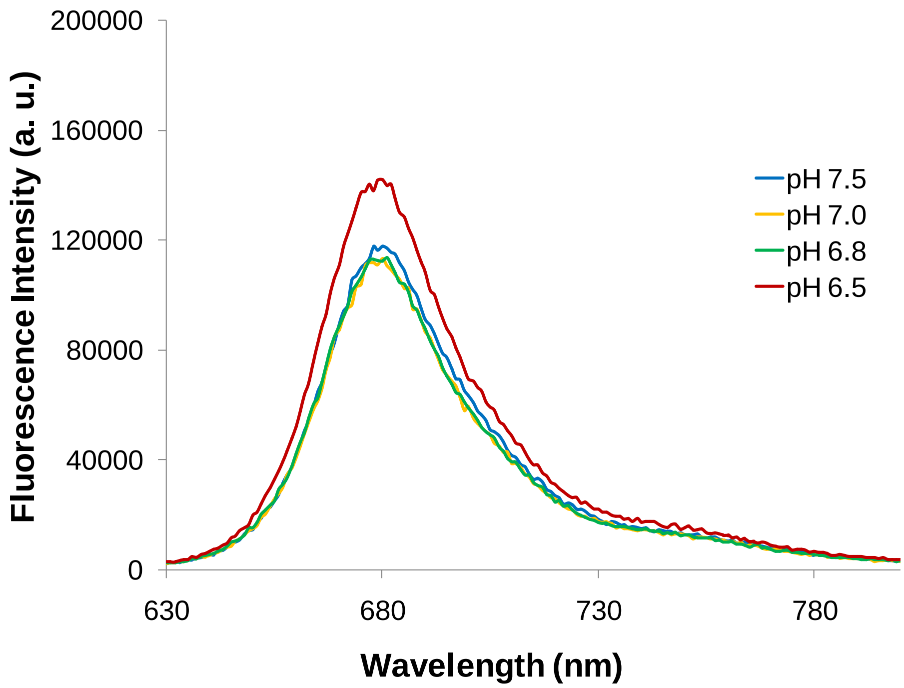
Effect of pH on the fluorescence response of K_{NIR}-1. A 15 μL aliquot of a 2.8 mM **K_{NIR}-1** stock solution was transferred to a quartz cuvette containing 3.0 mL of HEPES buffer solution to obtain a 14 μM **K_{NIR}-1** solution (0.5% vehicle concentration), and its fluorescence was recorded. A 30 μL aliquot of a 0.50 M KCl stock solution was added to above solution to obtain a 5.0mM K^+ solution. The pH of the above solution was increased to 7.8 by adding 0.1 M NaOH (aq). The pH of the above solution was then adjusted to 7.0, 6.8 and 6.0 by adding 0.1 M HCl (aq). Fluorescence intensity of the solution was measured at each pH.

Figure S1. Effect of pH on fluorescence emission of **K_{NIR}-1** (10 mM HEPES, 5 mM K^+). The pH was adjusted by adding either 0.1 M HCl or 0.1 M NaOH.



Quantum yield of K_{NIR}-1. The fluorescence quantum yield (Φ_f^i) was calculated according to equation **1**, using the absorption factor $f_x(\lambda_{ex})$ and the integrated fluorescence response $F^x(\lambda_{em})$ of both sample ($x = i$) and standard ($x = s$). The refractive index of solvent for sample was assumed to be equal to that of standard ($n_i = n_x$). The absorption factor $f_x(\lambda_{ex})$ was calculated using equation **2** where $A_x(\lambda_{ex})$ is the absorbance of sample ($x = i$) and standard ($x = s$) at the excitation wavelength.

$$\Phi_f^i = \Phi_f^s \frac{f_s(\lambda_{ex})}{f_i(\lambda_{ex})} \frac{\int \lambda_{em} F^i(\lambda_{em})}{\int \lambda_{em} F^s(\lambda_{em})} \frac{n_i^2}{n_s^2} \quad (\mathbf{1})$$

$$f_x(\lambda_{ex}) = 1 - 10^{-A_x(\lambda_{ex})} \quad (\mathbf{2})$$

A 9.3 mM stock solution of the fluorescence reference standard was prepared by dissolving 1.2 mg of oxazine-170 in 300 μL of acetonitrile. The absorbance and fluorescence emission (630 – 800 nM; 620 nm excitation) of a 14 μM oxazine-170 solution in 10 mM HEPES pH7.4 in the presence (200 mM) and absence (0 mM) of K^+. The quantum yields for **K_{NIR}-1** for apo: Φ = 0.0597 ± 0.0003 and K^+ bound: Φ = 0.289 ± 0.005 were determined from three experiments ± SEM. The vehicle (acetonitrile) concentration was 0.5% for all measurements.

Cell culture and DNA transfections: Chinese Hamster Ovary (CHO-K1) cells were cultured in F-12K nutrient mixture (Gibco – Invitrogen). All media was supplemented with 10% fetal bovine serum (Hyclone) and 102 U/mL penicillin/streptomycin (Gibco – Invitrogen). Cells were plated at 60-75% confluency in 35 mm dishes. After 24 h, cells were transiently transfected at RT with 1 μg of either empty plasmid DNA (pCDNA) or Shaker-IR DNA with 5 μL Lipofectamine (Invitrogen). Voltage-clamp fluorometry was performed 24 h post-transfection.

