Supplementary Materials

Fluorescent Vitamin B_{12}-Platinum(II) Derivatives as Potential Metallotheranostic Agents for the Treatment and Imaging of Tumors

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Materials and methods

Abbreviations

B_{12}: vitamin B_{12} (cyanocobalamin)

L1: (2*R*,5*S*)-2-methyl-5-(1-methylethenyl)-2-(phenylamino)-cyclohexanone oxime

L2: (2*S*,5*R*)-2-methyl-5-(1-methylethenyl)-2-(phenylamino)-cyclohexanone oxime

Pt1: dichloro[(2*R*,5*S*)-2-methyl-5-(1-methylethenyl)-2-(phenylamino-κ*N*)-cyclohexanone oxime-κ*N*(OH))]platinum(II)

Pt2: dichloro[(2*S*,5*R*)-2-methyl-5-(1-methylethenyl)-2-(phenylamino-κ*N*)-cyclohexanone oxime-κ*N*(OH))]platinum(II)

R6G: rhodamine 6G

R6G*: rhodamine 6G–*trans*-1,4-diaminocyclohexane

Materials

Cyanocobalamin (Fisher), rhodamine 6G, *trans*-1,4-diaminocyclohexane, 1,1'-carbonyldiimidazole (CDI), $Al(CH_3)_3$ 2.0 M toluene solution (Sigma-Aldrich), diisopropylamine (DIPEA) (Fluorochem), $AgNO_3$, $K_2[PtCl_4]$ (Alfa Aesar), trifluoroacetic acid (TFA) (Acros) and all deuterated solvents for NMR analysis (Deutero) were of reagent grade or comparable purity and were used as supplied. Anhydrous DCM was obtained by distillation over CaH_2 and subsequently stored over 4 Å activated molecular sieves under an inert atmosphere of nitrogen. All other reagents and solvents were used as purchased without any further purification.

Instrumentation

Thin layer chromatography (TLC) was performed on silica gel Merck $60F_{254}$ pre-coated aluminum sheets. Spots were visualized by direct UV irradiation at 254 nm or developed by exposure to either *p*-anisaldehyde or potassium permanganate staining solutions as appropriate.

Flash column chromatography was performed on Sigma Aldrich 60 Å silica gel (40-63 μm, 230-400 mesh) as stationary phase using the appropriate eluent.

Semi-preparative HPLC was performed using either a Waters Alliance 2695 chromatography system or a Varian Prostar 215 system equipped with a Waters UV Dual λ Absorbance detector 2487 at 210 and 254 nm. All solvents used were of HPLC grade.

Elemental analyses (carbon, hydrogen and nitrogen) were performed with a LECO CHNS-932 analyzer.

HRMS was carried out on an Agilent 1290 Infinity-QTOF 6540 UPLC/MS instrument equipped with a dual AJS ESI source. Samples were prepared in HPLC grade methanol (0.5 mg mL^{-1}) and mass

spectra were acquired in positive and negative ion modes over a mass range of *m/z* 100-1500. Data processing was carried out using MassHunter version B.07.00 (Agilent Technologies Inc.).

FT-IR spectra were recorded from CsI disks at room temperature on a Perkin Elmer Frontier FT-IR/FIR spectrophotometer in the range 4000-600 cm^{-1} (32 scans, resolution 4 cm^{-1}) and in the range 600-200 cm^{-1} (32 scans, resolution 2 cm^{-1}). Data processing was carried out using OMNIC version 5.1 (Nicolet Instrument Corporation).

All ^{1}H, $^{13}C\{^{1}H\}$ and $^{31}P\{^{1}H\}$ NMR spectra were acquired in the appropriate deuterated solvent at room temperature on a on either a Jeol ECX 400 MHz spectrometer or a Varian VNMRS 500 MHz spectrometer or an Agilent DD2 600 MHz spectrometer equipped with a cold probe. ^{1}H and ^{13}C signals were assigned with the aid of $[^{1}H,^{1}H]$ COSY, $^{13}C\{^{1}H\}$ DEPT, $[^{1}H,^{13}C]$ HMQC and $[^{1}H,^{13}C]$ HSQC experiments where appropriate. Additional $[^{1}H,^{13}C]$ HMBC and $[^{1}H,^{15}N]$ HMBC experiments were carried out in $CDCl_3$ on an Ultrashield Bruker Avance AV400 spectrometer. ^{1}H and ^{13}C chemical shifts were referenced to TMS at 0.00 ppm *via* internal referencing to the residual peak of the deuterated solvent employed. ^{31}P chemical shifts were referenced to an external standard of 85% H_3PO_4 at 0 ppm. ^{15}N chemical shifts were referenced to an external standard of liquid NH_3 at 0 ppm. Data processing was carried out using MestReNova version 12.0 (Mestrelab Research S.L.).

Pink crystals of **R6G*** suitable for X-ray crystallography were obtained upon slow evaporation of a CH_2Cl_2/MeOH solution of the compound. Single crystal X-ray diffraction data for **R6G*** were collected using an Oxford Diffraction Xcalibur CCD diffractometer (Mo-K_α radiation: λ = 0.71073 Å) at room temperature. Empirical absorption corrections were applied using the CrysAlis RED software [89], based on multi-scan symmetry-related measurements. The crystal structure was solved with the SHELXL structure solution program [90] and refined by full matrix least-squares using SHELXL refinement package [91], embedded in the OSCAIL software [92]. The compound crystallized as a chloride salt with two water molecules of crystallization. One of them showed disorder and was described in two positions: O(4)A and O(4)B with constrained occupancies of 75% and 25% respectively. Non-hydrogen atoms were treated anisotropically, except the oxygen atoms of the solvent molecules (O(3), O(4)A and O(4)B) which were refined isotropically. All the hydrogen atoms were positioned geometrically and refined by using a riding model, except those linked to nitrogen which were isotropically refined. Additionally, the hydrogen atoms of the water molecules were not included in the model. Moreover, a DFIX constraint was employed for the distance N(2)-H(2).

Colorless crystals of ***R*$_N$-Pt1** were grown at room temperature by slow evaporation from a chloroform solution. Crystals were removed from the vial and covered with a layer of a viscous perfluoropolyether (FomblinY). A suitable crystal was selected with the aid of a microscope,

mounted on a cryoloop, and immediately placed in the low temperature nitrogen stream of the diffractometer. The intensity data sets were collected at 200 K on a Bruker-Nonius KappaCCD diffractometer equipped with an Oxford Cryostream 700 unit. The structure was solved using the WINGX package [93] by intrinsic phasing methods (SHELXT) [90], and refined by least-squares against F2 (SHELXL-2018/3) [91]. All non-hydrogen atoms were anisotropically refined. All the hydrogen atoms were positioned geometrically and refined by using a riding model, except that linked to N(2) which was isotropically refined.

Crystallographic data of both **R6G*** and ***R*$_N$-Pt1** are reported on page S67. CCDC 2321413 (**R6G***) and CCDC 2321414 (***R*$_N$-Pt1**) contain the supplementary data for this paper.

UV-Vis and fluorescence measurements

UV-Vis spectra of **Pt1** and **Pt2** were acquired for 50 μM samples in water at room temperature on a Perkin-Elmer Lambda 35 Spectrophotometer in the range 200-840 nm.

Absorbance and emission spectra were recorded for 25 μM samples in DMSO at 25°C using an Agilent Cary Eclipse Fluorescence Spectrophotometer incorporating a xenon flash lamp and acquiring a data point every 12.5 ms at a scan rate of 24,000 nm min^{-1}. Wavelength range: 275-750 nm. Polarization measured as a function of temperature using the thermal software application Agilent WinFLR. Angle selections include 0°, 90°, 55° (magic angle), and 35°. Probe range: -10°C to +100°C. Probe size: 1.5 mm diameter, 15 mm length. Photometric performance: fiber optics. Temperature control: 20-60°C, using a water thermostatic cell holder. Reproducible temperature control: ±0.05°C. Absorption and emission spectra are reported on page S64.

Measurement of lipophilicity (Log*D*$_{7.4}$)

The *n*-octanol/water partition coefficients at pH 7.4 (Log*D*$_{7.4}$) were determined using the shake-flask method [78]. 100 mL of phosphate buffered saline (PBS) solution (pH 7.4, 0.01 M phosphate buffer, 0.0027 M KCl, 0.137 M NaCl) and 100 mL of analytical grade *n*-octanol were mutually saturated and mechanically shaken for 72 h using a Stuart STR4 Rotator Drive unit. A calculated amount of a freshly prepared 5 mM DMSO solution of each individual vitamin B$_{12}$-platinum(II) complex was diluted with the appropriate volume of an *n*-octanol/PBS-saturated solution to obtain a final stock 25 μM solution of the metal complex (the final DMSO content in solution being 0.5% v/v). 5 mL of this stock solution was then added to 5 mL of a PBS/*n*-octanol-saturated solution and the resulting mixture was mechanically shaken for 1 h. The biphasic mixture was centrifuged at 3000 rpm for 5 min to allow full separation of the two phases. Eventually, the concentration of each metal derivative in the various phases was measured by UV-Vis spectrophotometry. Measurements were carried out in triplicate for each sample.

For all complexes **B_{12}-Pt1**, **B_{12}-Pt2**, **R6G*-B_{12}-Pt1** and **R6G*-B_{12}-Pt2** absorbance was measured at 361 nm in PBS before and after partitioning. Log$D_{7.4}$ was calculated using the following equation:

$$\mathrm{Log}D_{7.4} = \mathrm{Log}\left(\frac{\mathrm{C_{oct}}}{\mathrm{C_{PBS}}}\right) \cong \mathrm{Log}\left(\frac{\mathrm{A_{oct}}}{\mathrm{A_{PBS}}}\right) = \mathrm{Log}\left(\frac{\mathrm{A_0 - A_{PBS}}}{\mathrm{A_{PBS}}}\right)$$

where:

- C_{oct} and C_{PBS} are the molar concentrations of each individual metal complex in the octanolic and PBS fractions, respectively, after partitioning;
- A_{oct} and A_{PBS} are the absorbances of each individual metal complex in the octanolic and PBS fractions, respectively, after partitioning;
- A_0 is the absorbance of each individual metal complex in the 25 μM *n*-octanol/PBS-saturated stock solution.

***In vitro* biological studies**

Cell lines, culture conditions and samples preparation. The androgen-unresponsive prostate cancer PC-3, cervical carcinoma HeLa and breast adenocarcinoma MCF-7 human cell lines were obtained from the American Type Culture Collection (Manassas, VA). HeLa and MCF-7 cells were grown in high-glucose Dulbecco's Modified Eagle´s Medium (DMEM), whereas PC-3 cells were grown in Roswell Park Memorial Institute (RPMI-1640) medium, and the culture medium was changed every 3 days.

After reaching 70-80% confluence, cells were washed with PBS, detached with 0.25% trypsin/0.2% ethylendiaminetetraacetic acid (EDTA) and seeded at 30,000-40,000 cells·cm^{-2}. Cells were cultured at 37°C in 5% CO_2 and moisture-enriched atmosphere in the respective medium supplemented with 5% heat-inactivated fetal bovine serum (FBS), 200 U mL^{-1} penicillin, 100 μg mL^{-1} streptomycin, and 2.0 mM L-glutamine. Adherent cells were allowed to attach for 24 h prior to treatment with the various tested compounds.

Stock solutions in the 500-1000 μM concentration range of the starting platinum(II) complexes (**Pt1** and **Pt2**) and their fluorescent vitamin B_{12} conjugate counterparts (**R6G*-B_{12}-Pt1** and **R6G*-B_{12}-Pt2**) were freshly prepared in 10% DMSO/cell culture medium and subsequently sequentially diluted with the appropriate complete cell culture medium to the desired concentration in such a way that the final amount of organic solvent did not exceed 0.2% v/v. Stock solutions in the 500-1000 μM concentration range of cisplatin and the non-fluorescent vitamin B_{12}-platinum(II) complexes (**B_{12}-Pt1** and **B_{12}-Pt2**) were freshly prepared directly in the appropriate complete cell culture medium and sequentially diluted with the same medium to the desired concentration.

Evaluation of the antiproliferative activity in vitro. Cells (5×10^4 per well) were seeded in 96-well cell culture plates (100 μL per well) in the appropriate complete medium and incubated at 37°C in

5% CO_2 humidified atmosphere. After allowing to adhere for 24 h, cells were treated with the test compounds at different concentrations and incubated for further 72 h at 37°C. The medium was then discarded, cells were washed with fresh PBS solution and harvested. All experiments were carried out in quadruplicate and control groups with and without 0.2% DMSO were included.

A modified MTT-reduction assay was performed to determine cell viability. 10 µL of a 5 $mg \cdot mL^{-1}$ solution of [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] (MTT) in PBS were added to each well and cells were incubated for further 1-3 h at 37 °C. The formazan crystals formed were then dissolved by adding to each well 100 µL of DMSO. The inhibition of cell growth induced by the tested compounds was assessed by measuring the absorbance of each well at 550 nm using a 96-well multi-scanner auto-reader FluOstar Omega (BMG Labtech). Antiproliferative data are expressed as IC_{50} values, that is, the concentration (µM) of the test agent inhibiting cell growth by 50% compared with control cultures. Results were calculated as the mean values ± standard deviation (SD) of at least three independent experiments.

Cell cycle distribution analysis. PC-3 cells (5×10^5) were seeded in 6-well plates in RPMI-1640 medium containing 5% FBS and 1% antibiotic/antimycotic (penicillin/streptomycin/amphotericin B) and incubated at 37°C in 5% CO_2 humidified atmosphere. After 24 h, cells were treated with the test compounds at the indicated concentrations and incubated for further 72 h at 37°C. Cells were then washed with PBS, detached with 0.25% trypsin/0.2% EDTA, harvested and centrifuged at 500G for 5 min at 4°C. Pellets were mixed with ice-cold 70% w/v ethanol and kept at 4°C for 30 min. After removing the ethanol by centrifugation, the pellets were washed with PBS and centrifuged once again. The supernatant was discarded and the pellets were suspended in PBS solution containing 0.2 mg mL^{-1} RNAse A and 20 µg mL^{-1} propidium iodide (PI). The analysis of DNA content was performed on a MACSQuant Analyzer 10 flow cytometer (Miltenyi Biotech) at λ_{ex} = 488 nm/λ_{em} = 620 nm. Data processing and analysis was carried out using MacsQuantify version 2.13.1.

Cell internalization studies. PC-3 cells (5×10^4) were seeded in ibidi (60 µ dish, 35 mm high glass) in RPMI-1640 medium and incubated at 37°C in 5% CO_2 humidified atmosphere. After 72 h, cells were treated with the test compounds at the concentration of 10 µM (for 24 or 72 h, 37°C) or 200 µM (for 3 h at 37°C or 4°C). After the indicated exposure times, the culture medium was removed, cells were washed with sterile PBS, and fresh medium containing 2 µL of Hoechst (Invitrogen H5370) per dish was added. Visualization of compounds was carried out with a confocal microscope Leica SP5 (Hoechst: λ_{ex} = 405 nm/λ_{em} = 420-501 nm; samples: λ_{ex} = 561 nm/λ_{em} = 570-620 nm). The acquired images were obtained using individual filters. A combined image, overlaying the fluorescence acquired with the two filters, was processed and analyzed using the Leica AF microscope software. The confocal microscopy analysis was performed with ICTS NANBIOSIS (Singular Scientific and

Technological Infrastructures for the Production and Characterization of Nanomaterials, Biomaterials and Nanomedicine), specifically at the Confocal Microscopy Service: Ciber in Bioengineering Biomaterials and nanomedicine (CIBER-BNN) at the University of Alcalá (CAI Medicine and Biology).

Statistical analysis. Results were subjected to computer-assisted statistical analysis using One-Way Analysis of Variance ANOVA, Bonferroni's post-test and Student´s *t*-test to determine statistical significance. Data are shown as the means of individual experiments ± SD (Standard Deviation). Differences of $P < 0.05$ were considered statistically significant.

DNA interaction studies

Equilibrium dialysis. Calf thymus double-stranded DNA (CT dsDNA, deoxyribonucleic acid, activated, type XV) and the aqueous solutions of surfactant sodium dodecyl sulfate (SDS, 10%) were purchased from Sigma Aldrich and used as provided. The buffering system was: 10 mM phosphate buffer NaH_2PO_4/Na_2HPO_4, pH = 7.2. The solutions of DNA were prepared in the working phosphate buffer at 75 μM monomeric unit (mu) concentrations in base pairs (bp). After 24 h of hydration in milliQ water dialysis bags were filled with 75 μM (bp) of DNA (200 μL each bag) and placed in a beaker containing 225 mL of *ca.* 2 μM solution of the tested compound. The beaker was sealed with parafilm, kept in the dark covered with aluminum foil and the solution allowed to equilibrate under continuous stirring over 24 h at room temperature. Experiments were run in replicate, each using three dialysis compartments. The content of each dialysis bag was then transferred to an Eppendorf tube and mixed with a 10% aqueous solution of SDS to a final 1% (v/v) concentration. The solutions were subsequently allowed to equilibrate for additional 2 h, after which the total concentration of the ligand (C_t) was determined by UV-Vis analysis using the extinction coefficient determined for conjugates **B_{12}-Pt1**, **B_{12}-Pt2, R6G*-B_{12}-Pt1** and **R6G*-B_{12}-Pt2** in the presence of 1% SDS. The concentration of free compound (C_f) was also determined spectrophotometrically using an aliquot of the dialysate. The DNA-bound compound concentrations (C_b) were then calculated by difference ($C_b = C_t - C_f$) and the corresponding apparent association constants (K_{app}) were determined.

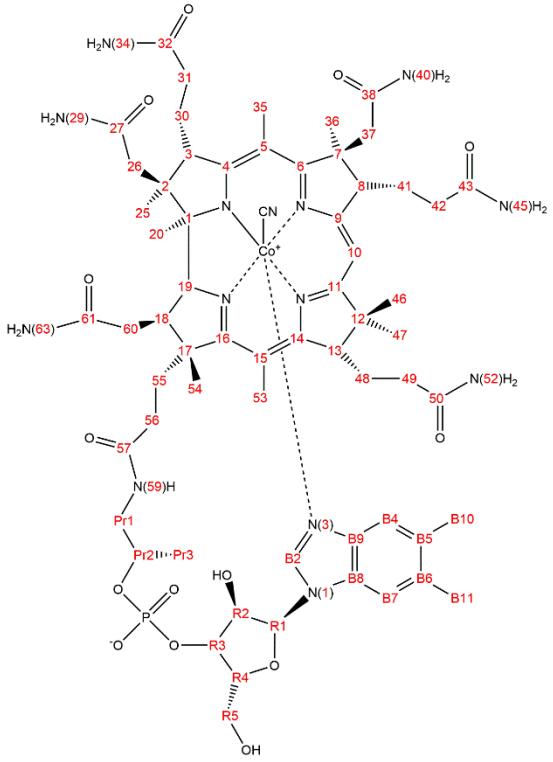
DNA FRET melting assay. The DNA melting assay was conducted on a QuantStudio 6 PRO 0.2 mL, 96-well package instrument (Applied Biosystems) using 96-Well Optical MicroAmp Reaction Plates (Life Technologies Corporation). The oligonucleotide sequence employed in this experiment, F10T (5'-FAM-AGC TAT TA /sp18/ TA GCT ATA-TAMRA-3'; FAM: 6-carboxyfluorescein; TAMRA: carboxytetramethylrhodamine) was produced, HPLC-purified and desalted by IDT™. The buffering system was: 10 mM sodium cacodylate, 100 mM LiCl, pH = 7.3). For the experiments performed in the absence of chloride anions the buffering system was: 10 mM Na_2HPO_4/NaH_2PO_4, pH = 7.2. The duplex-forming oligonucleotide was first dissolved in BPC grade water

(Biotechnology Performance Certified) to prepare a 50 μM stock solution, which was then diluted to 0.5 μM and subsequently mixed with the working buffer (2×) and water (BPC grade). The DNA solution was heated at 90°C for 10 min, slowly cooled down for 3 h and left at 4°C overnight. Compounds to be tested were dissolved in water and approximately 1 mM stock solutions were prepared. The exact concentrations were determined by UV-Vis analysis. The stock solutions were then diluted with buffer to a final 50 μM concentration of each compound. DNA solutions were mixed in a 96-well microplate with solutions of the tested compounds and buffer to achieve a total volume of 50 μL with a F10T concentration of 0.2 μM and a compound concentration ranging between 1.0 and 10 μM.

The melting process program consisted of an incubation step at 24°C for 5 min and then a temperature ramp (1°C min^{-1} rate) with fluorescence readings at each degree up to 95°C. Subsequently, the microplate was incubated for 5 min at 96°C and then gradually cooled down at -1°C min^{-1} rate, with fluorescence readings at each degree down to 25°C. The melting and refolding (or renaturing) curves were registered based on the change in emission of the FAM fluorophore (λ_{ex} = 492 nm/λ_{em} = 516 nm). Melting temperature values (T_m) were determined from replicate experiments using normalized curves and considering mid-transition ($T_{½}$) temperatures.

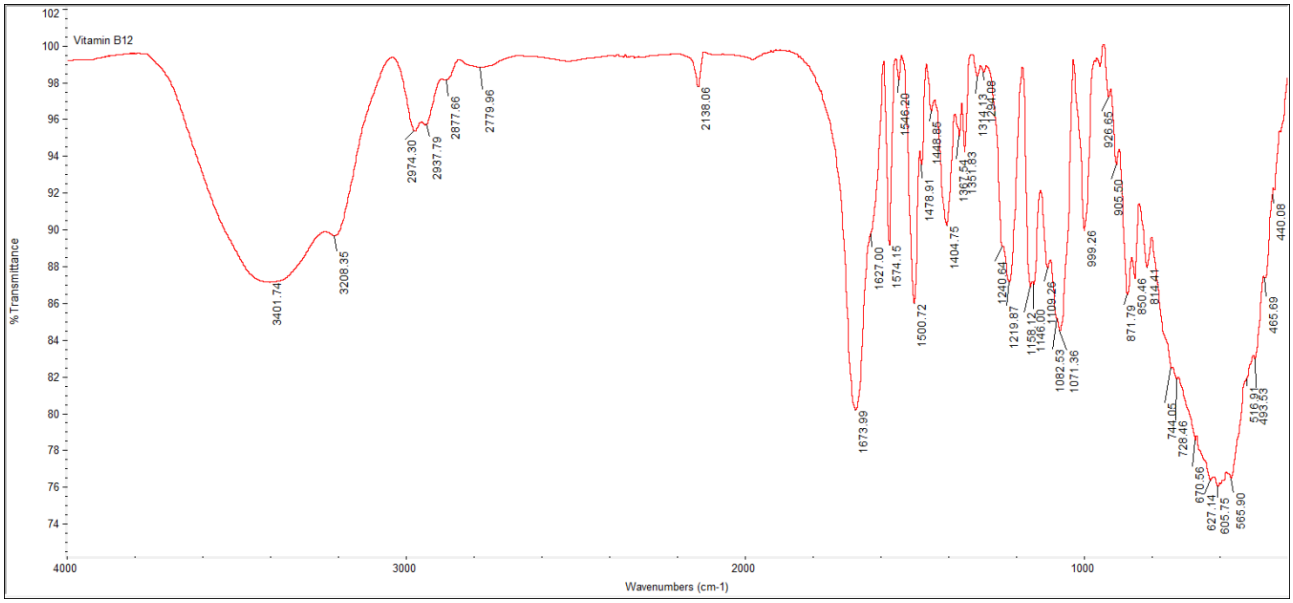
Spectroscopic characterization of the starting reagents

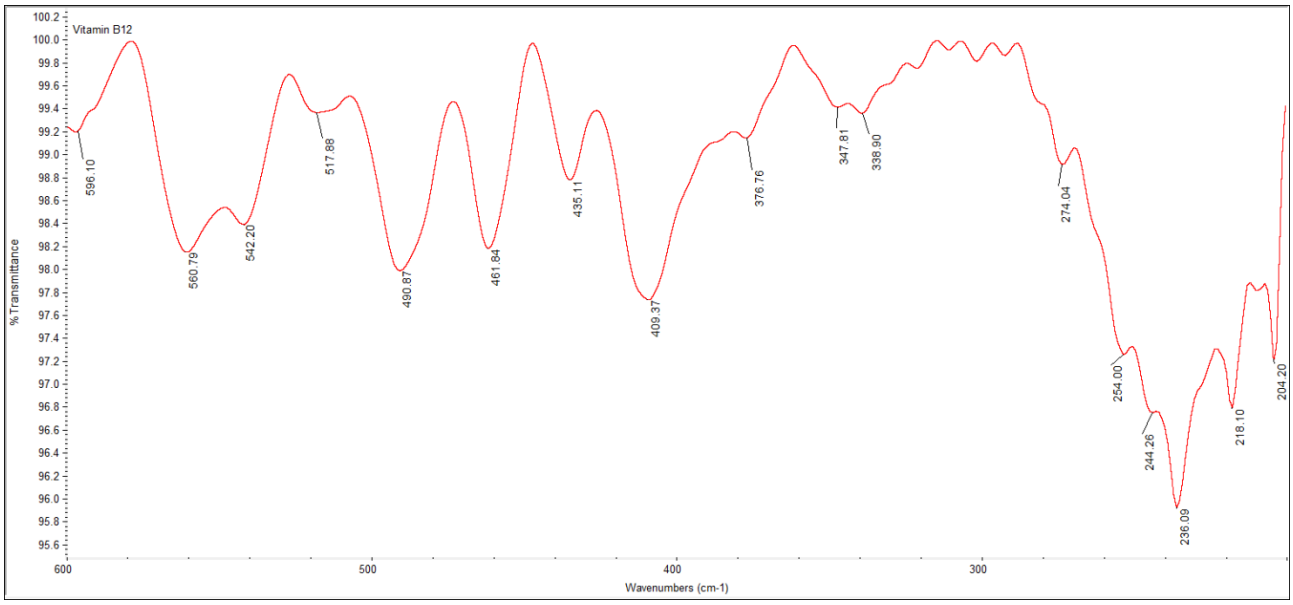
Vitamin B_{12} (cyanocobalamin, B_{12})

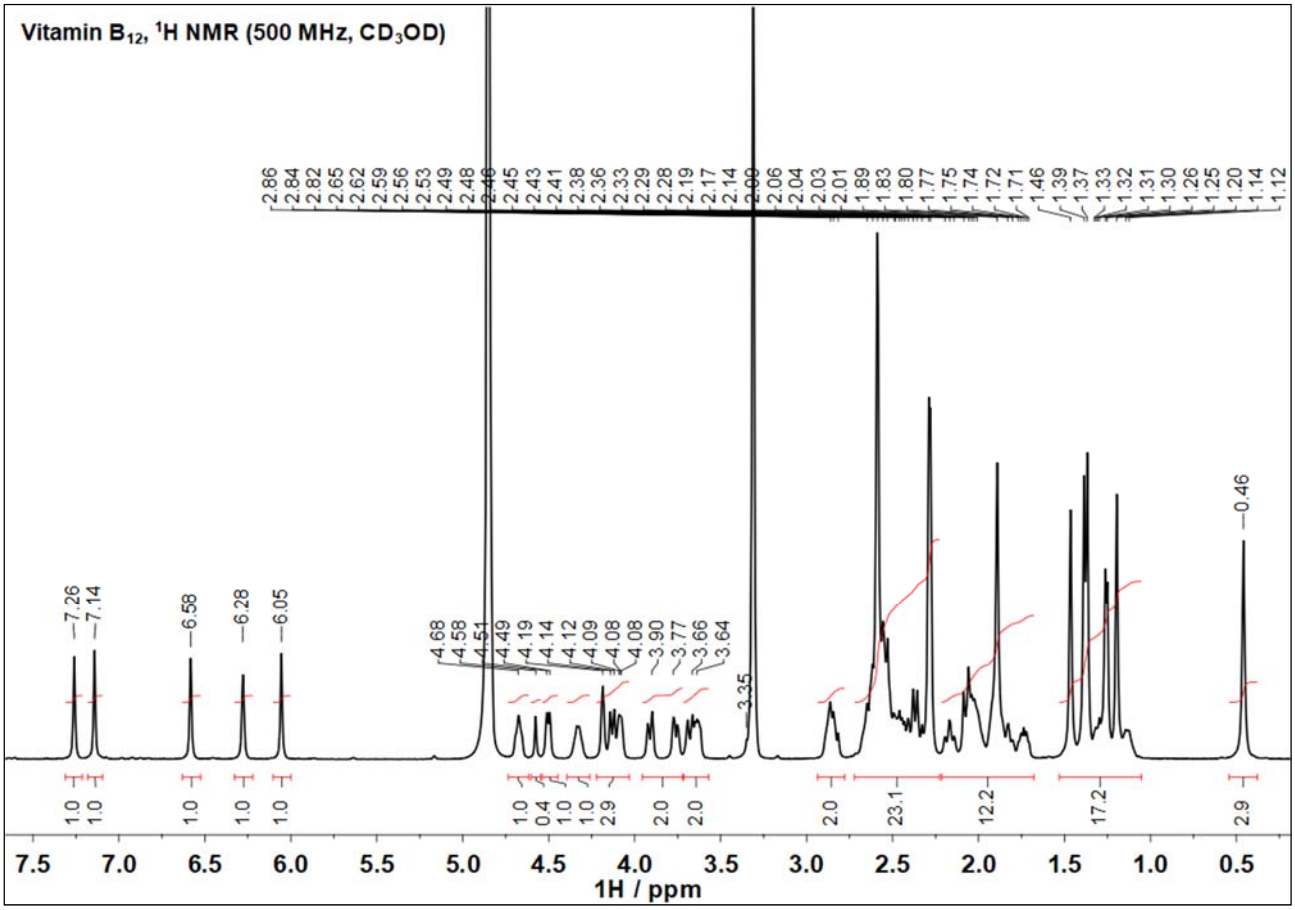


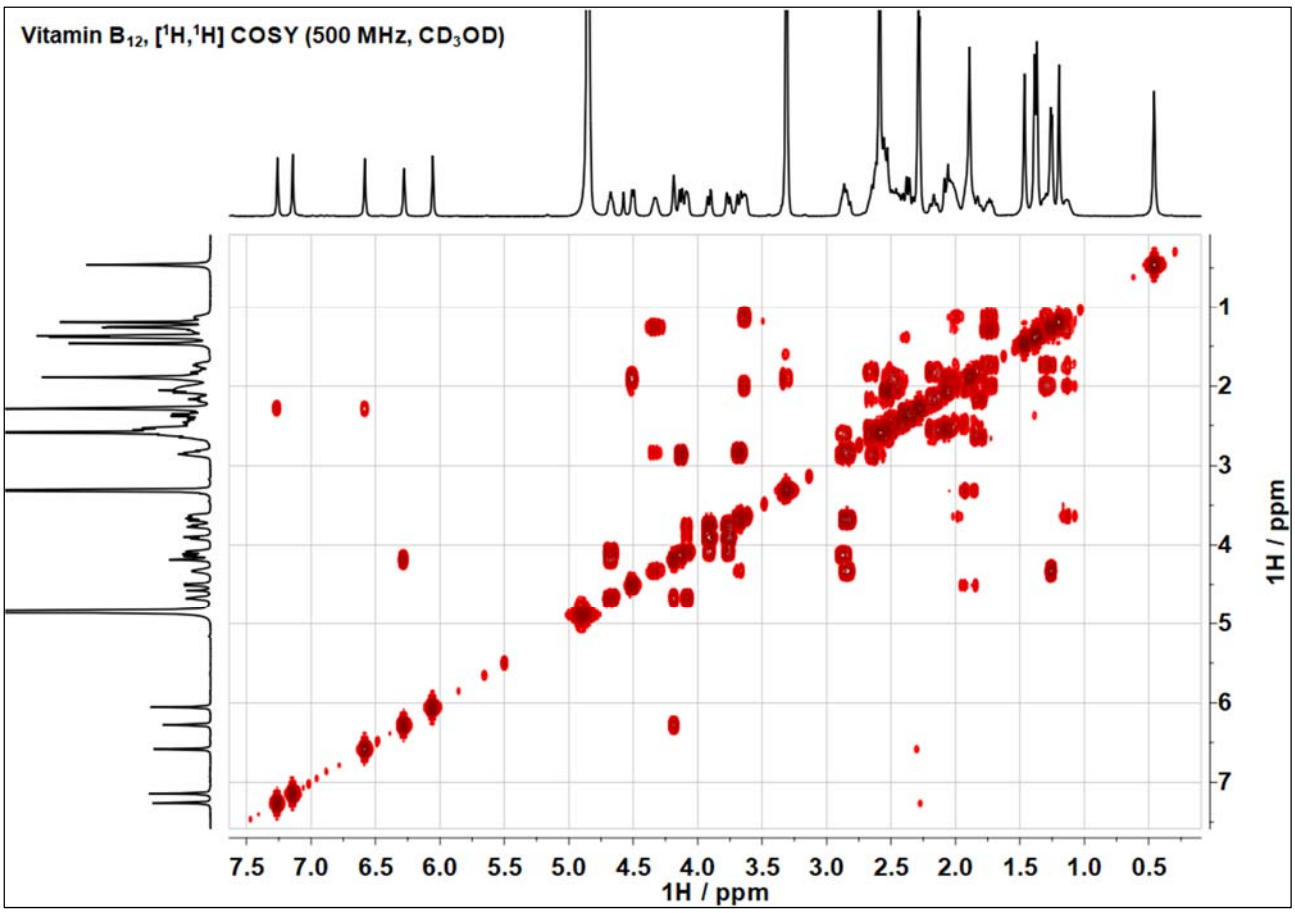
Commercially available (98+%). FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3402/3208 ($\nu_{a,s}(NH_2)_{amide}$ + $\nu(NH)_{amide}$ + $\nu(OH)_{ribose}$ overlapped), 2138 ($\nu(C{\equiv}N)$), 1674 ($\nu(C{=}O)$/amide I), 1627 ($\delta(NH_2)$/amide II), 1574/1546 (corrin ring breathing mode), 1500/1479 ($\nu(C{=}N)$ + $\nu(C{=}C)$ overlapped), 1158/1146 ($\nu_{oop}(PO_2^-)$), 1083/1071 ($\nu_{ip}(PO_2^-)$), 999 ($\nu(P{-}O{-}C)$), 561 ($\delta(Co{-}C{\equiv}N)$), 491 ($\nu(Co{-}N)$), 409 ($\nu(Co{-}CN)$), 348/339 ($\nu(Co{-}N)$) cm^{-1}[94]. ^{1}H NMR (500 MHz; CD_3OD; 298 K): δ 0.46 (3 H, s, $C^{20}\boldsymbol{H}_3$), 1.09-1.17 (1 H, m, $C^{41}\boldsymbol{H}'$), 1.20 (3 H, s, $C^{46}\boldsymbol{H}_3$), 1.26 (3 H, d, $C^{Pr3}\boldsymbol{H}_3$, $^3J_{H,H}$ = 5.6 Hz), 1.28-1.34 (1 H, m, $C^{42}\boldsymbol{H}'$), 1.37 (3 H, s, $C^{25}\boldsymbol{H}_3$), 1.39 (3 H, s, $C^{54}\boldsymbol{H}_3$), 1.46 (3 H, s, $C^{47}\boldsymbol{H}_3$), 1.71-1.77 (1 H, m, $C^{42}\boldsymbol{H}''$), 1.80-1.86 (1 H, m, $C^{55}\boldsymbol{H}'$), 1.87-1.95 (2 H, m, $C^{48}\boldsymbol{H}_{2/diastereotopic}$), 1.89 (3 H, s, $C^{36}\boldsymbol{H}_3$), 1.95-2.04 (3 H, m, $C^{41}\boldsymbol{H}''$ + $C^{30}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 2.06-2.09 (1 H, m, $C^{26}\boldsymbol{H}'$), 2.14-2.19 (1 H, m, $C^{56}\boldsymbol{H}'$), 2.28 (3 H, s, $C^{B11}\boldsymbol{H}_3$), 2.29 (3 H, s, $C^{B10}\boldsymbol{H}_3$), 2.33-2.41 (2 H, m, $C^{37}\boldsymbol{H}_{2/diastereotopic}$), 2.43-2.49 (4 H, m, $C^{31}\boldsymbol{H}_{2/diastereotopic}$ + $C^{49}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 2.53-2.56 (1 H, m, $C^{26}\boldsymbol{H}''$), 2.57-2.67 (4 H, m, $C^{55}\boldsymbol{H}''$ + $C^{56}\boldsymbol{H}''$ + $C^{60}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 2.59 (6 H, br s, $C^{35}\boldsymbol{H}_3$ + $C^{53}\boldsymbol{H}_3$ overlapped), 2.82-2.90 (2 H, m, $C^{Pr1}\boldsymbol{H}'$ + $C^{18}\boldsymbol{H}$ overlapped), 3.30-3.35 (1 H, m, $C^{13}\boldsymbol{H}$), 3.62-3.66 (1 H, m, $C^{8}\boldsymbol{H}$), 3.66-3.69 (1 H, m, $C^{Pr1}\boldsymbol{H}''$), 3.75-3.92 (2 H, m, $C^{R5}\boldsymbol{H}_{2/diastereotopic}$), 4.08-4.09 (1 H, m, $C^{R4}\boldsymbol{H}$), 4.13 (1 H, d, $C^{19}\boldsymbol{H}$, $^3J_{H,H}$ = 11.3 Hz), 4.19 (1 H, br s, $C^{R2}\boldsymbol{H}$), 4.31-4.35 (1 H, m, $C^{Pr2}\boldsymbol{H}$), 4.49-4.51 (1 H, m, $C^{3}\boldsymbol{H}$), 4.65-4.69 (1 H, m, $C^{R3}\boldsymbol{H}$), 6.05 (1 H, s, $C^{10}\boldsymbol{H}$), 6.28 (1 H, br s, $C^{R1}\boldsymbol{H}$), 6.58 (1 H, s, $C^{B4}\boldsymbol{H}$), 7.14 (1 H, s, $C^{B2}\boldsymbol{H}$), 7.26 (1 H, s, $C^{B7}\boldsymbol{H}$) ppm [95–99]. $^{13}C\{^1H\}$ NMR (125 MHz; CD_3OD; 298 K): δ