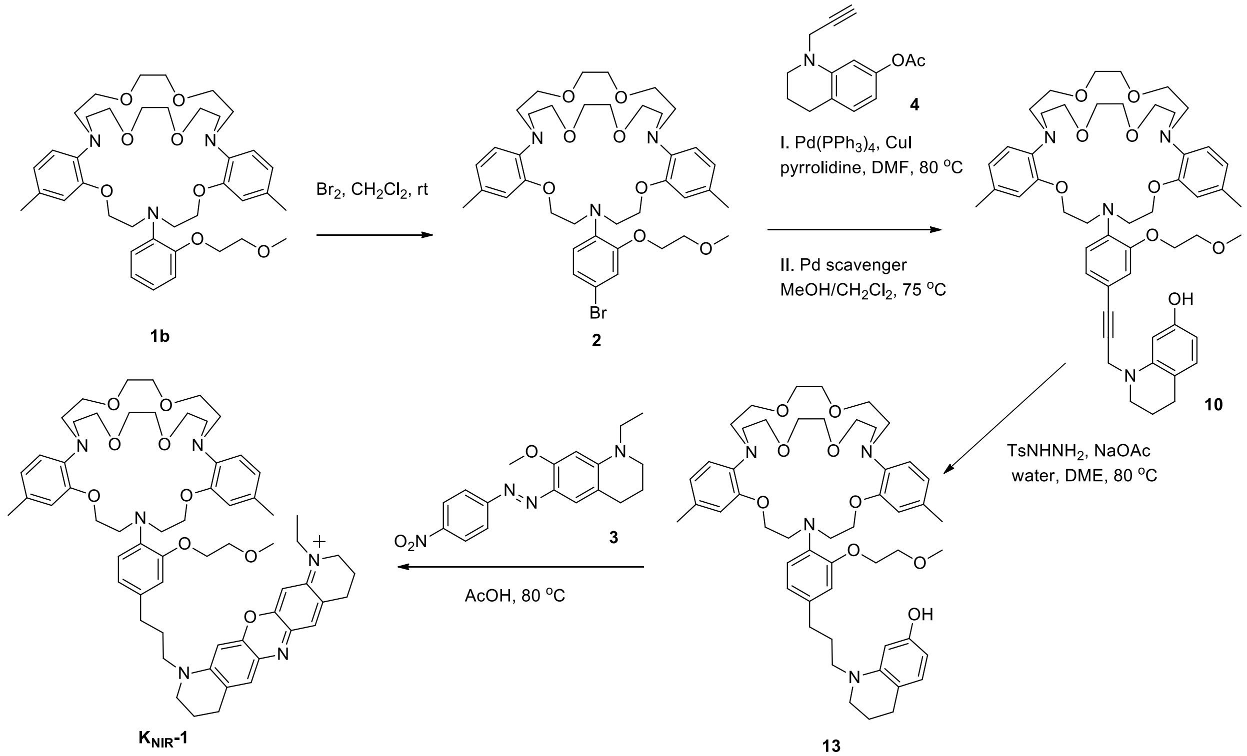


Figure S2. Synthesis of near-IR fluorescent K^+ sensor, $\mathbf{K_{NIR}}$**-1**

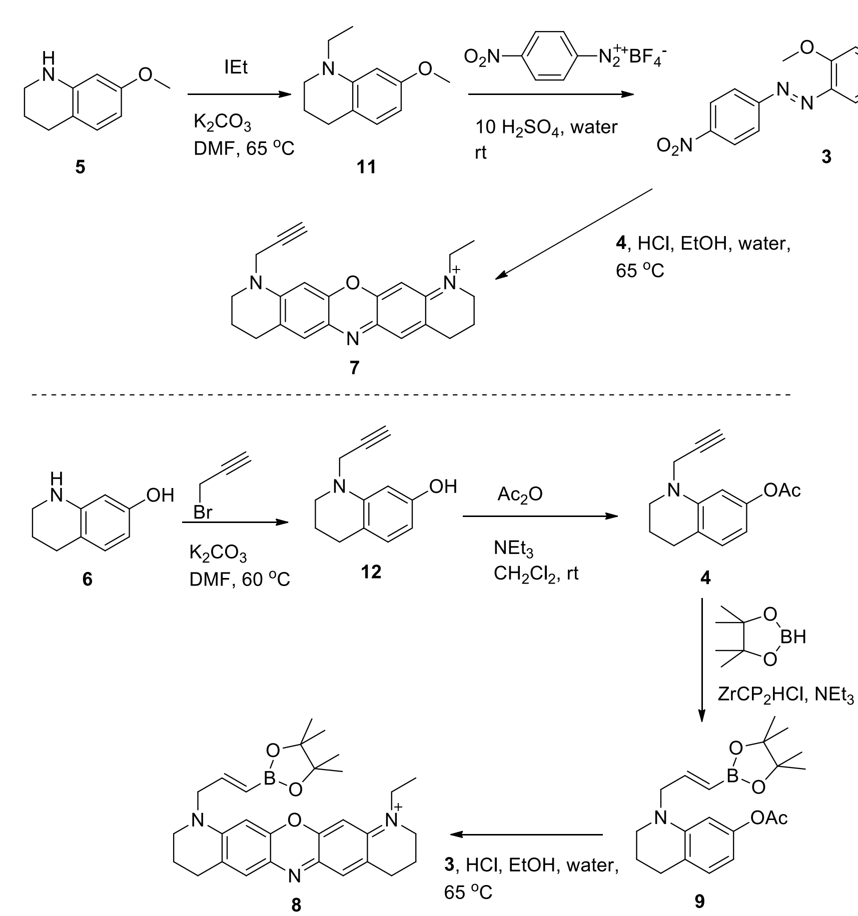
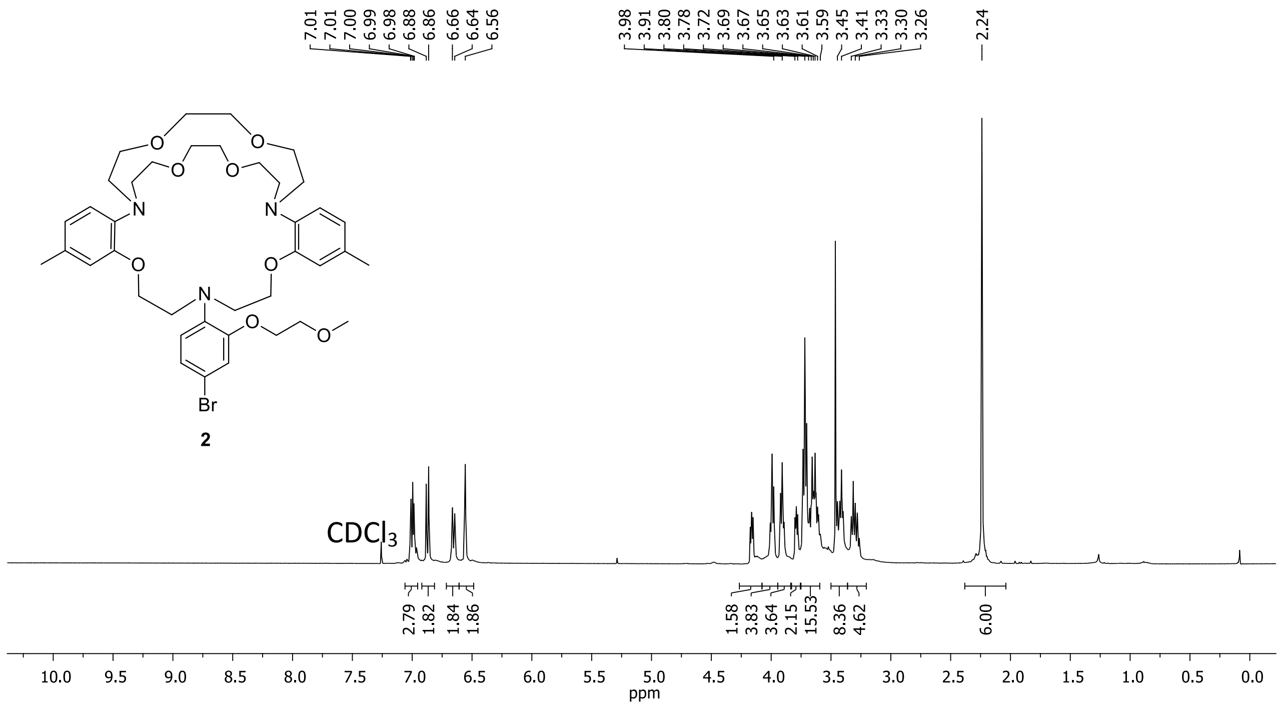


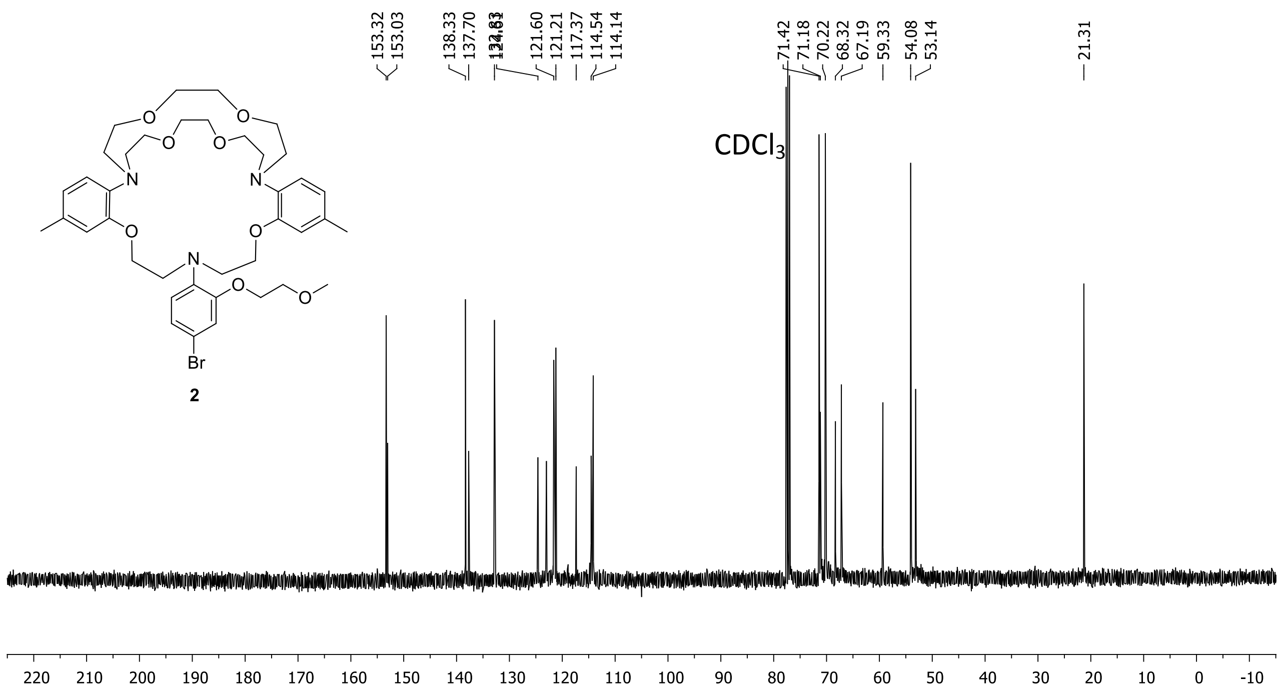
Figure S3. Synthesis of oxazine dyes and precursors for palladium-based coupling reactions