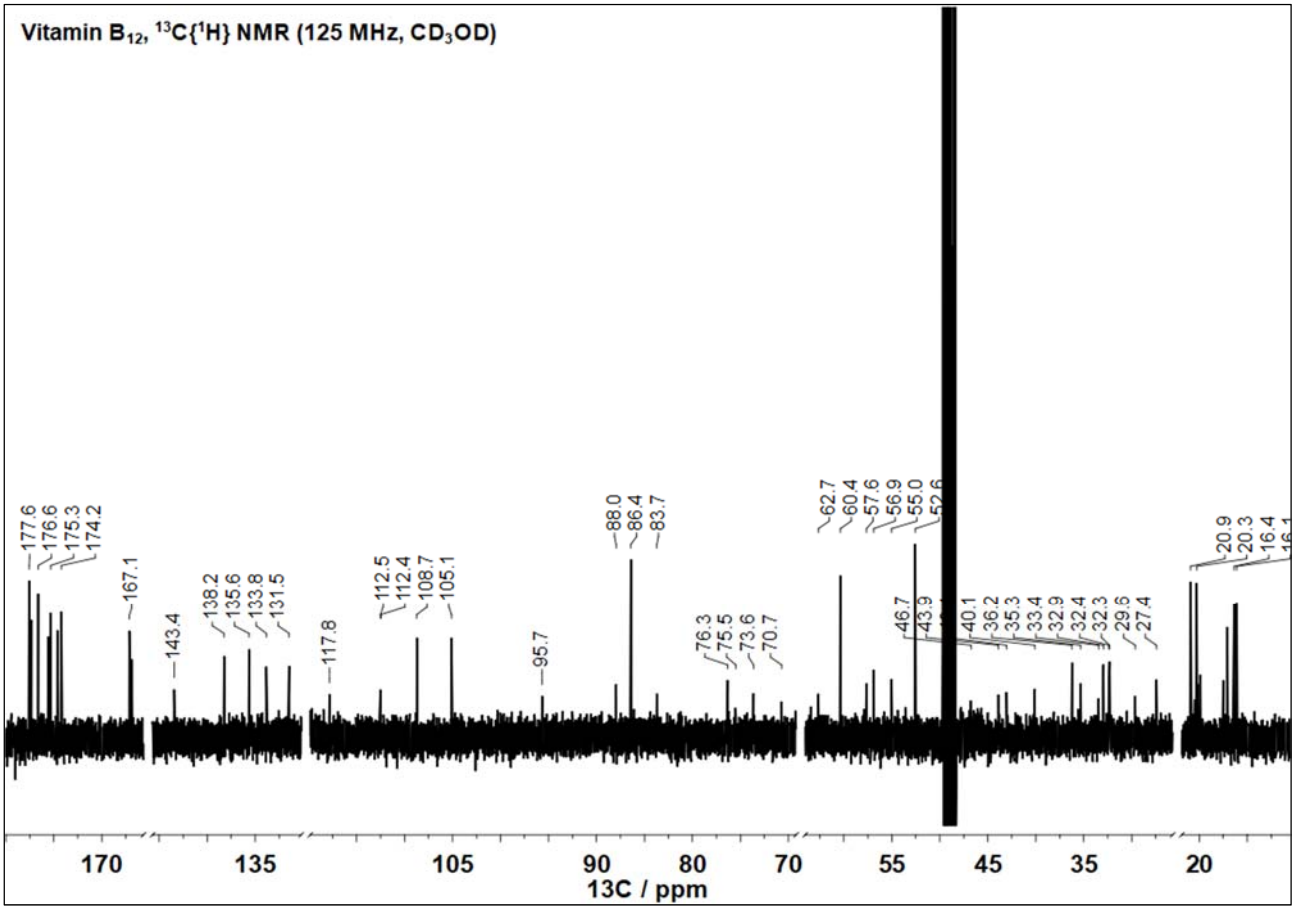
16.1 (***C***^{53}H$_3$), 16.4 (***C***^{35}H$_3$), 17.1 (***C***^{25}H$_3$), 17.5 (***C***^{54}H$_3$), 19.9 (***C***^{36}H$_3$), 20.1 (***C***^{Pr3}H$_3$), 20.2 (***C***^{47}H$_3$), 20.3 (***C***^{B11}H$_3$), 20.4 (***C***^{20}H$_3$), 20.9 (***C***^{B10}H$_3$), 27.4 (***C***^{30}H$_2$ + ***C***^{41}H$_2$ overlapped), 29.6 (***C***^{48}H$_2$), 32.3 (***C***^{46}H$_3$), 32.4 (***C***^{42}H$_2$), 32.9 (***C***^{56}H$_2$ + ***C***^{60}H$_2$ overlapped), 33.4 (***C***^{55}H$_2$), 35.3 (***C***^{49}H$_2$), 36.2 (***C***^{31}H$_2$), 40.1 (***C***^{18}H), 43.1 (***C***^{37}H$_2$), 43.9 (***C***^{26}H$_2$), 46.7 (***C***^{Pr1}H$_2$), ~49 (***C***^{2} + ***C***^{12} overlapped with the residual peak of CD$_3$OD), 52.6 (***C***^{7}), 55.0 (***C***^{13}H), 56.9 (***C***^{8}H), 57.6 (***C***^{3}H), 60.4 (***C***^{17}), 62.7 (***C***^{R5}H$_2$), 70.7 (***C***^{R2}H), 73.6 (***C***^{Pr2}H), 75.5 (***C***^{R3}H), 76.3 (***C***^{19}H), 83.7 (***C***^{R4}H), 86.4 (***C***^{1}), 88.0 (***C***^{R1}H), 95.7 (***C***^{10}H), 105.1 (***C***^{15}), 108.7 (***C***^{5}), 112.4 (***C***N), 112.5 (***C***^{B7}H), 117.8 (***C***^{B4}H), 131.5 (***C***^{B8}), 133.8 (***C***^{B5}), 135.6 (***C***^{B6}), 138.2 (***C***^{B9}), 143.4 (***C***^{B2}H), 166.9 (***C***^{14}), 167.1 (***C***^{6}), 174.2 (***C***^{57}), 174.6 (***C***^{38}), 175.3 (***C***^{61}), 175.5 (***C***^{27}), 175.6 (***C***^{43}), 176.6 (***C***^{32}), 177.4 (***C***^{9}), 177.6 (***C***^{50}), 177.7 (***C***^{11}), 180.1 (***C***^{16}), 181.6 (***C***^{4}) ppm [95–99]. ^{31}P{^{1}H} NMR (162 MHz; CD$_3$OD; 298 K): δ 0.3 (***P***O$_4^-$) ppm.

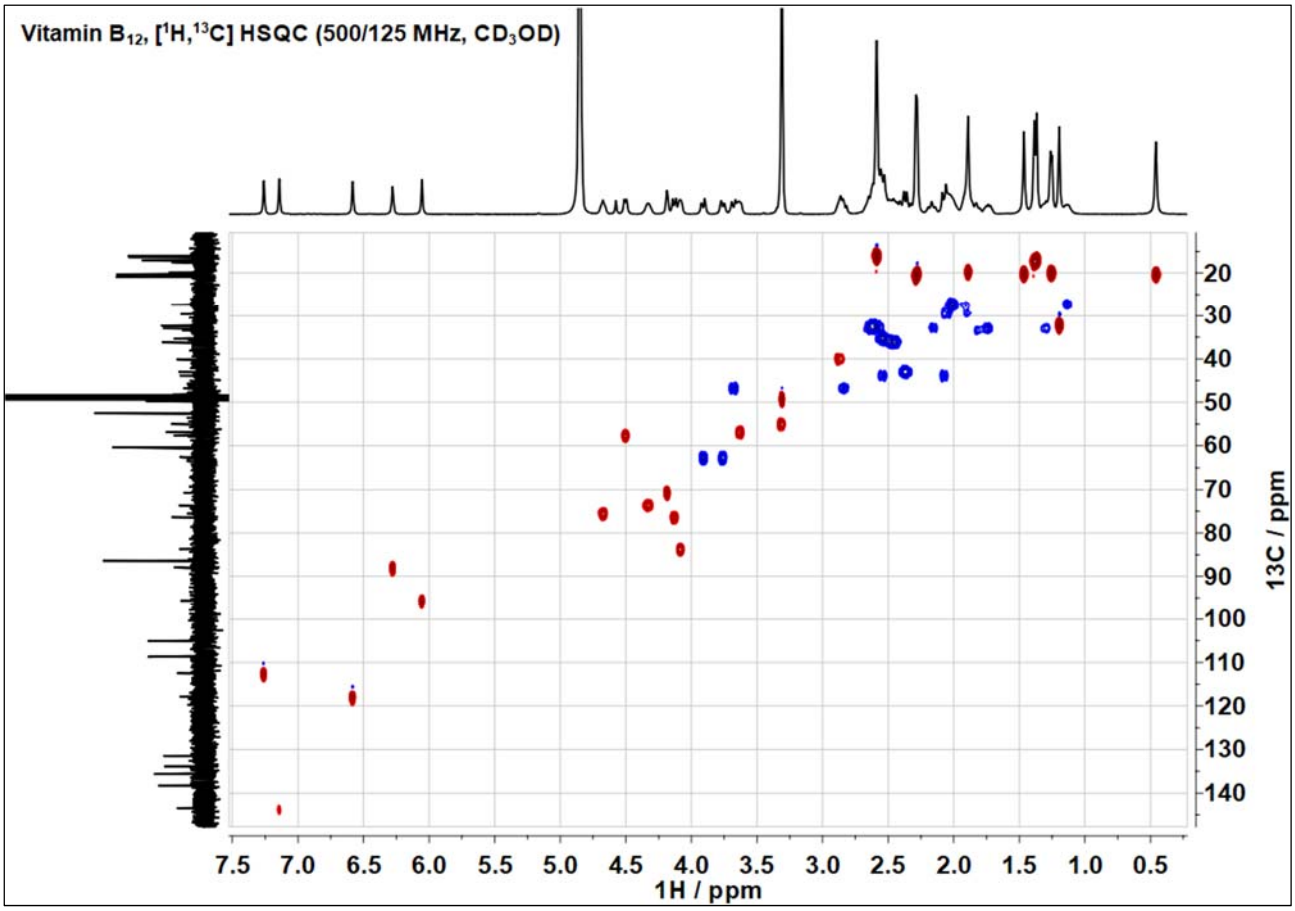


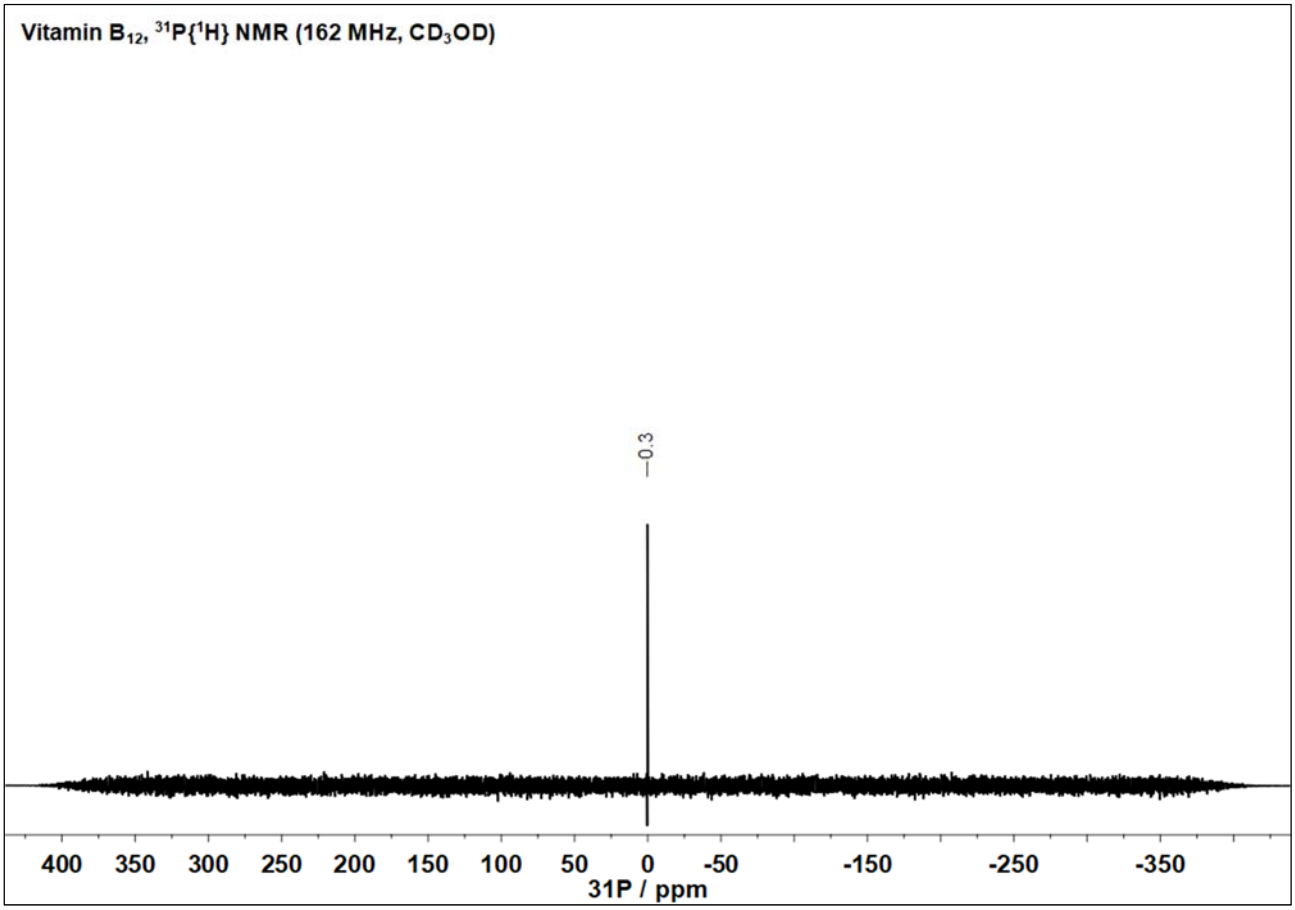



Vitamin B12, 1H NMR (500 MHz, CD3OD)
7.26
7.14
6.58
6.28
6.05
4.68
4.58
4.51
4.49
4.19
4.14
4.12
4.09
4.08
3.90
3.77
3.66
3.64
3.35
0.46
1.0
1.0
1.0
1.0
1.0
1.0
0.4
1.0
1.0
2.9
2.0
2.0
2.0
23.1
12.2
17.2
2.9
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1H / ppm

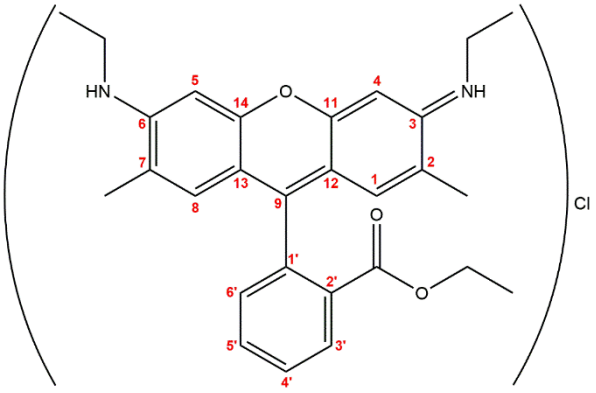

Vitamin B12, [1H,1H] COSY (500 MHz, CD3OD)
1
2
3
4
5
6
7
1H / ppm
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1H / ppm


Vitamin B12, 13C{1H} NMR (125 MHz, CD3OD)
177.6
176.6
175.3
174.2
167.1
143.4
138.2
135.6
133.8
131.5
117.8
112.5
112.4
108.7
105.1
95.7
88.0
86.4
83.7
76.3
75.5
73.6
70.7
62.7
60.4
57.6
56.9
55.0
52.6
46.7
43.9
40.1
36.2
35.3
33.4
32.9
32.4
32.3
29.6
27.4
20.9
20.3
16.4
170
135
105
90
80
70
55
45
35
20
13C / ppm


Vitamin B12, [1H,13C] HSQC (500/125 MHz, CD3OD)
20
30
40
50
60
70
80
90
100
110
120
130
140
13C / ppm
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1H / ppm

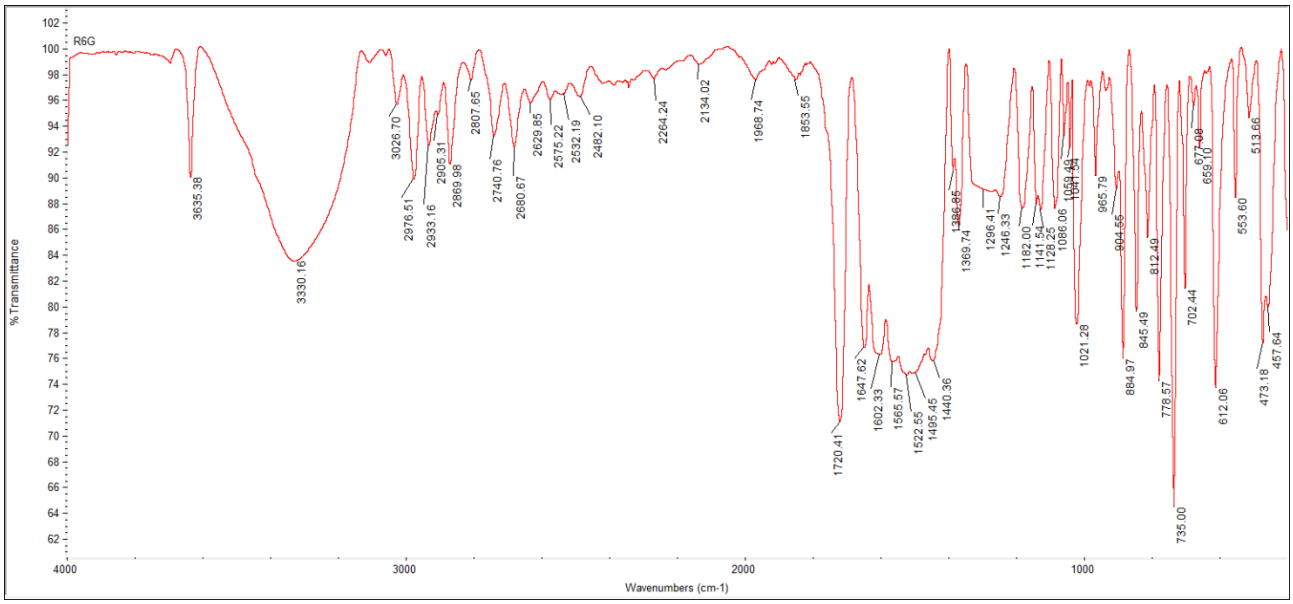


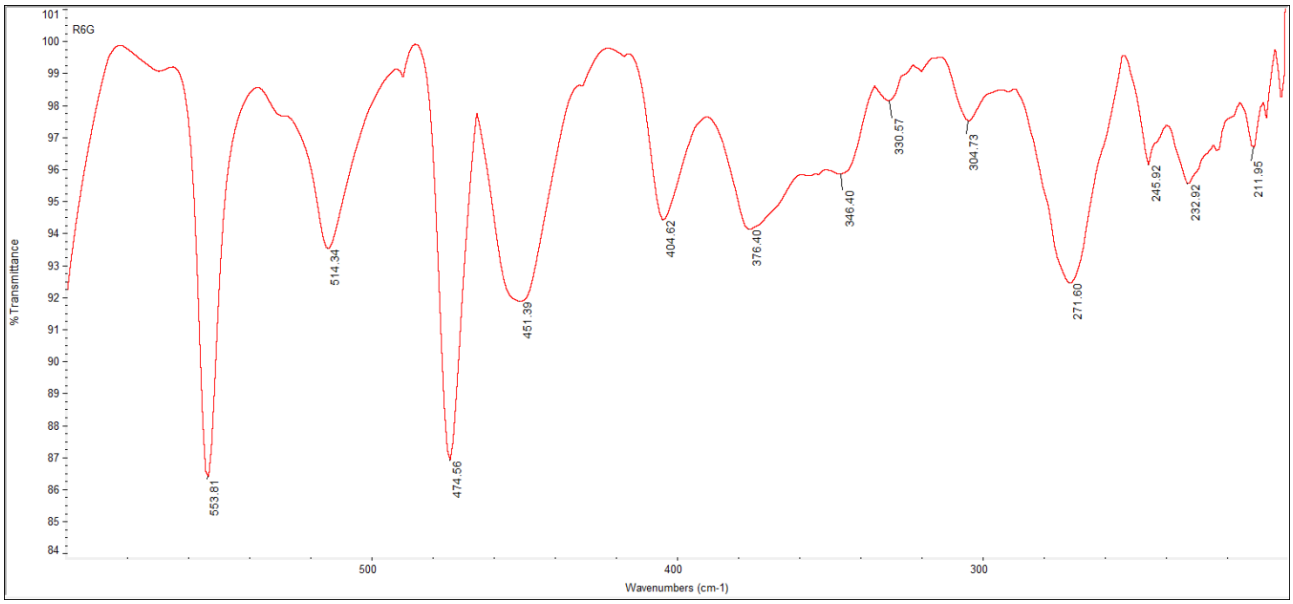
Rhodamine 6G (9-[2-(ethoxycarbonyl)phenyl]-3,6-bis(ethylamino)-2,7-dimethyl-xanthylium chloride, R6G)

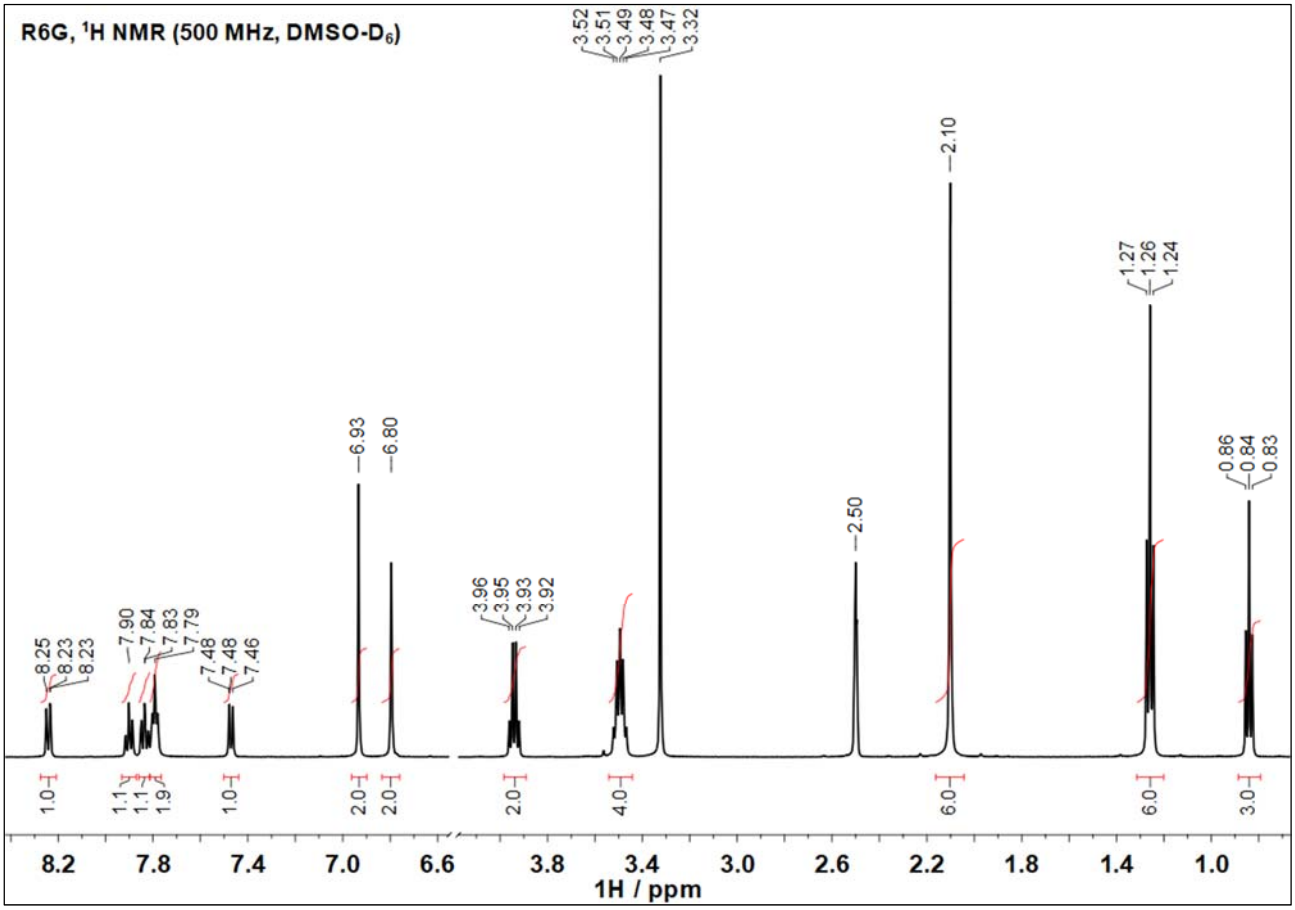


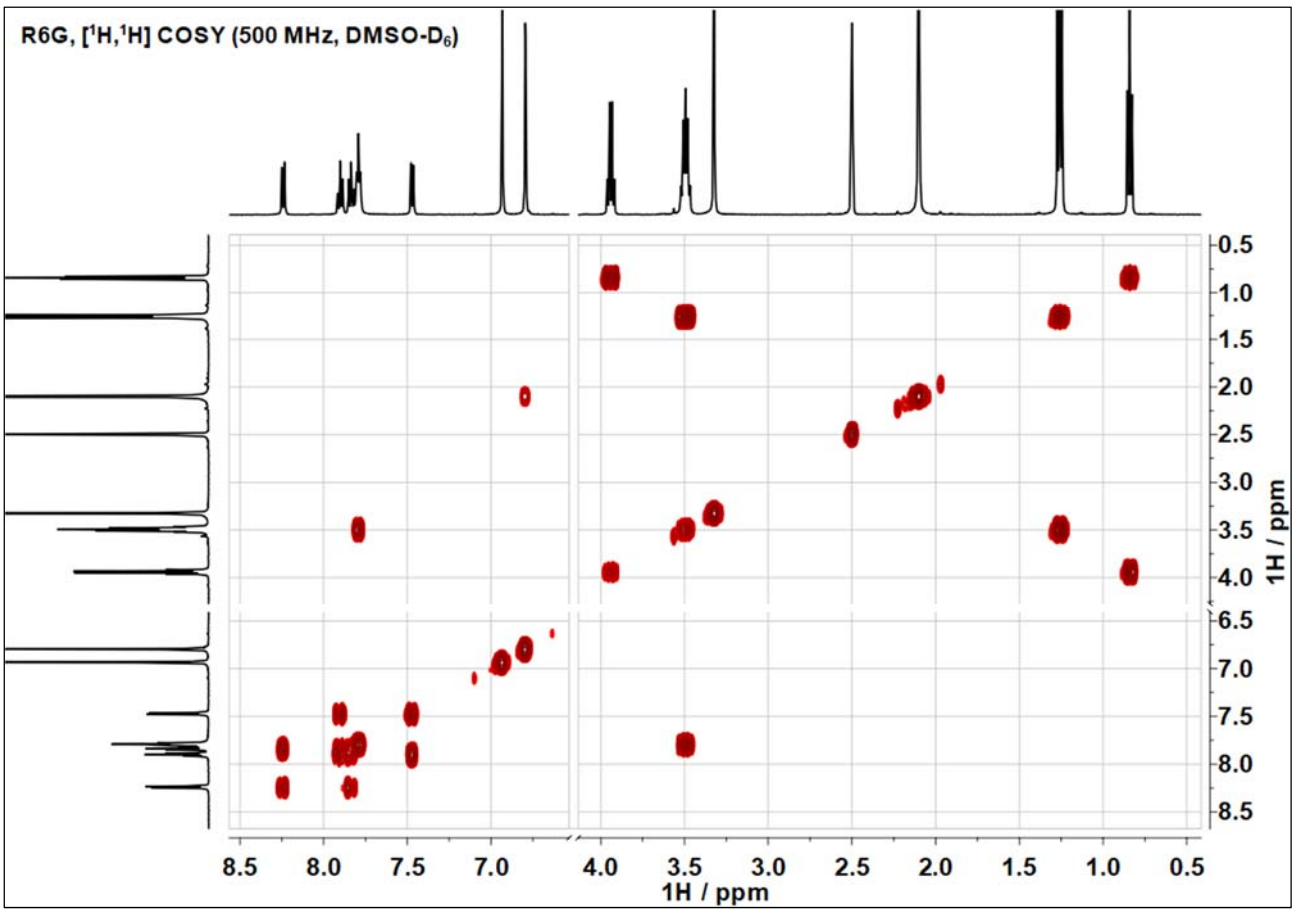
Commercially available (99+%). FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3636 (ν(NH)$_{iminium}$), 3330 (ν(NH)$_{amine}$), 1720 (ν(C=O)), 1648 (ν(C=N)$_{iminium}$), 1566 (δ(C=N–H)$_{iminium}$), 1523 (ν(C–N)$_{amine}$), 1296 (ν(C–Ph)$_{xanthene}$ + ν(C–O–C)$_{xanthene}$ + ν(N–Ph)$_{amine}$ overlapped), 1246 (ν(C–OEt)$_{ester}$), 1086 (ν(O–Et)$_{ester}$) cm^{-1} [29,32,100]. ^{1}H NMR (500 MHz; DMSO-D$_6$; 298 K): δ 0.84 (3 H, t, C$\boldsymbol{H}_{3(ester)}$, $^3J_{H,H}$ = 7.1 Hz), 1.26 (6 H, t, C$\boldsymbol{H}_{3(amine/iminium)}$, $^3J_{H,H}$ = 7.1 Hz), 2.10 (6 H, s, C^{2,7}C$\boldsymbol{H}_3$), 3.45-3.55 (4 H, m, C$\boldsymbol{H}_{2(amine/iminium)}$), 3.94 (2 H, q, C$\boldsymbol{H}_{2(ester)}$, $^3J_{H,H}$ = 7.1 Hz), 6.80 (2 H, s, C$^{1,8}\boldsymbol{H}$), 6.93 (2 H, s, C$^{4,5}\boldsymbol{H}$), 7.47 (1 H, dd, C$^{6'}\boldsymbol{H}$, , $^3J_{H,H}$ = 7.5 Hz, $^4J_{H,H}$ = 0.8 Hz), 7.79 (2 H, br t, N$\boldsymbol{H}_{(amine/iminium)}$, $^3J_{H,H}$ = 5.7 Hz),

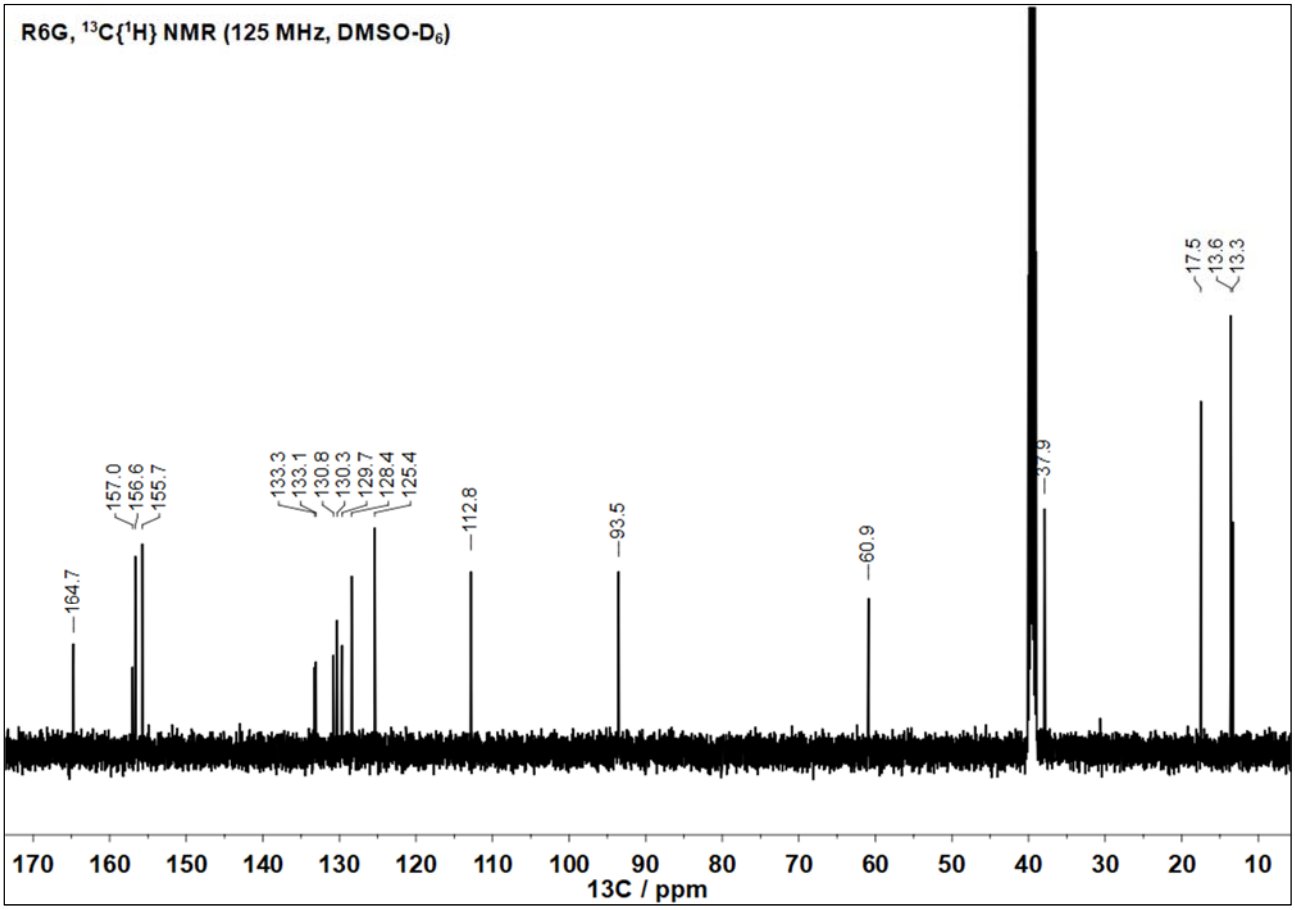
7.83 (1 H, td, C$^{4'}$***H***, $^{3}J_{H,H}$ = 7.7 Hz, $^{4}J_{H,H}$ = 1.2 Hz), 7.90 (1 H, td, C$^{5'}$***H***, $^{3}J_{H,H}$ = 7.5 Hz, $^{4}J_{H,H}$ = 1.2 Hz), 8.24 (1 H, dd, C$^{3'}$***H***, , $^{3}J_{H,H}$ = 7.8 Hz, $^{4}J_{H,H}$ = 0.9 Hz) ppm [100]. $^{13}C\{^{1}H\}$ NMR (125 MHz; DMSO-D$_6$; 298 K): δ 13.3 (***C***H$_{3(ester)}$), 13.6 (***C***H$_{3(amine/iminium)}$), 17.5 (C^{2,7}***C***H$_3$), 37.9 (***C***H$_{2(amine/iminium)}$), 60.9 (***C***H$_{2(ester)}$), 93.5 (***C***^{4,5}H), 112.8 (***C***^{12,13}), 125.4 (***C***^{2,7}), 128.4 (***C***^{1,8}H), 129.7 (***C***$^{4'}$H), 130.3 (***C***$^{6'}$H + ***C***$^{2'}$ overlapped), 130.8 (***C***$^{3'}$H), 133.1 (***C***$^{5'}$H), 133.3 (***C***$^{1'}$), 155.7 (***C***^{3,6}), 156.6 (***C***^{11,14}), 157.0 (***C***^{9}), 164.7 (***C***=O) ppm [100].