NMR spectra

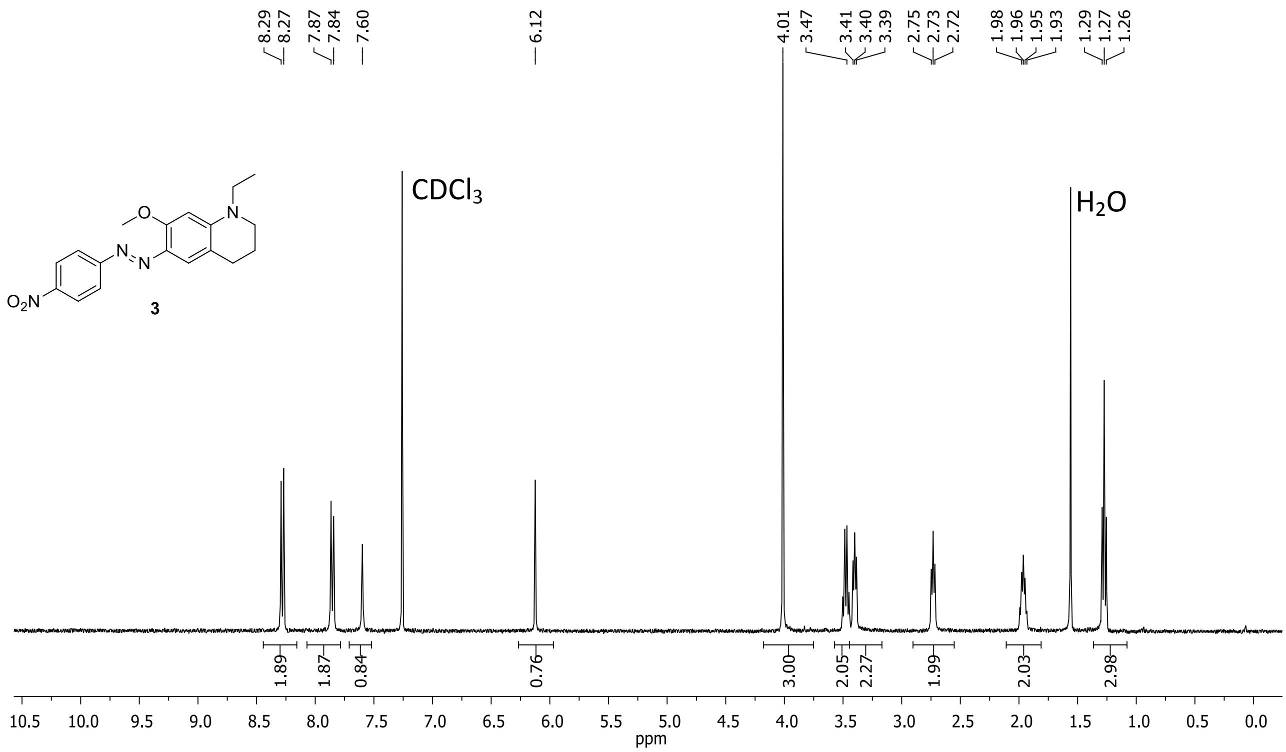
^{1}H NMR of compound **2** in $CDCl_3$



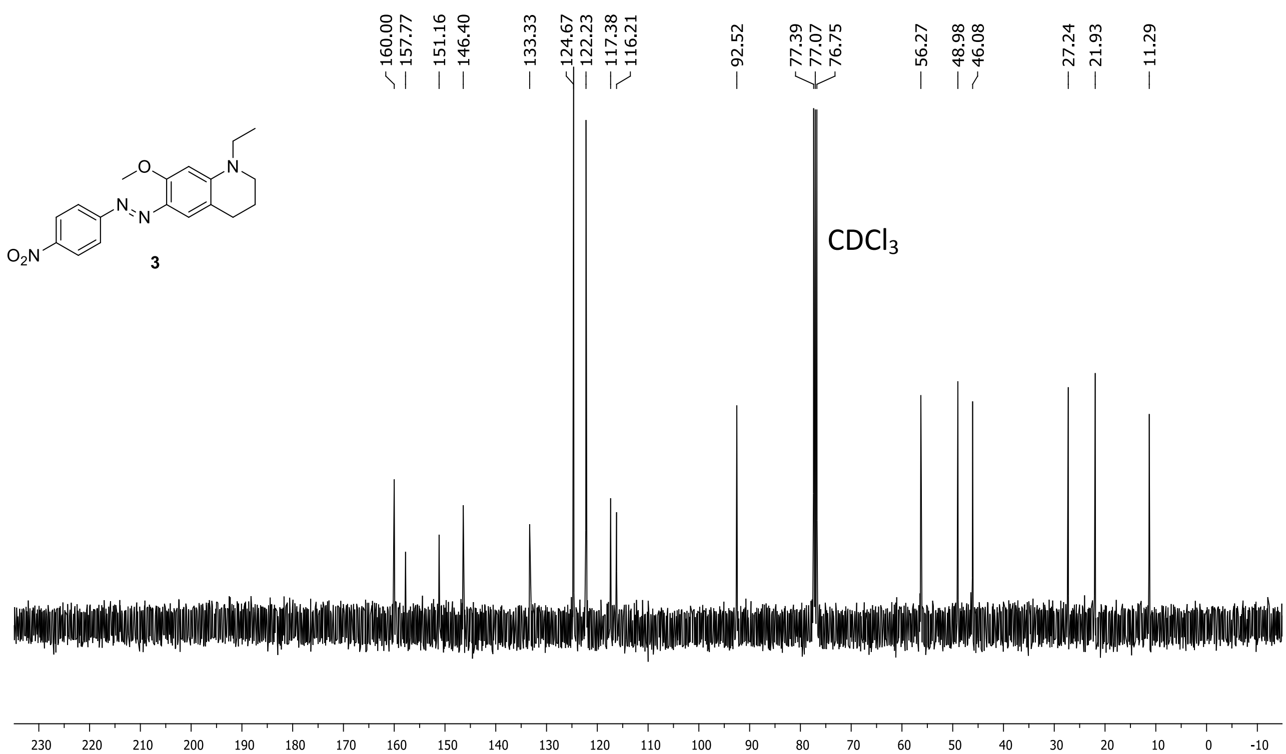
^{13}C NMR of compound **2** in $CDCl_3$



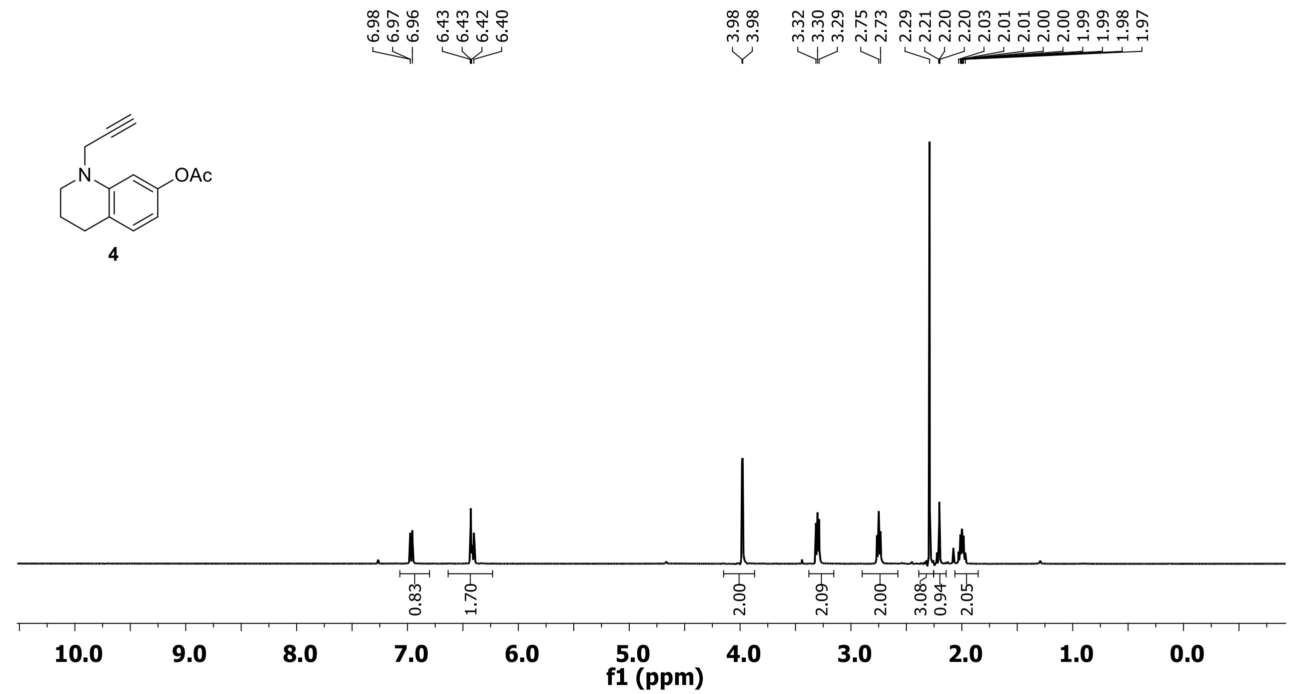
^{1}H NMR of compound **3** in $CDCl_3$



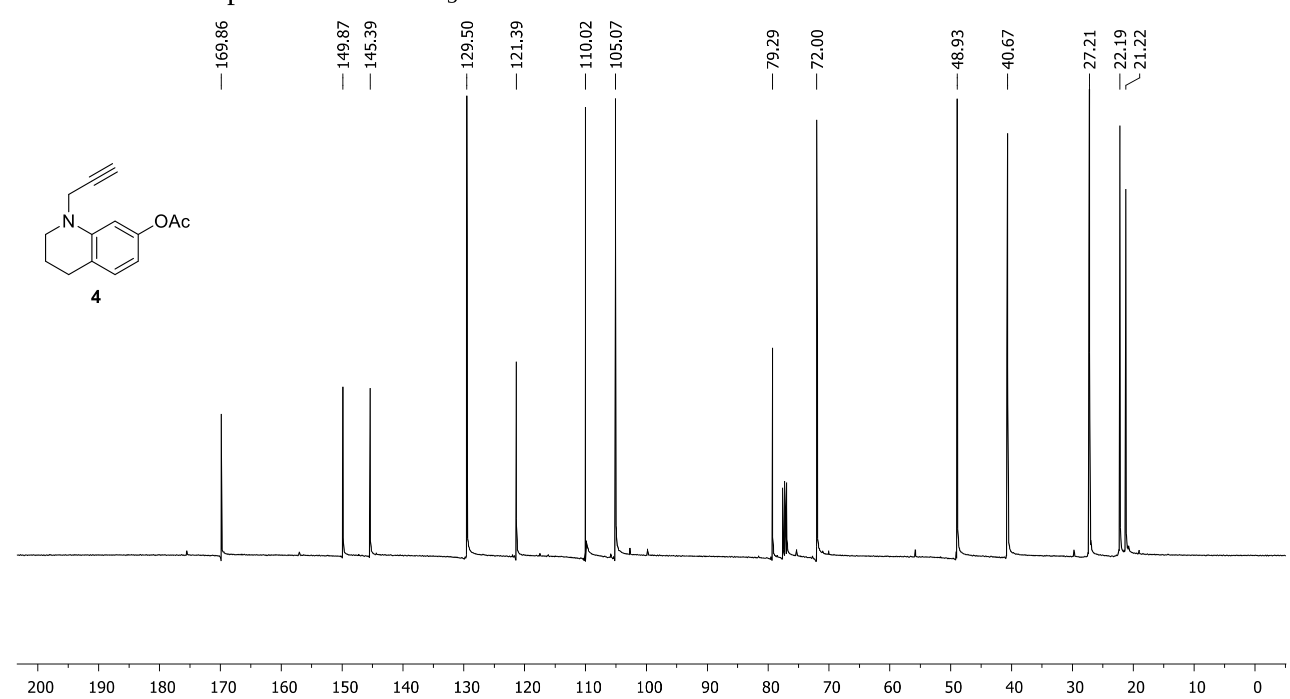
^{13}C NMR of compound **3** in $CDCl_3$



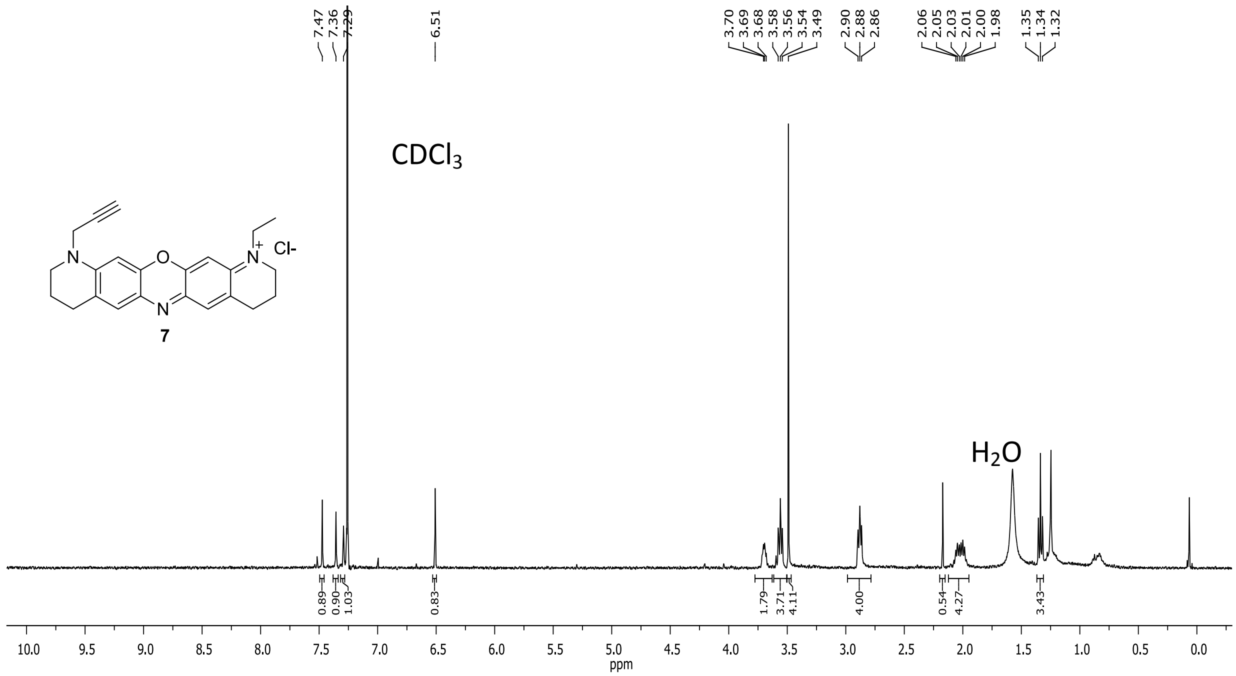
^{1}H NMR compound **4** in $CDCl_3$



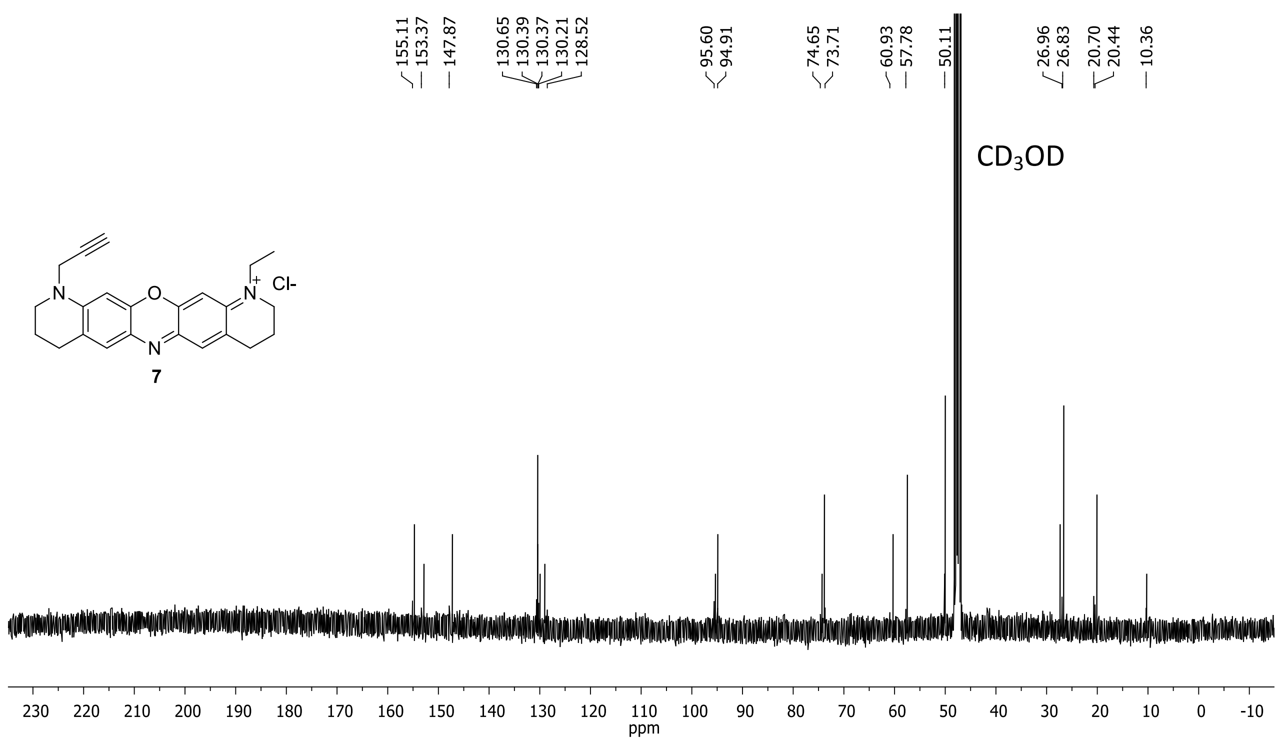
^{13}C NMR of compound **4** in $CDCl_3$



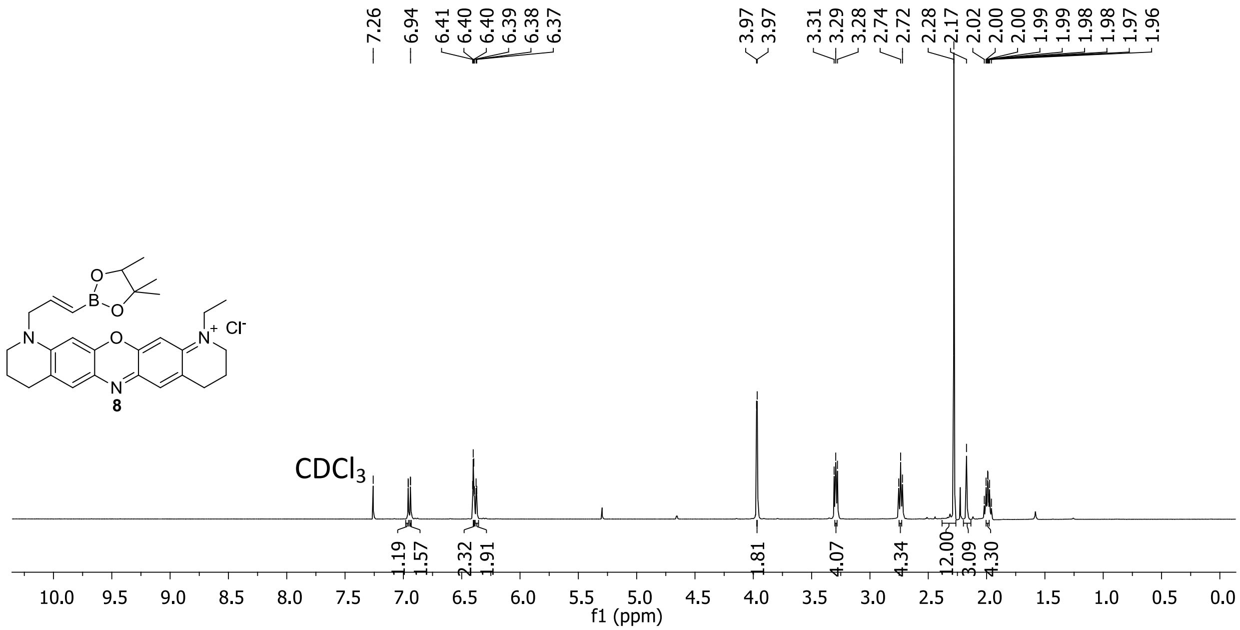
^{1}H NMR of compound **7chloride** in $CDCl_3$