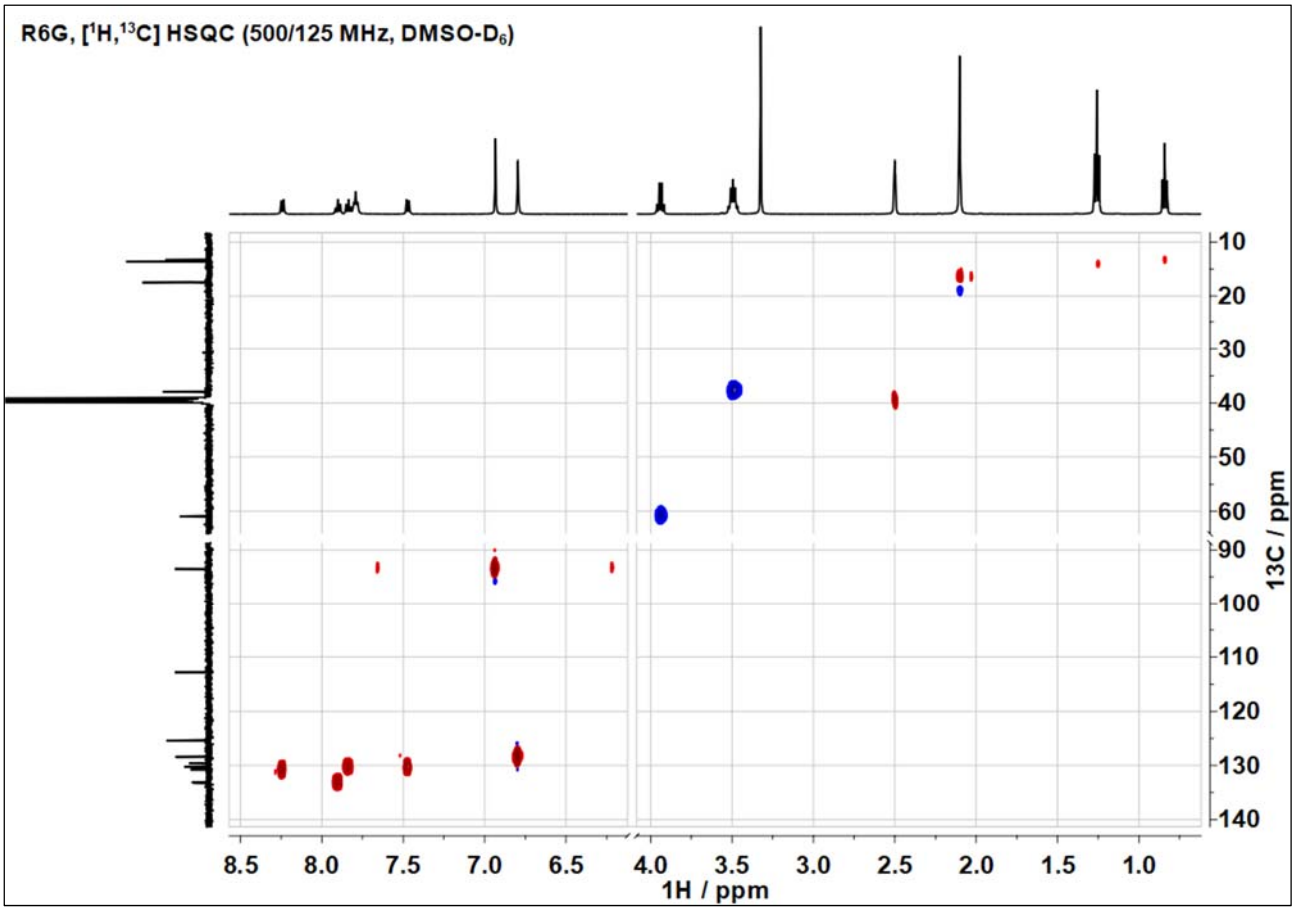




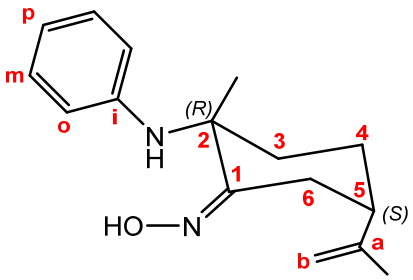

R6G, ¹H NMR (500 MHz, DMSO-D6)
3.52
3.51
3.49
3.48
3.47
3.32
2.10
1.27
1.26
1.24
6.93
6.80
0.86
0.84
0.83
2.50
3.96
3.95
3.93
3.92
8.25
8.23
8.23
7.90
7.84
7.83
7.79
7.48
7.48
7.46
1.0
1.1
1.1
1.9
1.0
2.0
2.0
2.0
4.0
6.0
6.0
3.0
8.2
7.8
7.4
7.0
6.6
3.8
3.4
3.0
2.6
2.2
1.8
1.4
1.0
1H / ppm


R6G, [¹H,¹H] COSY (500 MHz, DMSO-D6)
0.5
1.0
1.5
2.0
2.5
3.0
3.5
4.0
6.5
7.0
7.5
8.0
8.5
1H / ppm
8.5
8.0
7.5
7.0
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
1H / ppm

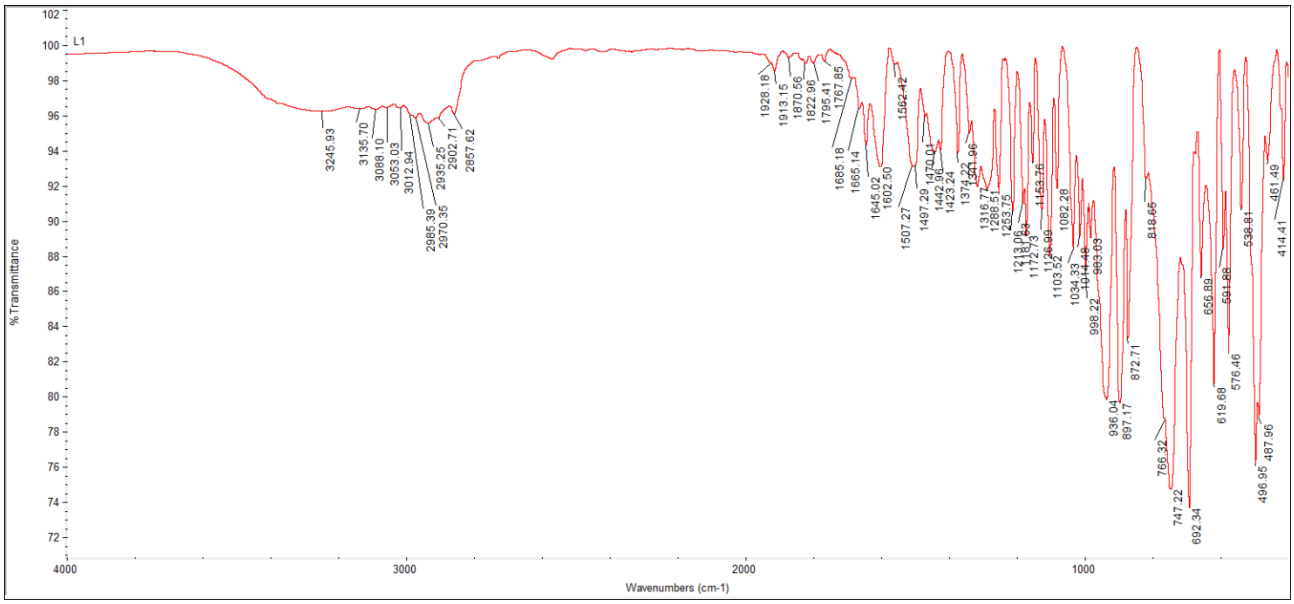

R6G, $^{13}C\{^{1}H\}$ NMR (125 MHz, DMSO-D_6)
164.7
157.0
156.6
155.7
133.3
133.1
130.8
130.3
129.7
128.4
125.4
112.8
93.5
60.9
37.9
17.5
13.6
13.3
170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10
13C / ppm

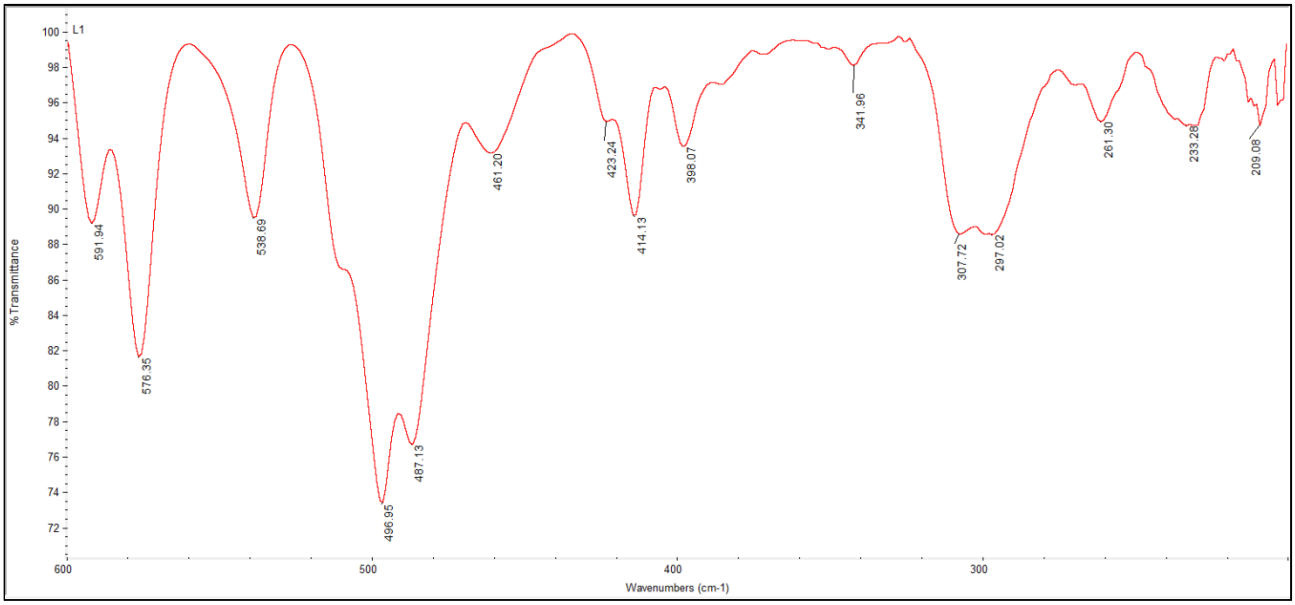

R6G, [^{1}H,^{13}C] HSQC (500/125 MHz, DMSO-D_6)
10 20 30 40 50 60 90 100 110 120 130 140
13C / ppm
8.5 8.0 7.5 7.0 6.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0
1H / ppm

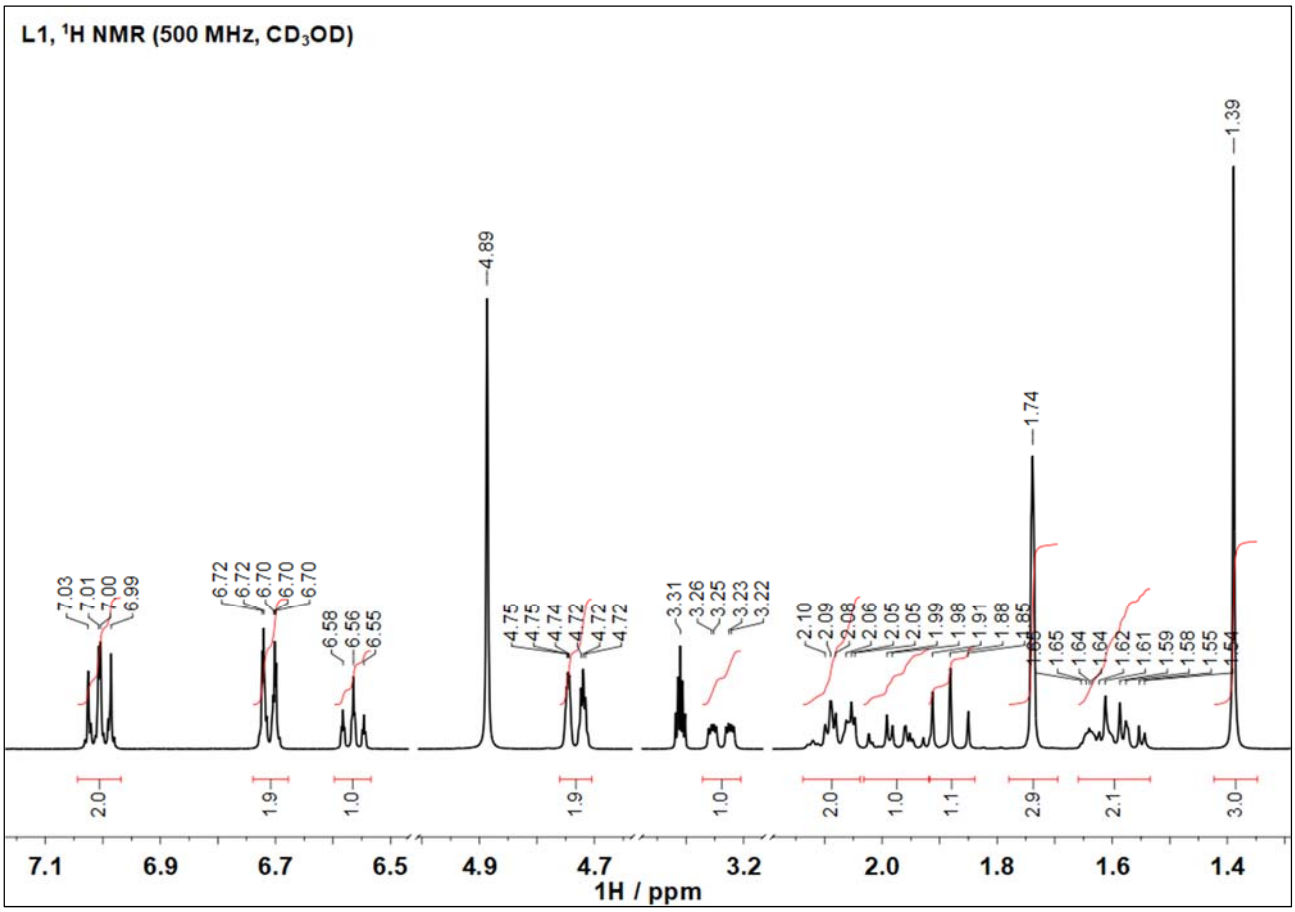
(2*R*,5*S*)-2-Methyl-5-(1-methylethenyl)-2-(phenylamino)-cyclohexanone oxime (L1)

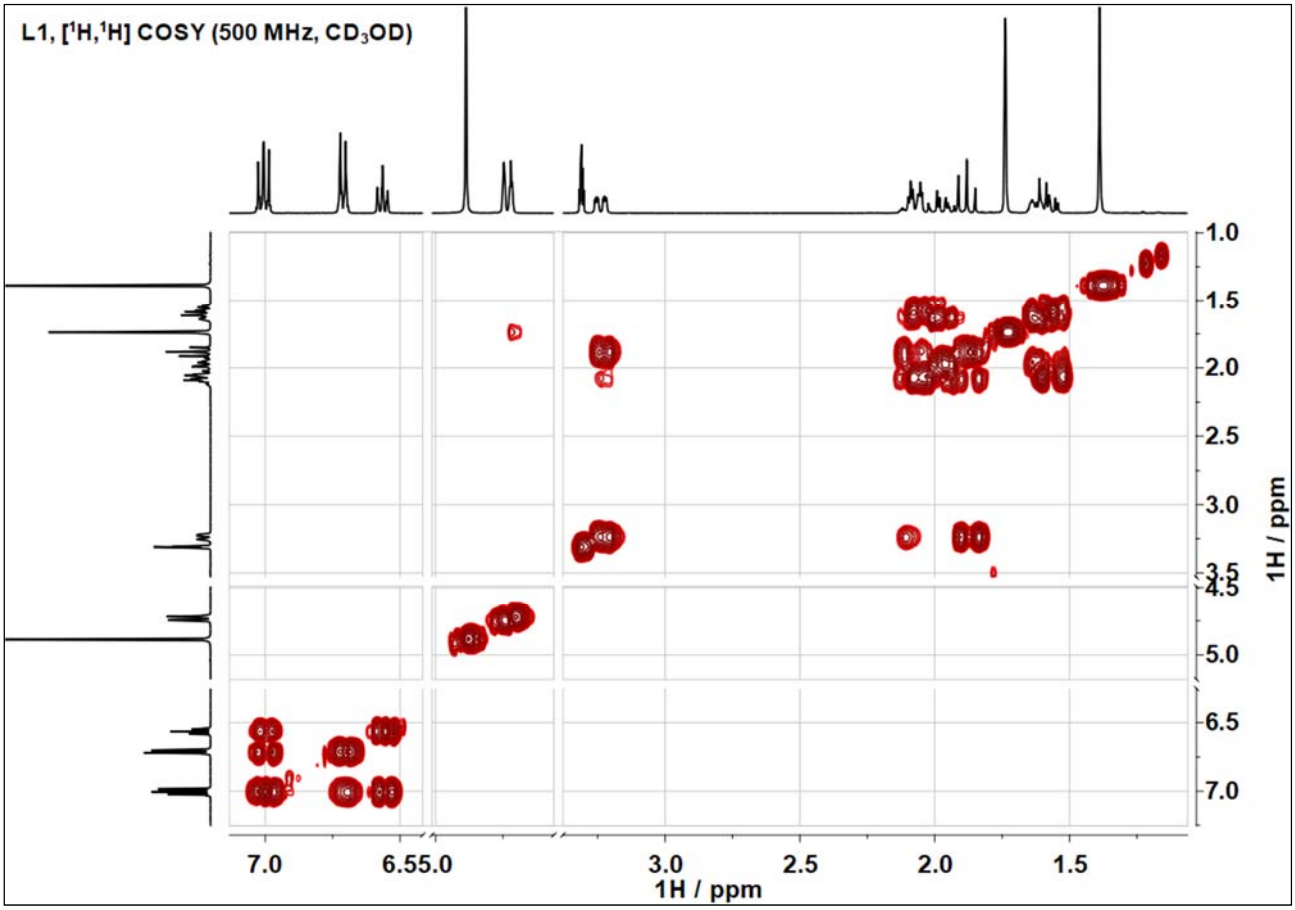


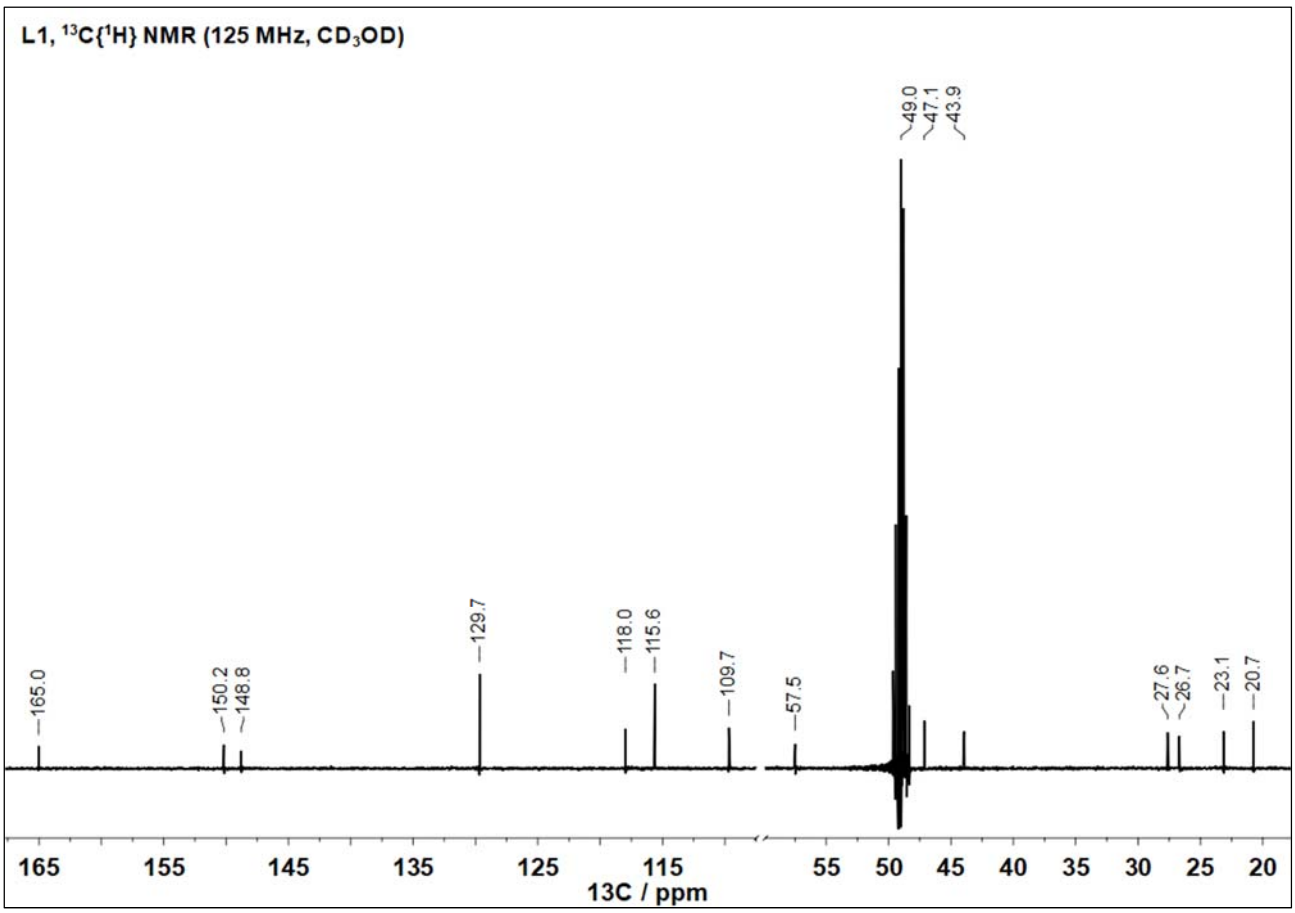
The ligand **L1** was synthesized as previously described [45]. FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3246/3136 (ν(OH)$_{oxime}$ + ν(NH)$_{amine}$ overlapped), 1665 (ν(C=N)$_{oxime}$), 1645 (ν(C=C)$_{alkene}$), 1289 (ν(Ph–N)), 936 (ν(N–OH)$_{oxime}$) cm^{-1} [101]. ^{1}H NMR (500 MHz; CD_3OD; 298 K): δ 1.39 (3 H, s, C^{2}C***H***$_3$), 1.54-1.61 (1 H, m, C^{3}***H***$_{ax}$), 1.62-1.66 (1 H, m, C^{4}***H***$_{ax}$), 1.74 (3 H, s, C^{a}C***H***$_3$), 1.85-1.91 (1 H, m, C^{6}***H***$_{ax}$), 1.96-2.03 (1 H, m, C^{4}***H***$_{eq}$), 2.04-2.06 (1 H, m, C^{3}***H***$_{eq}$), 2.08-2.10 (1 H, m, C^{5}***H***), 3.22-3.26 (1 H, m, C^{6}***H***$_{eq}$), 4.72-4.75 (2 H, m, C^{b}***H***$_{2/diastereotopic}$), 6.56 (1 H, tt, *p*-C***H***), 6.70-6.72 (2 H, m, *o*-C***H***), 6.99-7.03 (2 H, m, *m*-C***H***) ppm. ^{13}C{^{1}H} NMR (125 MHz; CD_3OD; 298 K): δ 20.7 (C^{a}***C***H$_3$), 23.1 (C^{2}***C***H$_3$), 26.7 (***C***^{6}H$_2$), 27.6 (***C***^{4}H$_2$), 43.9 (***C***^{3}H$_2$), 47.1 (***C***^{5}H), 57.5 (***C***^{2}), 109.7 (***C***^{b}H$_2$), 115.6 (*o*-***C***H), 118.0 (*p*-***C***H), 129.7 (*m*-***C***H), 148.8 (*i*-***C***), 150.2 (***C***^{a}), 165.0 (***C***^{1}) ppm [45]. [^{1}H,^{13}C] HMBC (100.6 MHz; $CDCl_3$; 293 K): δ 57.0 (***C***^{2}), 164.9 (***C***^{1}) ppm. [^{1}H,^{15}N] HMBC (40.5 MHz; $CDCl_3$; 293 K): δ 84.1 (C^{2}***N***(Ph)H), 343.5 (C^{1}***N***OH) ppm.

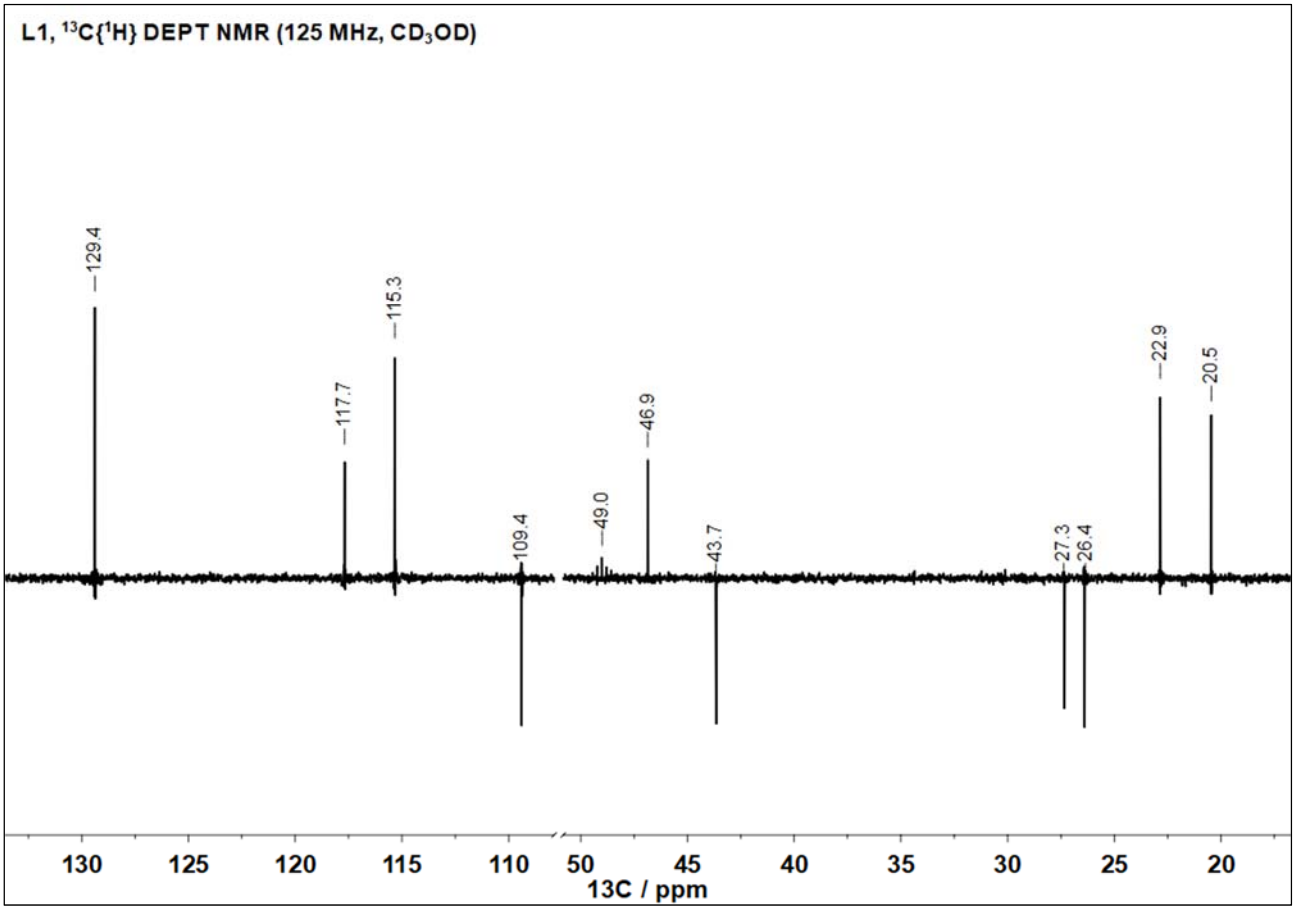


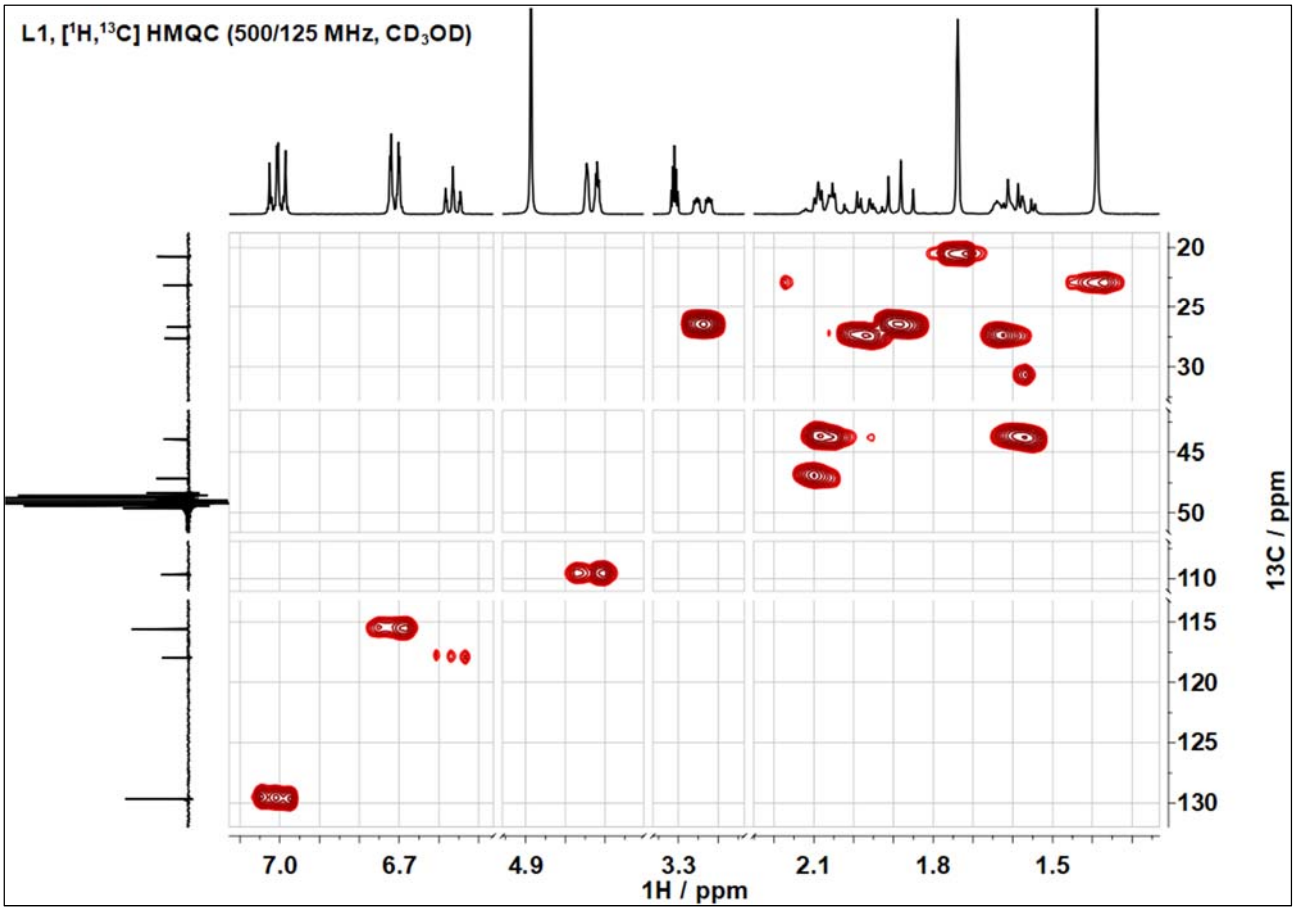

L1
% Transmittance
Wavenumbers (cm-1)
591.94
576.35
538.69
496.95
487.13
461.20
423.24
414.13
398.07
341.96
307.72
297.02
261.30
233.28
209.08


L1, ¹H NMR (500 MHz, CD3OD)
1H / ppm
4.89
1.74
1.39

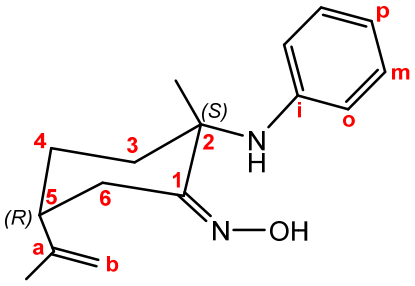

L1, [¹H,¹H] COSY (500 MHz, CD3OD)
1.0
1.5
2.0
2.5
3.0
3.5
4.5
5.0
6.5
7.0
1H / ppm
7.0
6.55.0
3.0
2.5
2.0
1.5
1H / ppm


L1, ¹³C{¹H} NMR (125 MHz, CD3OD)
165.0
150.2
148.8
129.7
118.0
115.6
109.7
57.5
49.0
47.1
43.9
27.6
26.7
23.1
20.7
165
155
145
135
125
115
55
50
45
40
35
30
25
20
13C / ppm

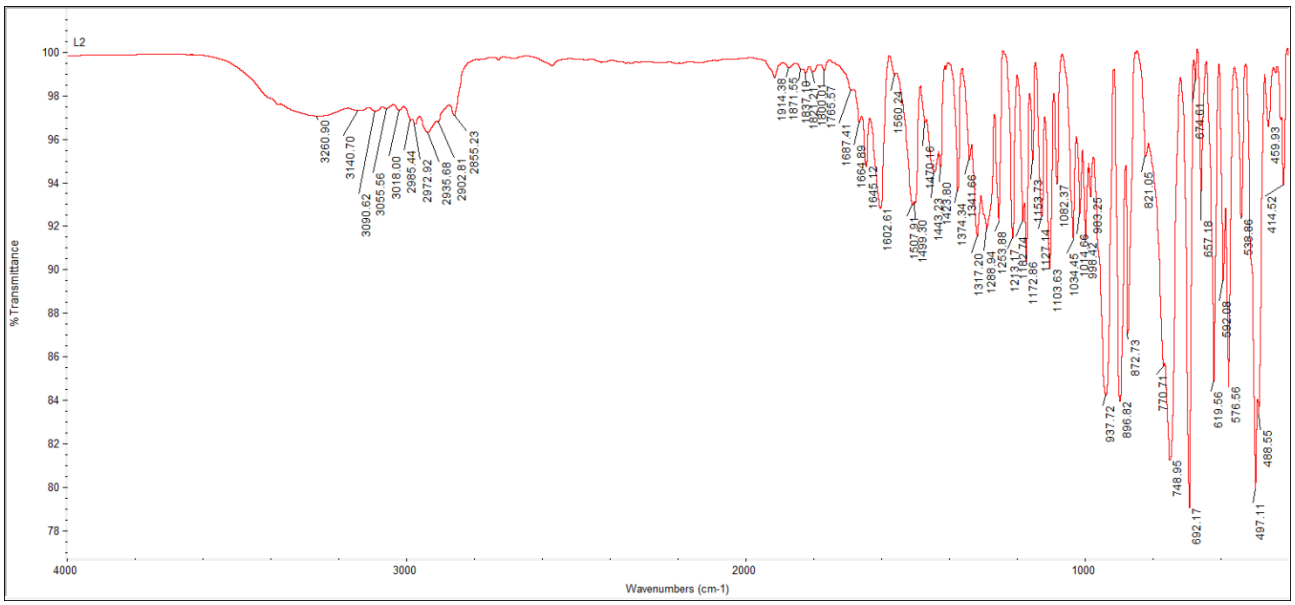

L1, $^{13}C\{^{1}H\}$ DEPT NMR (125 MHz, CD_3OD)
129.4
117.7
115.3
109.4
49.0
46.9
43.7
27.3
26.4
22.9
20.5
130
125
120
115
110
50
45
40
35
30
25
20
13C / ppm

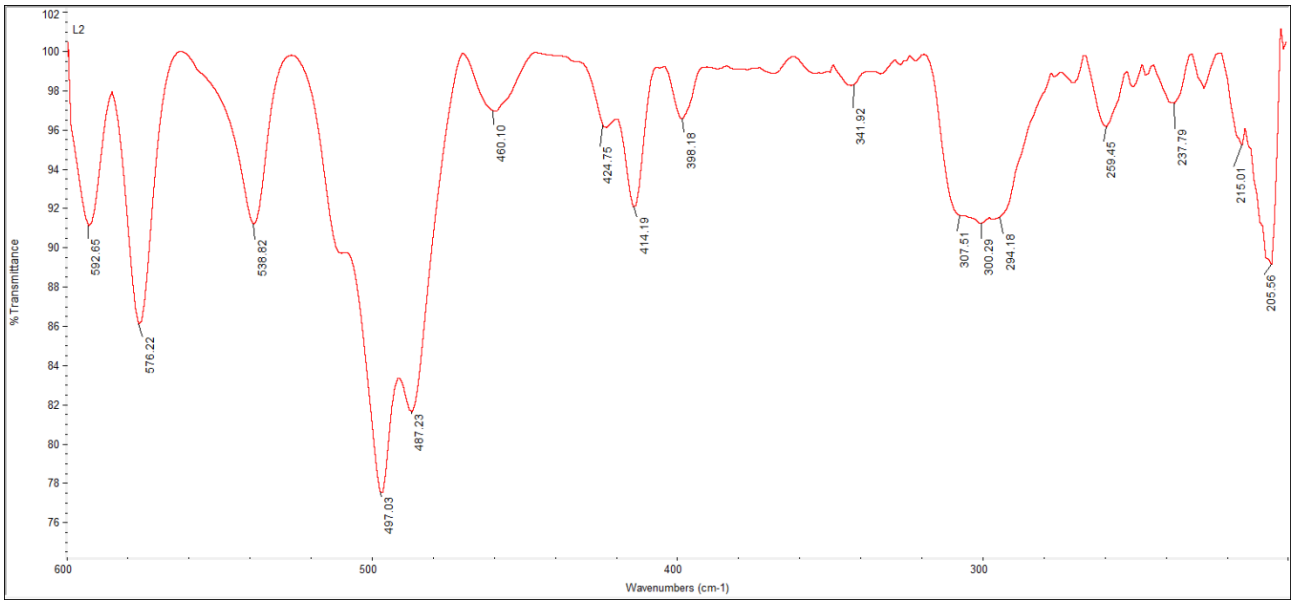

L1, [^{1}H,^{13}C] HMQC (500/125 MHz, CD_3OD)
20
25
30
45
50
110
115
120
125
130
13C / ppm
7.0
6.7
4.9
3.3
2.1
1.8
1.5
1H / ppm