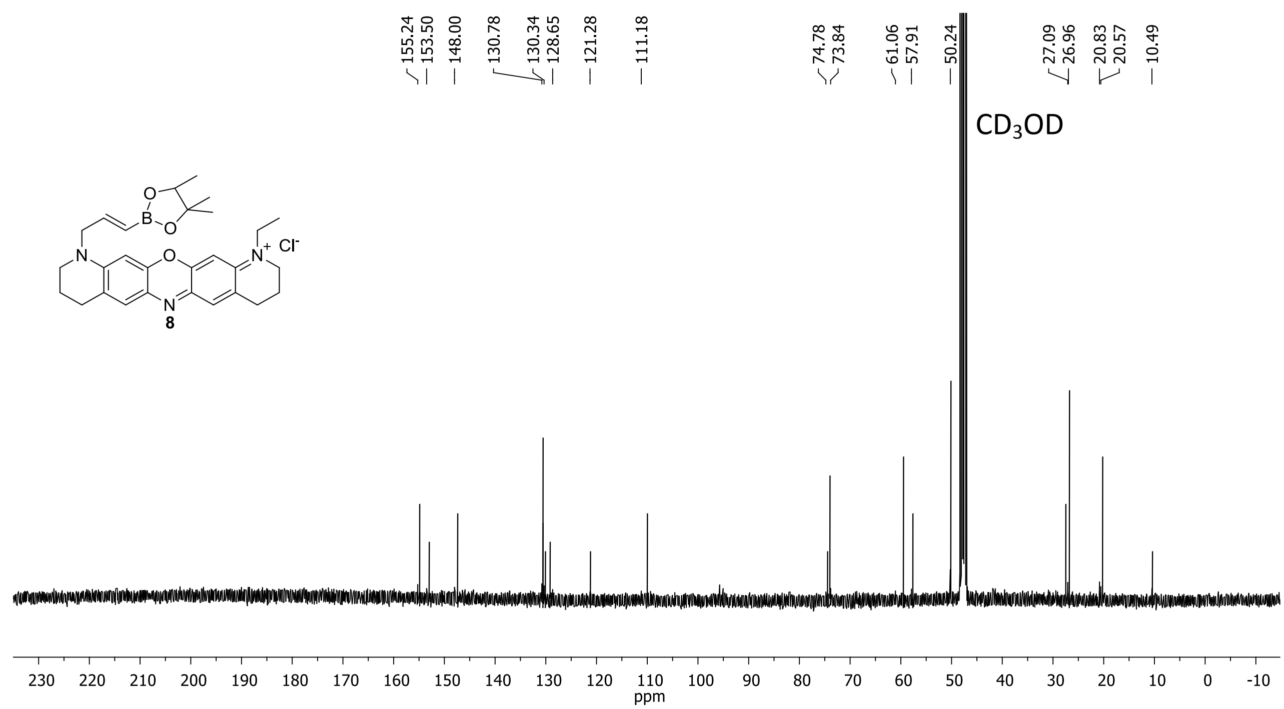
^{13}C NMR of compound **7 chloride** in CD_3OD



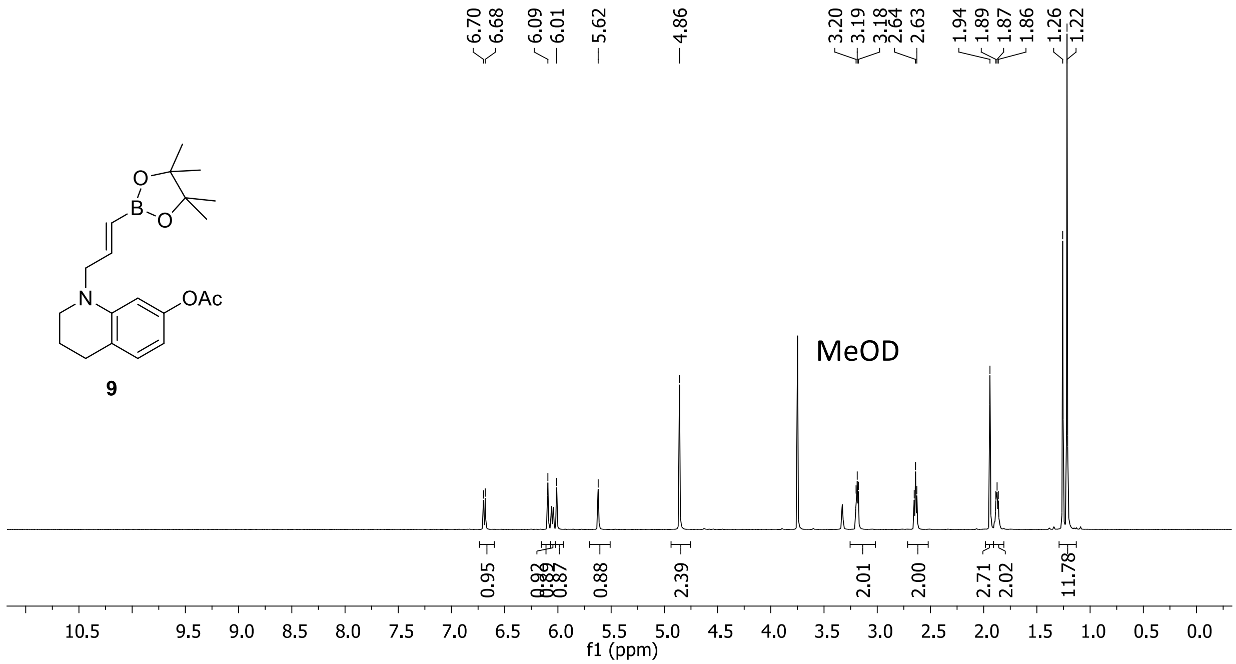
^{1}H NMR of compound **8** in $CDCl_3$



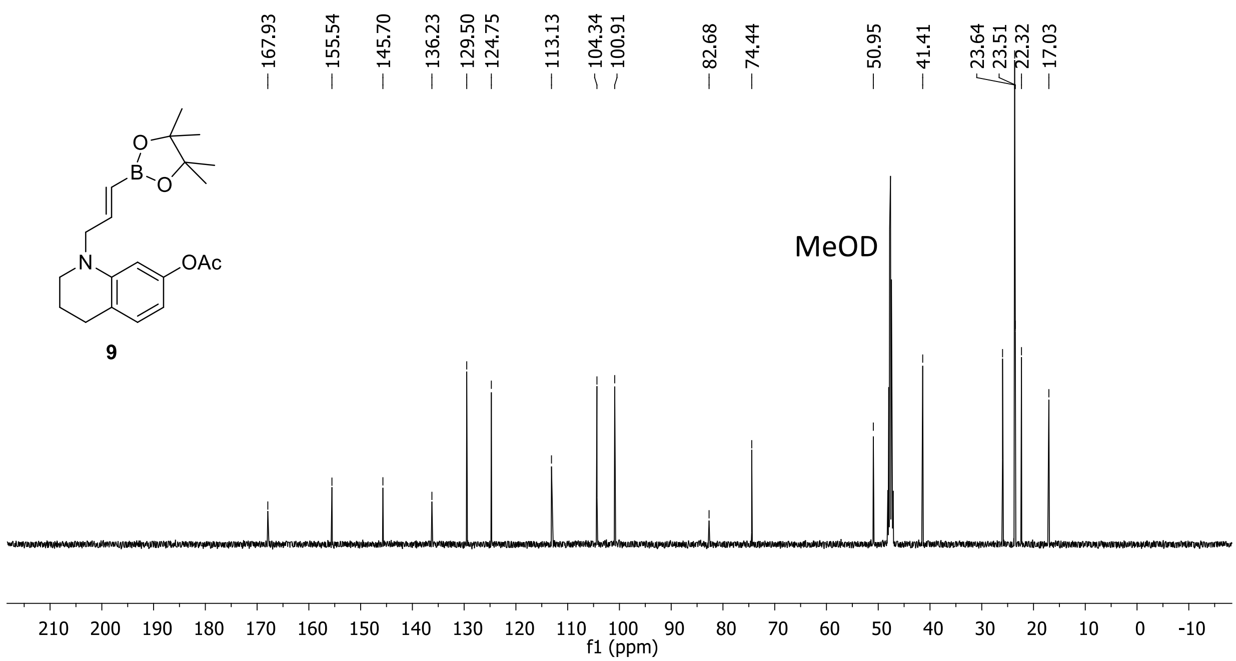
^{13}C NMR of compound **8** in CD_3OD



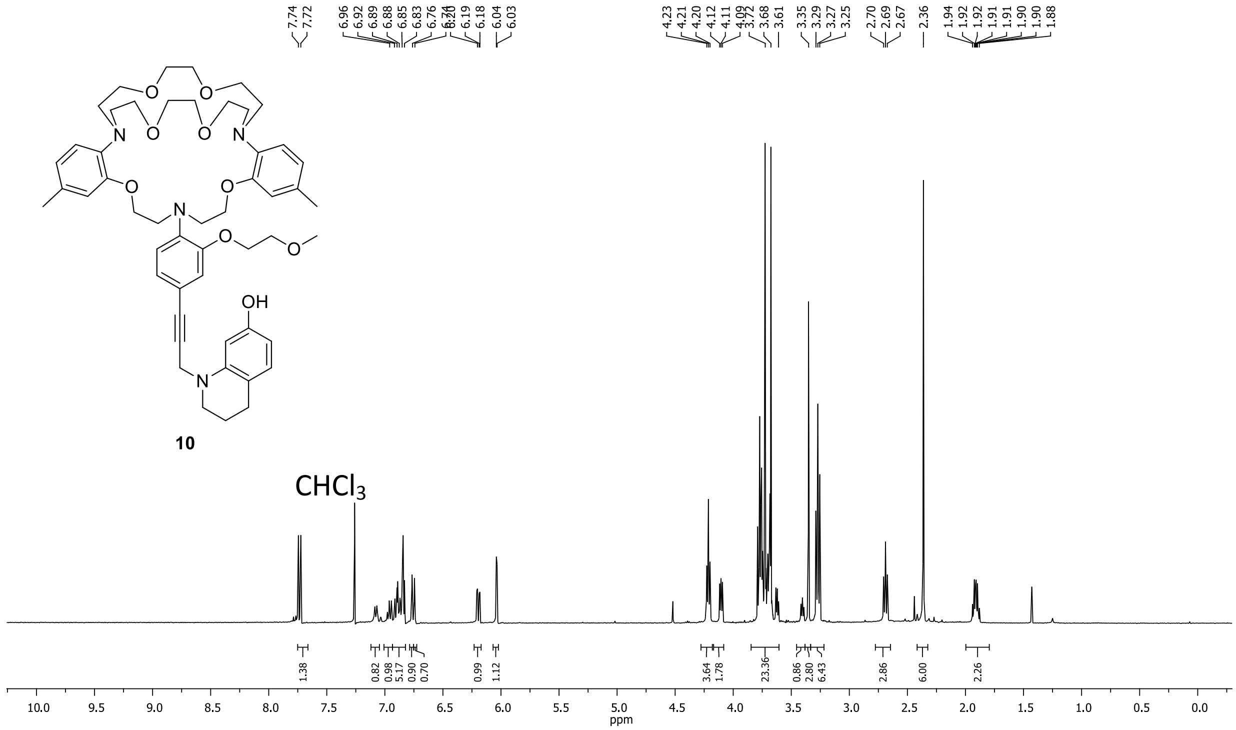
[1]H NMR of compound **9** in MeOD



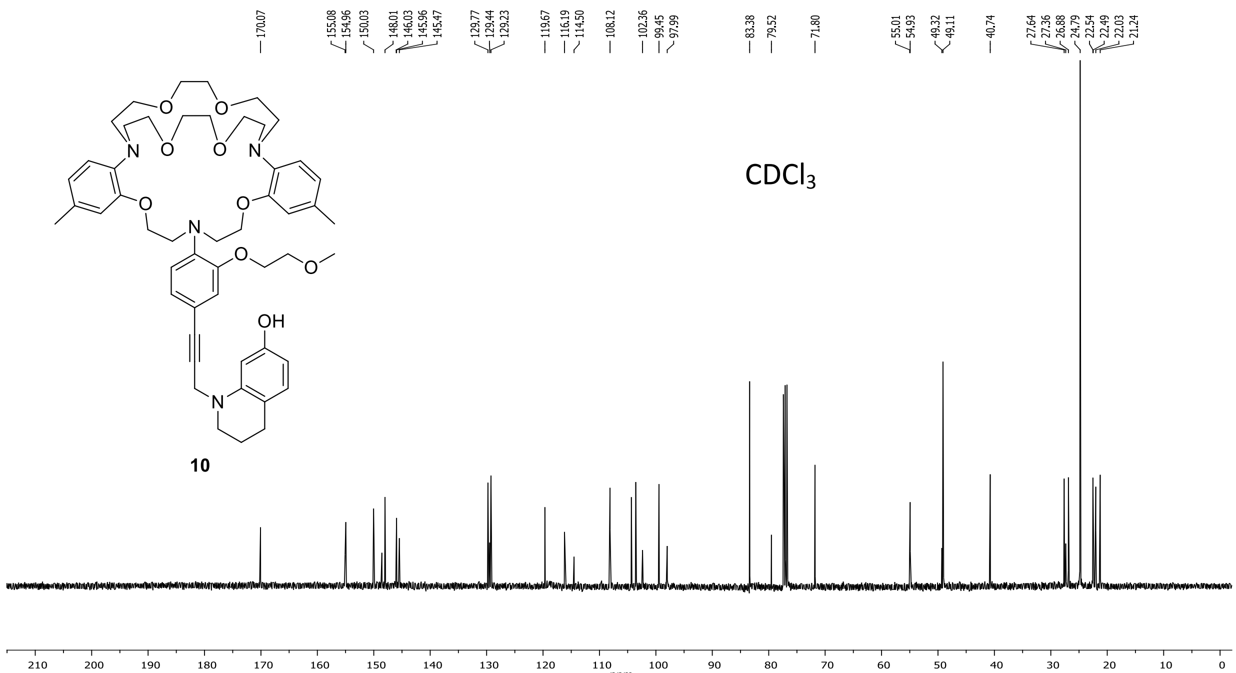
[13]C NMR of compound **9** in MeOD



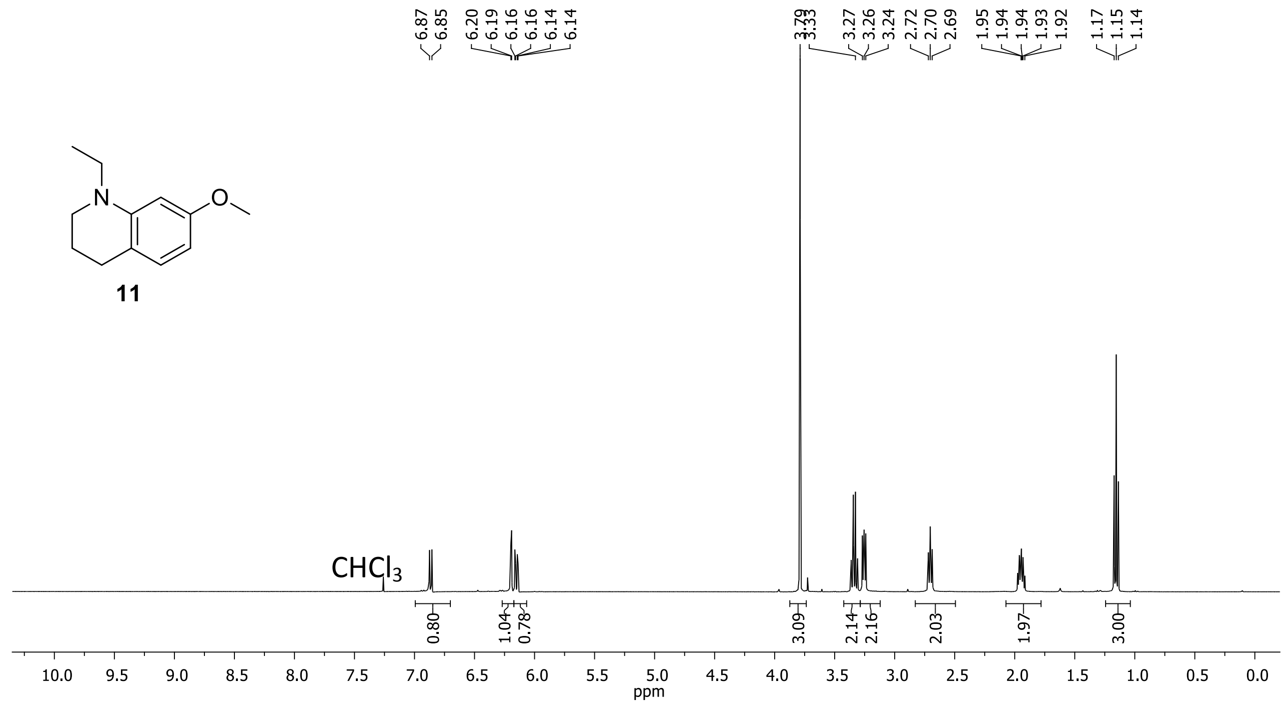
^{1}H NMR of compound **10** in $CDCl_3$