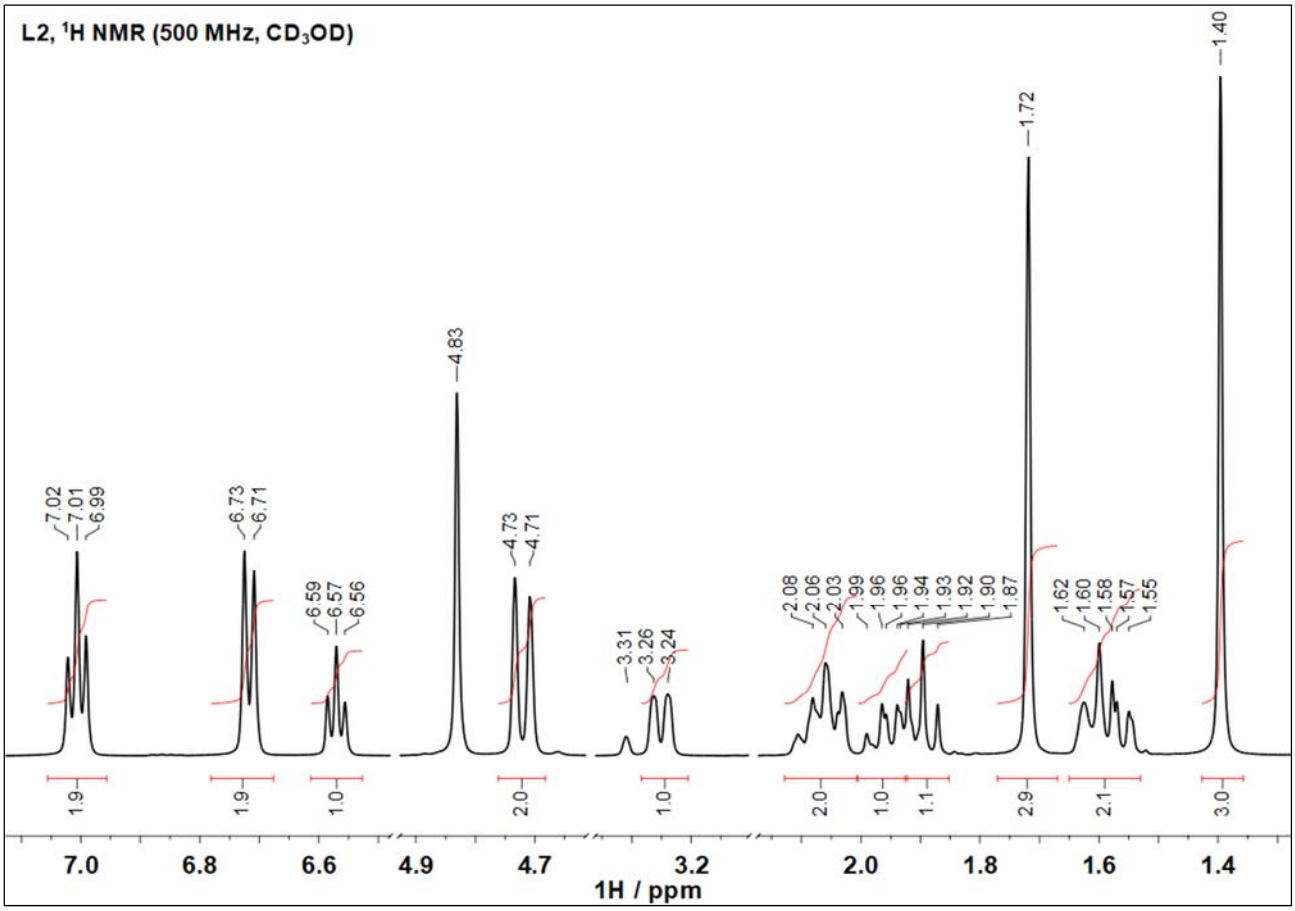
(2*S*,5*R*)-2-Methyl-5-(1-methylethenyl)-2-(phenylamino)-cyclohexanone oxime (L2)

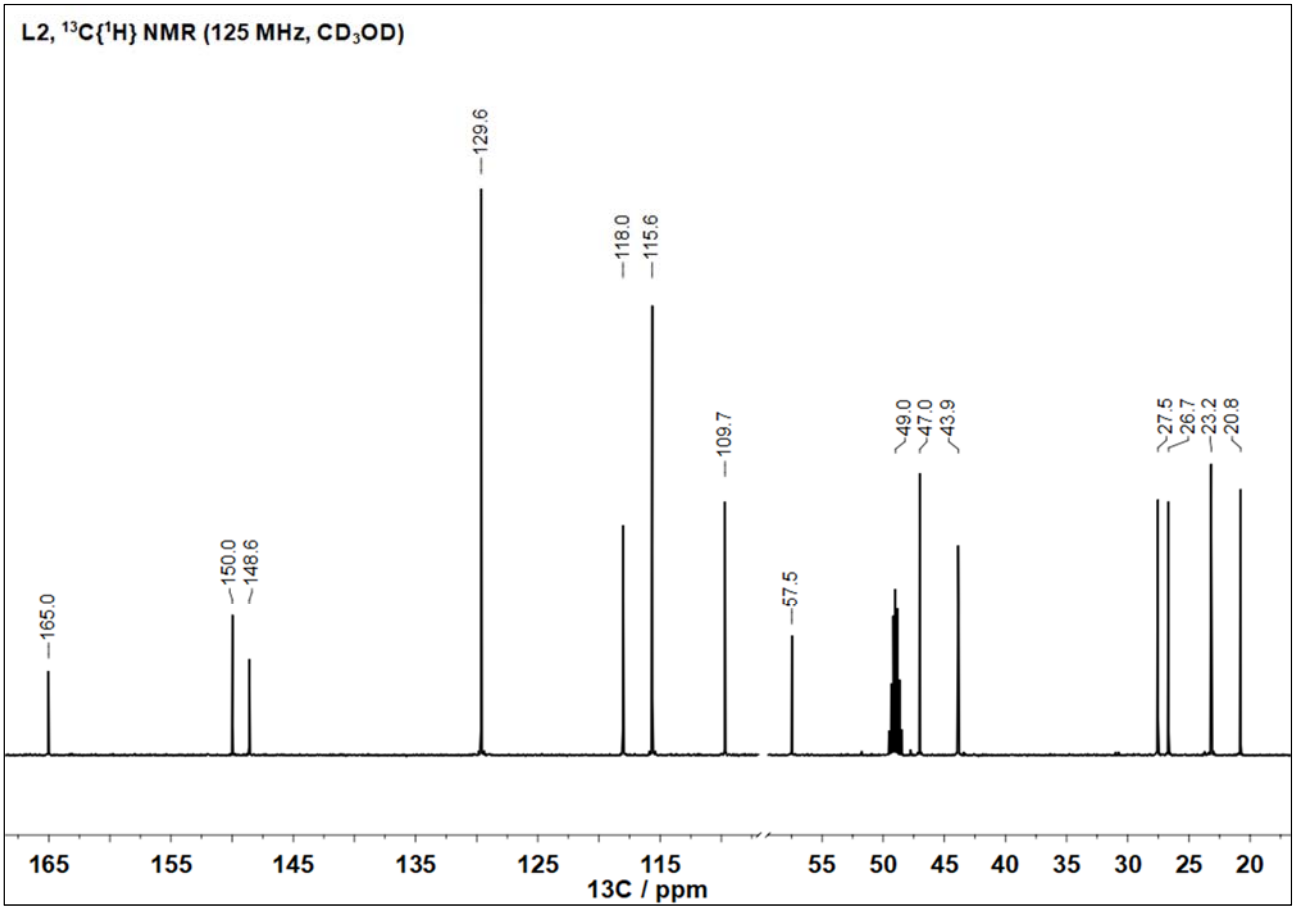


The ligand **L2** was synthesized as previously described [45] FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3261/3141 (ν(OH)$_{oxime}$ + ν(NH)$_{amine}$ overlapped), 1665 (ν(C=N)$_{oxime}$), 1645 (ν(C=C)$_{alkene}$), 1289 (ν(Ph–N)), 938 (ν(N–OH)$_{oxime}$) cm^{-1} [94,101]. ^{1}H NMR (500 MHz; CD_3OD; 298 K): δ 1.40 (3 H, s, C^{2}C***H***$_3$), 1.55-1.59 (1 H, m, C^{3}***H***$_{ax}$), 1.60-1.63 (1 H, m, C^{4}***H***$_{ax}$), 1.72 (3 H, s, C^{a}C***H***$_3$), 1.87-1.92 (1 H, m, C^{6}***H***$_{ax}$), 1.93-1.99 (1 H, m, C^{4}***H***$_{eq}$), 2.03-2.05 (1 H, m, C^{3}***H***$_{eq}$), 2.06-2.09 (1 H, m, C^{5}***H***), 3.24-3.26 (1 H, m, C^{6}***H***$_{eq}$), 4.71-4.73 (2 H, m, C^{b}***H***$_{2/diastereotopic}$), 6.57 (1 H, br tt, *p*-C***H***), 6.71-6.73 (2 H, m, *o*-C***H***), 6.99-7.02 (2 H, m, *m*-C***H***) ppm. ^{13}C{^{1}H} NMR (125 MHz; CD_3OD; 298 K): δ 20.8 (C^{a}***C***H$_3$), 23.2 (C^{2}***C***H$_3$), 26.7 (***C***^{6}H$_2$), 27.5 (***C***^{4}H$_2$), 43.9 (***C***^{3}H$_2$), 47.0 (***C***^{5}H), 57.5 (***C***^{2}), 109.7 (***C***^{b}H$_2$), 115.6 (*o*-***C***H), 118.0 (*p*-***C***H), 129.6 (*m*-***C***H), 148.6 (*i*-***C***), 150.0 (***C***^{a}), 165.0 (***C***^{1}) ppm [45]. [^{1}H,^{13}C] HMBC (100.6 MHz; $CDCl_3$; 293 K): δ 57.0 (***C***^{2}), 164.9 (***C***^{1}) ppm. [^{1}H,^{15}N] HMBC (40.5 MHz; $CDCl_3$; 293 K): δ 84.1 (C^{2}***N***(Ph)H), 343.5 (C^{1}***N***OH) ppm.




L2
% Transmittance
Wavenumbers (cm-1)
592.65
576.22
538.82
497.03
487.23
460.10
424.75
414.19
398.18
341.92
307.51
300.29
294.18
269.45
237.79
215.01
205.56

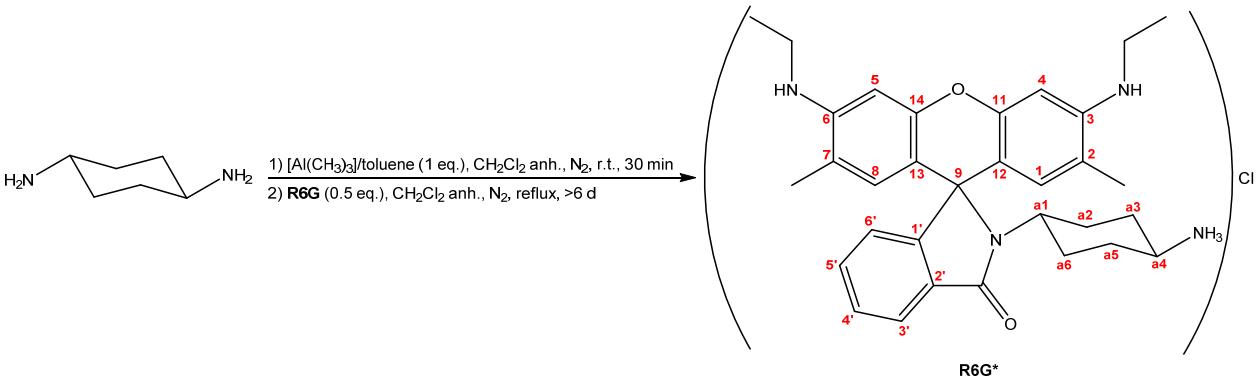

L2, ¹H NMR (500 MHz, CD3OD)
7.02
7.01
6.99
6.73
6.71
6.59
6.57
6.56
4.83
4.73
4.71
3.31
3.26
3.24
2.08
2.06
2.03
1.99
1.96
1.96
1.94
1.93
1.92
1.90
1.87
1.72
1.62
1.60
1.58
1.57
1.55
1.40
1.9
1.9
1.0
2.0
1.0
2.0
1.0
1.1
2.9
2.1
3.0
7.0
6.8
6.6
4.9
4.7
3.2
2.0
1.8
1.6
1.4
1H / ppm


L2, $^{13}C\{^{1}H\}$ NMR (125 MHz, CD_3OD)
165.0
150.0
148.6
129.6
118.0
115.6
109.7
57.5
49.0
47.0
43.9
27.5
26.7
23.2
20.8
165
155
145
135
125
115
55
50
45
40
35
30
25
20
13C / ppm

Synthesis of the vitamin B_{12}–rhodamine 6G adduct

1st Step. Synthesis of the rhodamine 6G–*trans*-1,4-diaminocyclohexane intermediate (**R6G***)

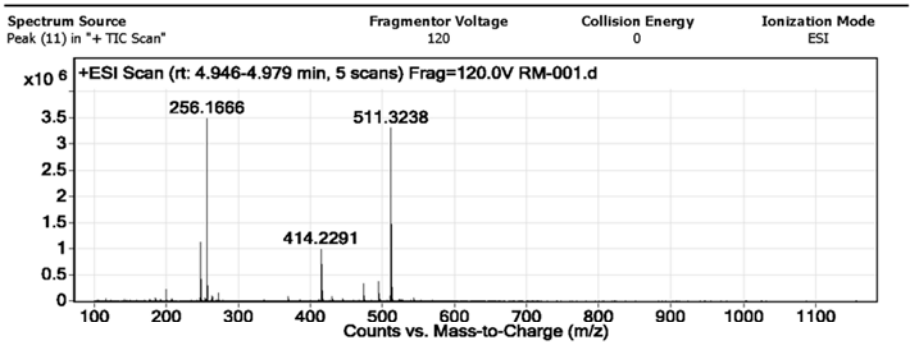
The reaction was carried out in a Schlenk line under an inert atmosphere of nitrogen following a modified literature procedure [28]. A 2 M solution of $Al(CH_3)_3$ in toluene (0.8 mL, 1.6 mmol) was added dropwise over 5 min to a dry DCM solution (125 mL) of *trans*-1,4-diaminocyclohexane (0.41 g, 1.6 mmol) under stirring. After stirring at room temperature for 30 min, rhodamine 6G (**R6G**, 0.38 g, 0.8 mmol) was added in one portion and the mixture was stirred under reflux for a minimum of 6 d. The mixture was then allowed to cool down to room temperature and 0.1 M HCl was added dropwise over 25 min until effervescence ceased. The insoluble residue was filtered off, washed with DCM (250 mL), and the combined filtered solution and washings were evaporated to dryness to give a pinkish solid. The residue was then treated with 0.01 M HCl saturated with NaCl (2 mL) and the aqueous layer was extracted with a 1:1 mixture of DCM and isopropanol (3 × 100 mL). The combined organic extracts were dried with anhydrous Na_2SO_4, filtered and evaporated to dryness. The crude solid was subsequently dissolved with $CHCl_3$ (100 mL), filtered, evaporated to dryness, purified by column chromatography (eluent: DCM/MeOH 6:1), and dried under vacuum over P_2O_5, yielding the title compound as a bright pink solid ($MM(C_{32}H_{39}ClN_4O_2)$ = 547.14 g mol^{-1}, 0.30 g, 69% yield).