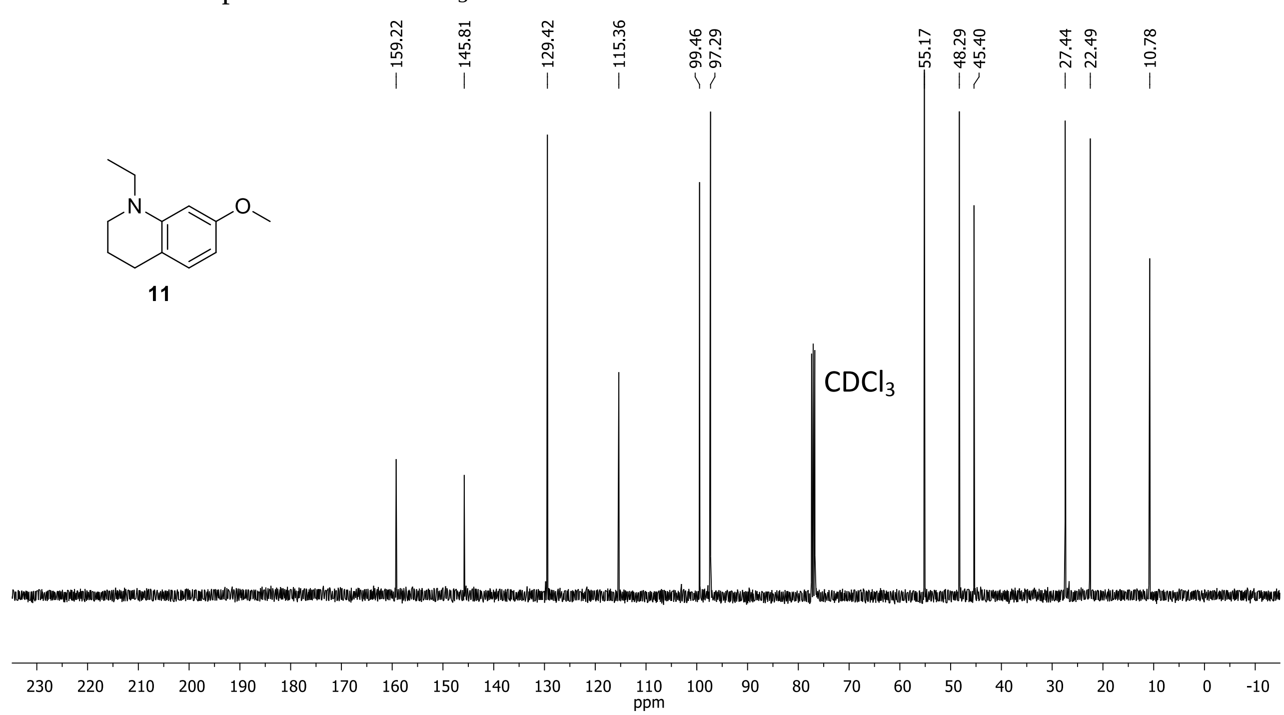
^{13}C NMR of compound **10** in $CDCl_3$



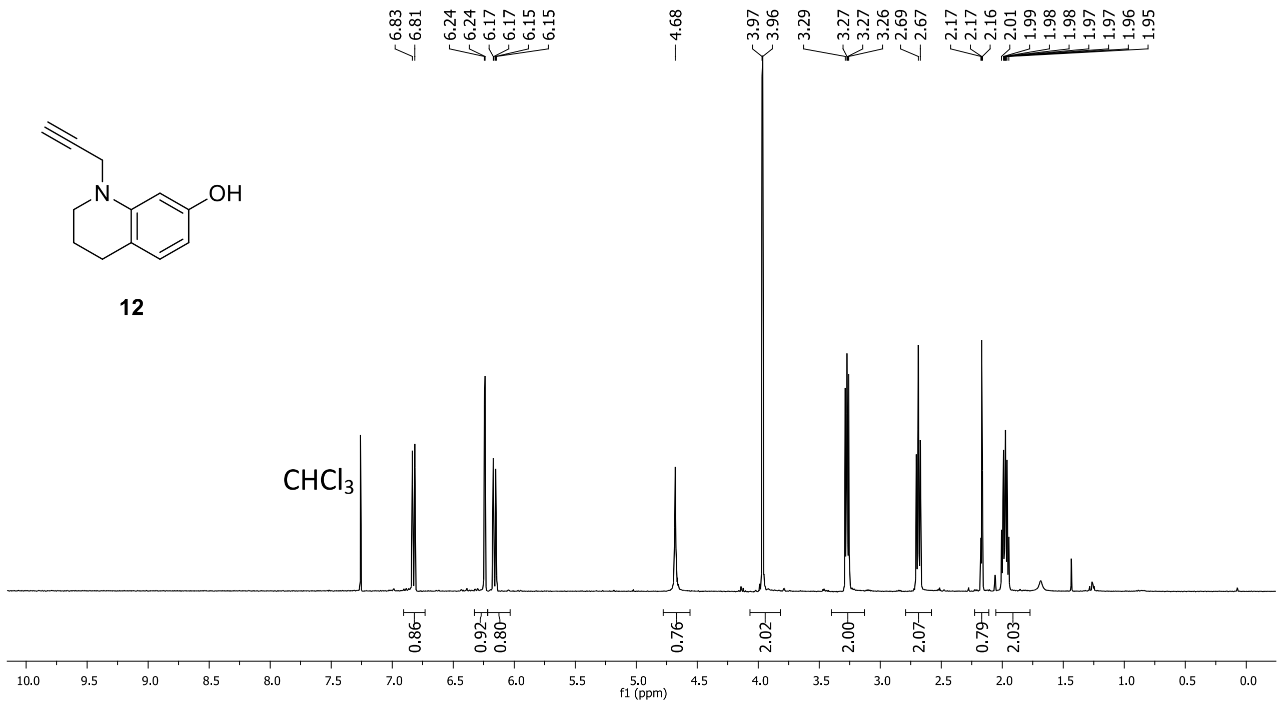
^{1}H NMR of compound **11** in $CDCl_3$



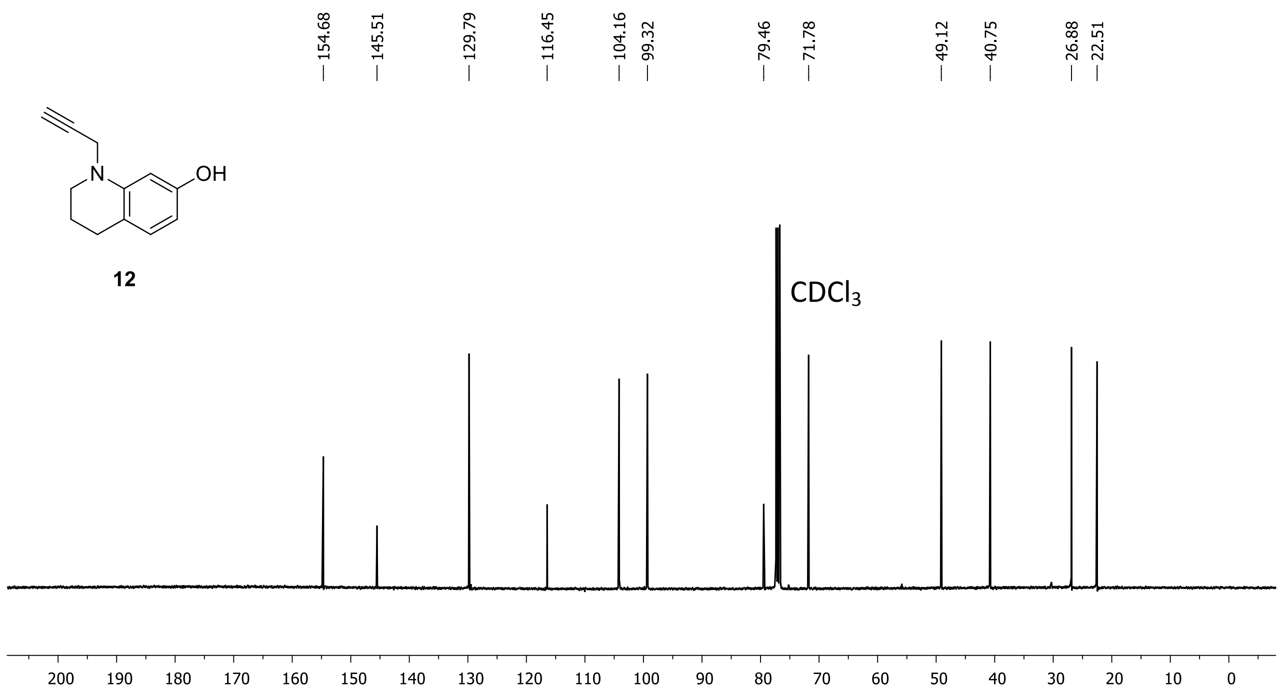
^{13}C NMR of compound **11** in $CDCl_3$



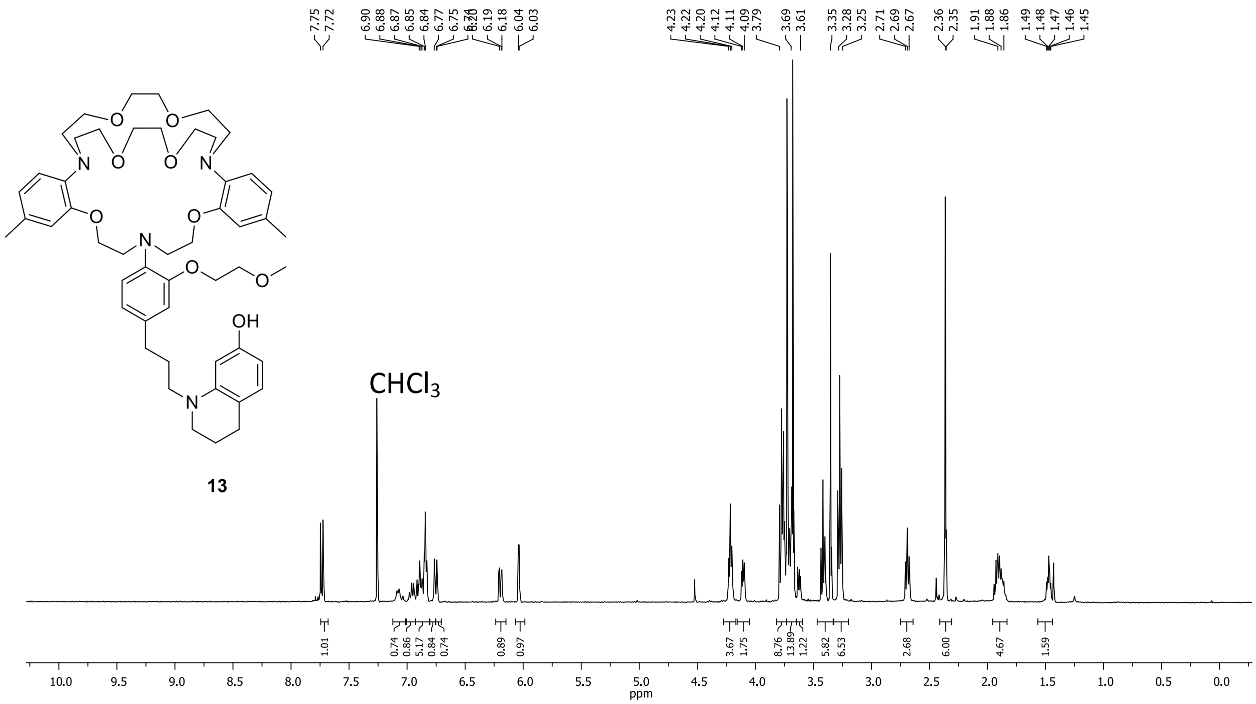
^{1}H NMR of compound **12** in $CDCl_3$



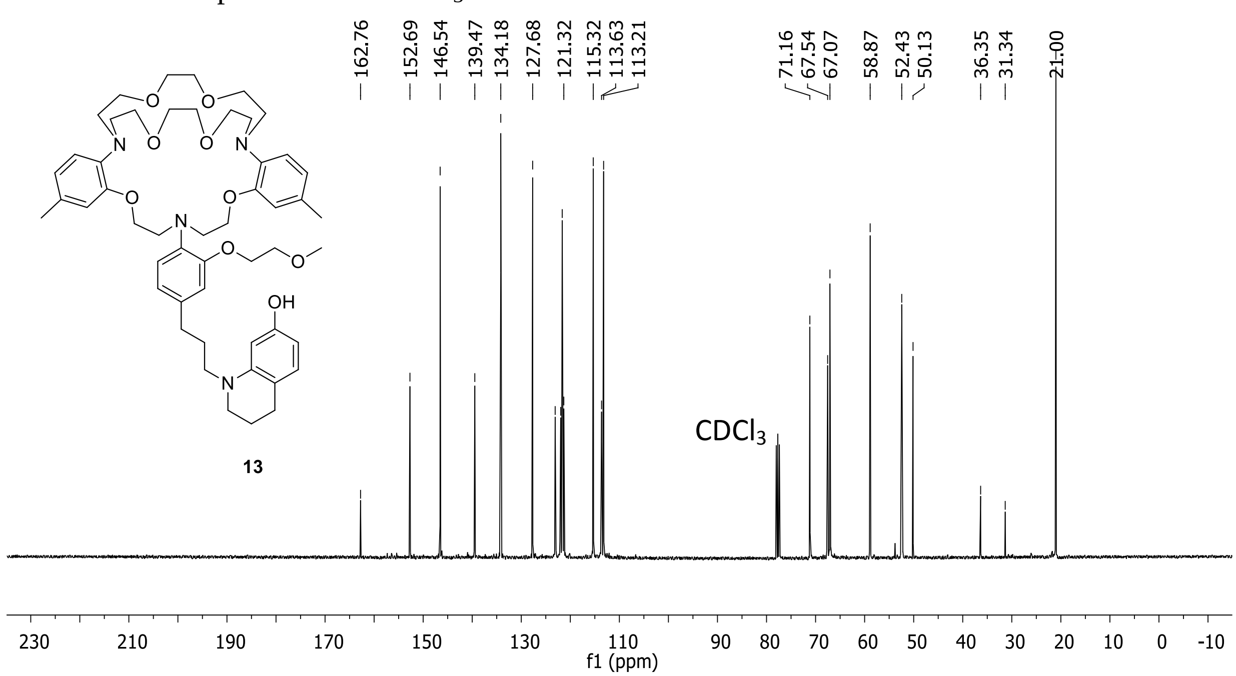
^{13}C NMR of compound **12** in $CDCl_3$



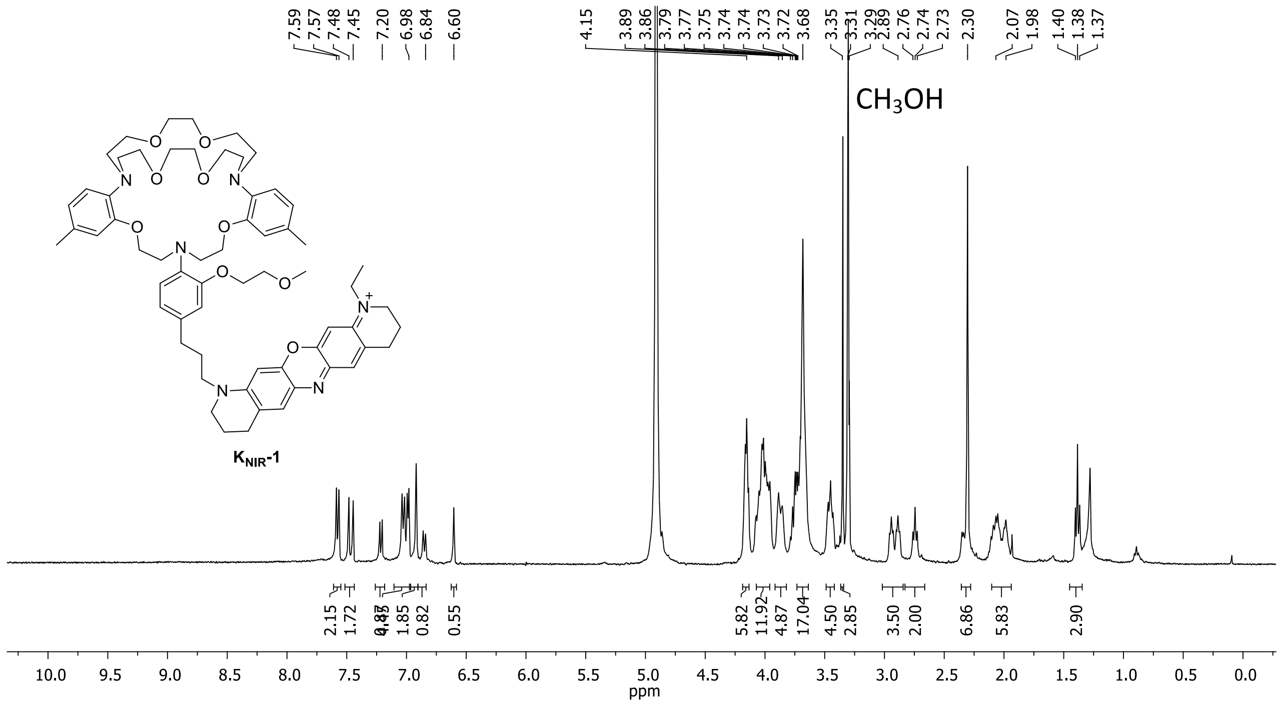
^{1}H NMR of compound **13** in $CDCl_3$



^{13}C NMR of compound **13** in $CDCl_3$



^{1}H NMR of **K$_{NIR}$-1acetate** in CD_3OD



^{13}C NMR of **K$_{NIR}$-1acetate** in DMSO-d6