R6G*

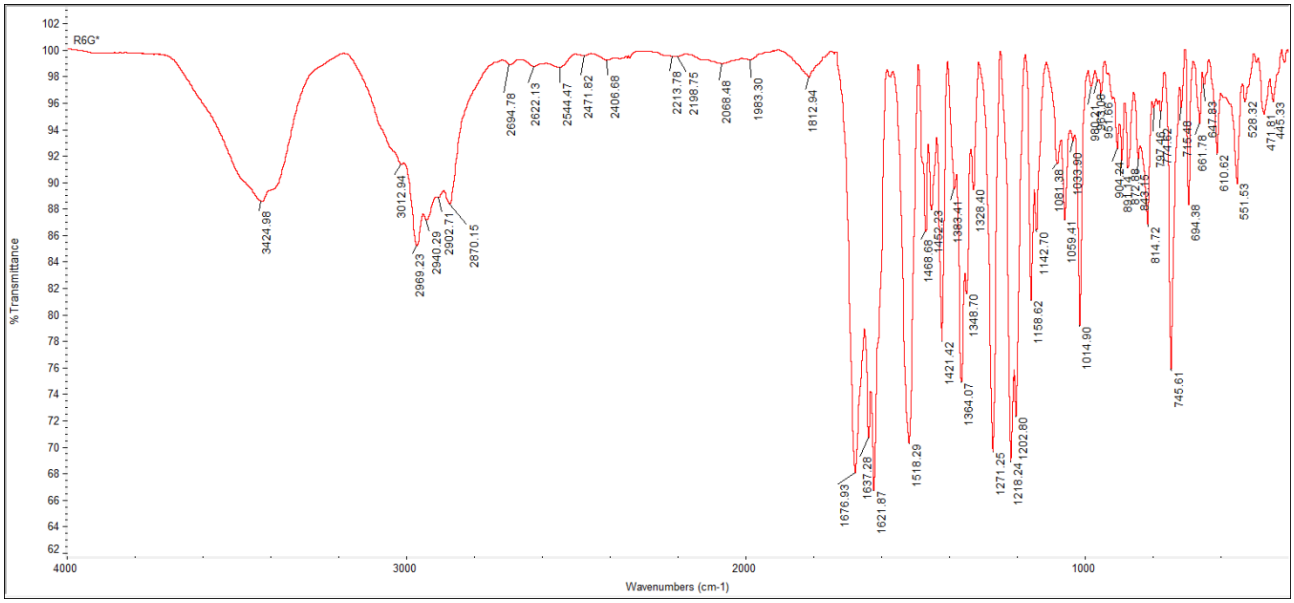
HRMS (CH_3OH, *m/z*) calcd. for $[M\text{-}Cl]^+$: 511.3268; found: 511.3238; $[M\text{-}Cl+H]^{2+}$: 256.1422; found: 256.1666. FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3425 (ν(NH)), ~3000-2800 ($\nu_{a,s}(NH_3^+)$), 1677 ($\nu(C{=}O)_{lactam}$), 1622 ($\delta_a(NH_3^+)$), 1518 (δ(C–N–H) + $\delta_s(NH_3^+)$ overlapped), 1349 ($\nu(N\text{–}Ph)_{amine}$), 1271 ($\nu(C\text{–}O\text{–}C)_{xanthene}$) cm^{-1}. ^{1}H NMR (400 MHz; $CDCl_3$; 298 K): δ 1.04-1.13 (2 H, m, $C^{a3,a5}\boldsymbol{H}_{ax}$), 1.14-1.24 (2 H, m, $C^{a2,a6}\boldsymbol{H}_{ax}$), 1.32 (6 H, t, $C\boldsymbol{H}_{3(amine)}$, $^3J_{H,H}$ = 7.1 Hz), 1.85 (6 H, s, $C^{2,7}C\boldsymbol{H}_3$), 1.88 (2 H, sh, $C^{a3,a5}\boldsymbol{H}_{eq}$), 2.11-2.20 (2 H, m, $C^{a2,a6}\boldsymbol{H}_{eq}$), 2.84-2.89 (1 H, m, $C^{a1}\boldsymbol{H}$), 2.90-2.94 (1 H, m, $C^{a4}\boldsymbol{H}$), 3.16-3.24 (4 H, m, $C\boldsymbol{H}_{2(amine)}$), 3.50 (2 H, br s, $N\boldsymbol{H}_{(amine)}$), 5.95-6.45 (3 H, br s, $N\boldsymbol{H}_3^+$), 6.20 (2 H, s, $C^{1,8}\boldsymbol{H}$), 6.30 (2 H, s, $C^{4,5}\boldsymbol{H}$), 6.93 (1 H, dd, $C^{6'}\boldsymbol{H}$, $^3J_{H,H}$ = 7.4 Hz, $^4J_{H,H}$ = 0.8 Hz), 7.36-7.41 (2 H, m, $C^{4'}\boldsymbol{H}$ + $C^{5'}\boldsymbol{H}$ overlapped), 7.82 (1 H, dd, $C^{3'}\boldsymbol{H}$, $^3J_{H,H}$ = 7.8 Hz, $^4J_{H,H}$ = 1.9 Hz) ppm [28]. $^{13}C\{^1H\}$

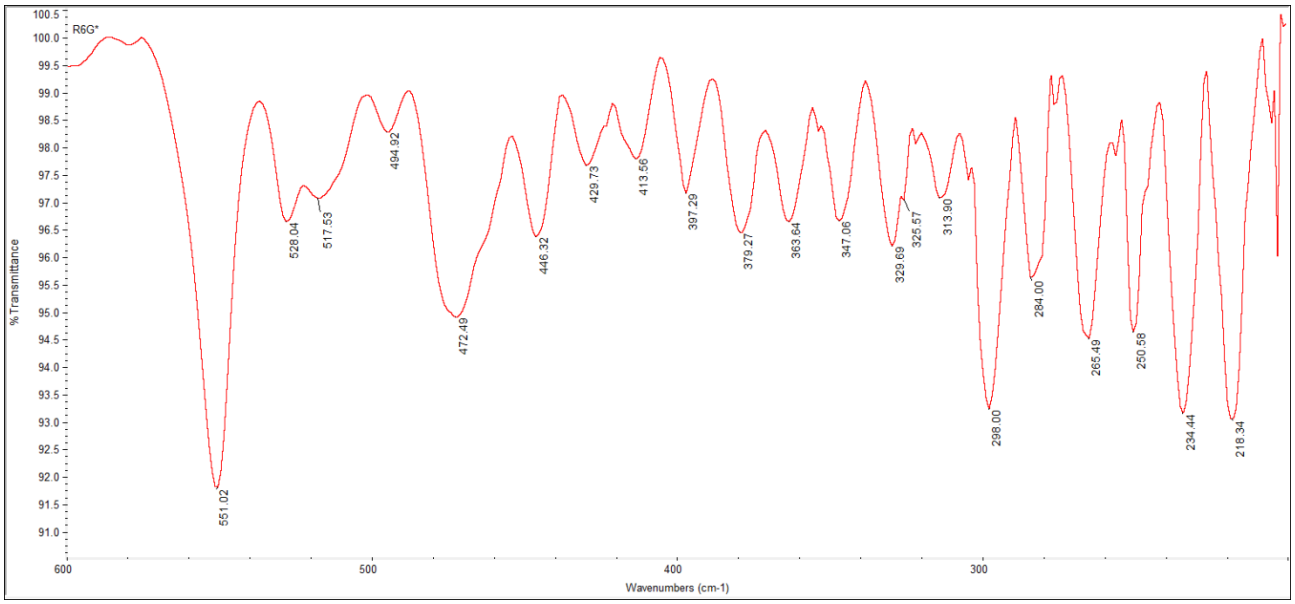
NMR (100 MHz; $CDCl_3$; 298 K): δ 14.8 (***C***$H_{3(amine)}$), 16.7 ($C^{2,7}$***C***H_3), 26.8 (***C***$^{a2,a6}H_2$), 31.3 (***C***$^{a3,a5}H_2$), 38.4 (***C***$H_{2(amine)}$), 49.7 (***C***^{a4}H), 51.8 (***C***^{a1}H), 65.8 (***C***^{9}), 96.7 (***C***^{4,5}H), 105.5 (***C***^{12,13}), 117.6 (***C***^{2,7}), 122.4 (***C***$^{3'}$H), 123.7 (***C***$^{6'}$H), 127.9 (***C***$^{4'}$H), 128.7 (***C***^{1,8}H), 131.8 (***C***$^{2'}$), 132.3 (***C***$^{5'}$H), 147.4 (***C***^{3,6}), 151.6 (***C***^{11,14}), 153.6 (***C***$^{1'}$), 167.6 (***C***=$O_{(lactam)}$) ppm [28].

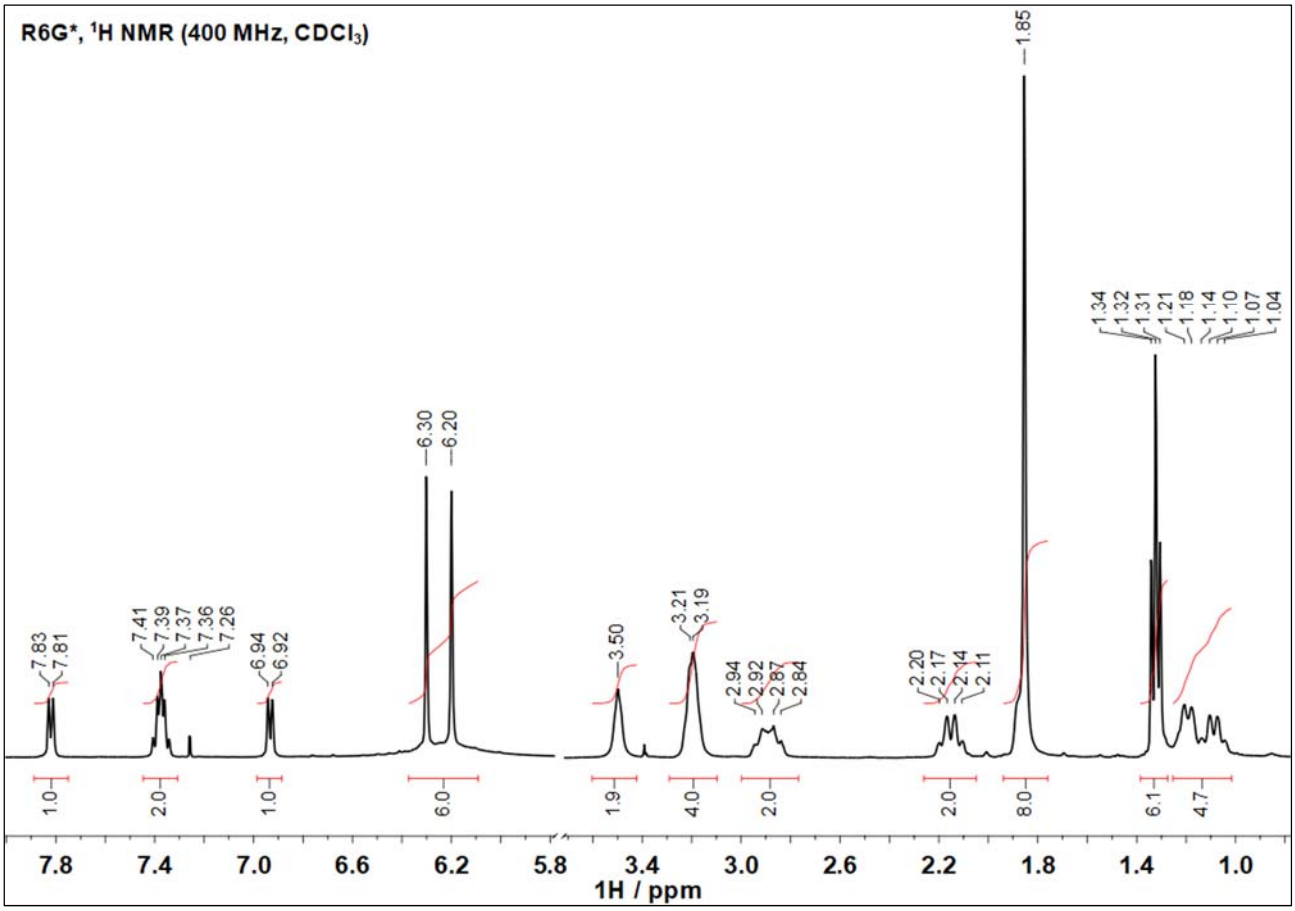


Peak List

m/z	z	Abund
247.651	2	1120574.88
248.1529	2	405233.78
256.1666	2	3485393.25
256.6665	2	1607518.63
414.2291		990494.69
415.2221	1	704549.56
473.2203	1	333023.75
494.2942	1	373483.06
511.3238	1	3301104.75
512.3246	1	1463571




R6G*
% Transmittance
Wavenumbers (cm-1)
551.02
528.04
517.53
494.92
472.49
446.32
429.73
413.56
397.29
379.27
363.64
347.06
329.69
325.57
313.90
298.00
284.00
265.49
250.58
234.44
218.34


R6G*, ¹H NMR (400 MHz, CDCl3)
1H / ppm
7.83
7.81
7.41
7.39
7.37
7.36
7.26
6.94
6.92
6.30
6.20
3.50
3.21
3.19
2.94
2.92
2.87
2.84
2.20
2.17
2.14
2.11
1.85
1.34
1.32
1.31
1.21
1.18
1.14
1.10
1.07
1.04
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1.0
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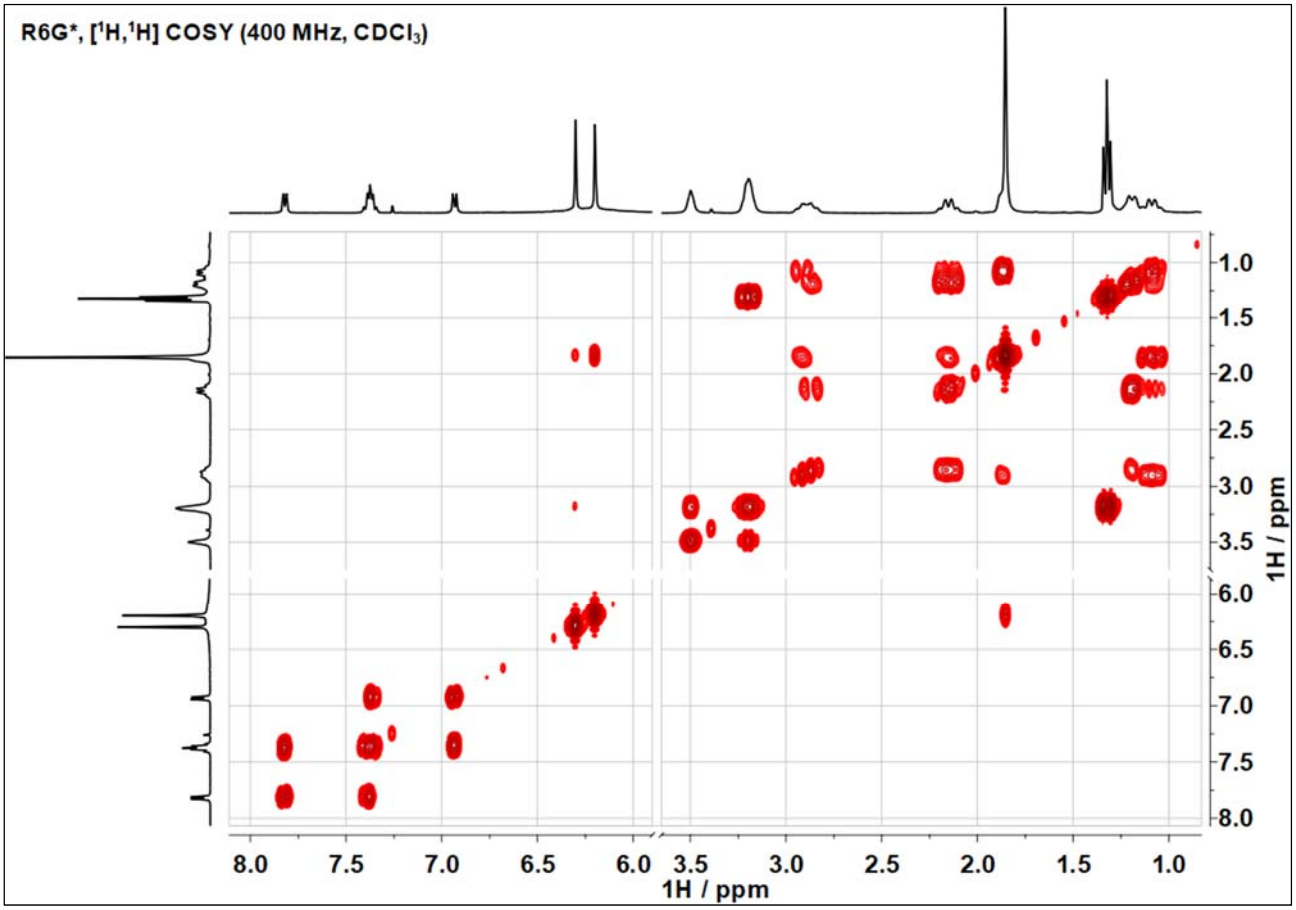
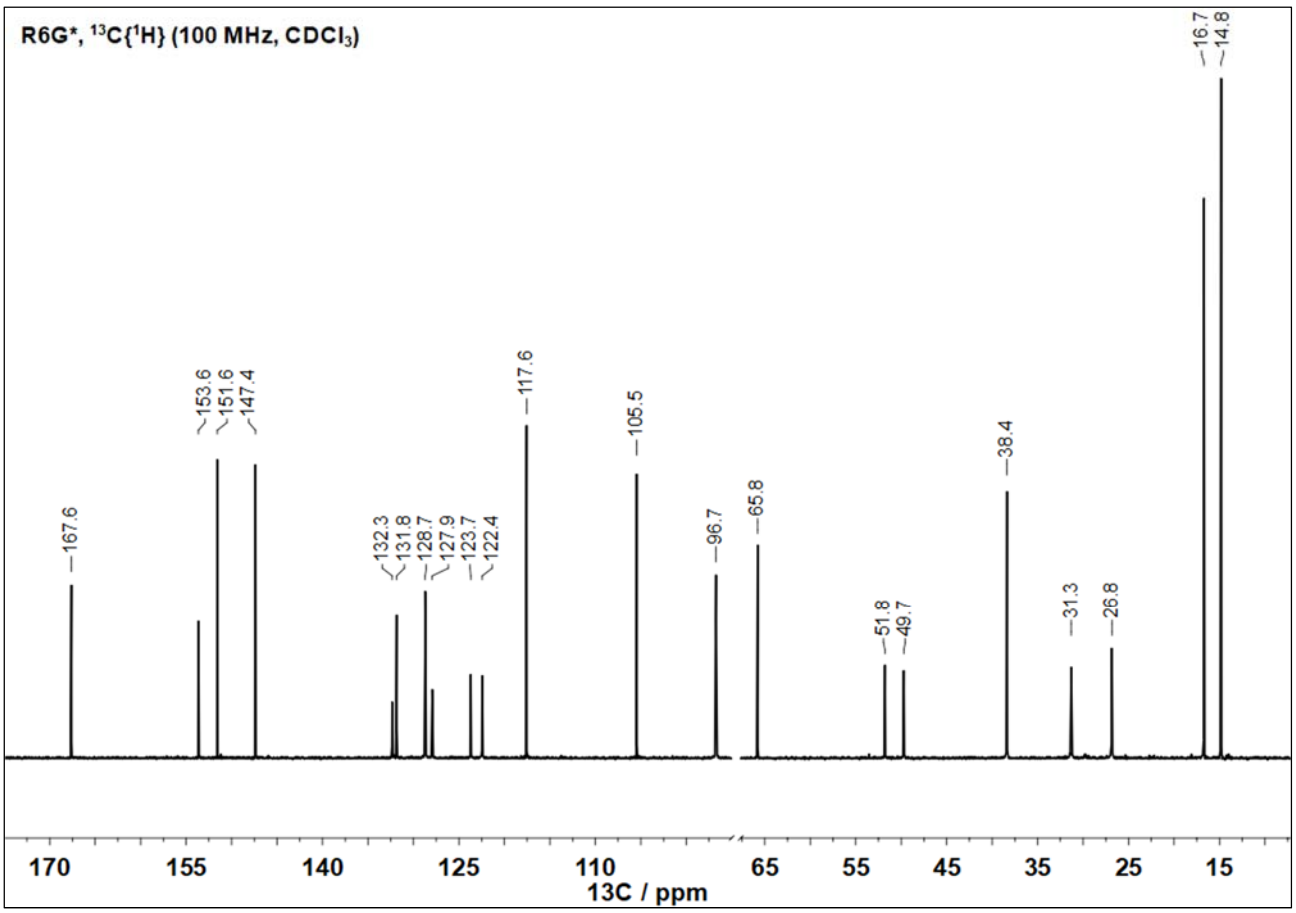
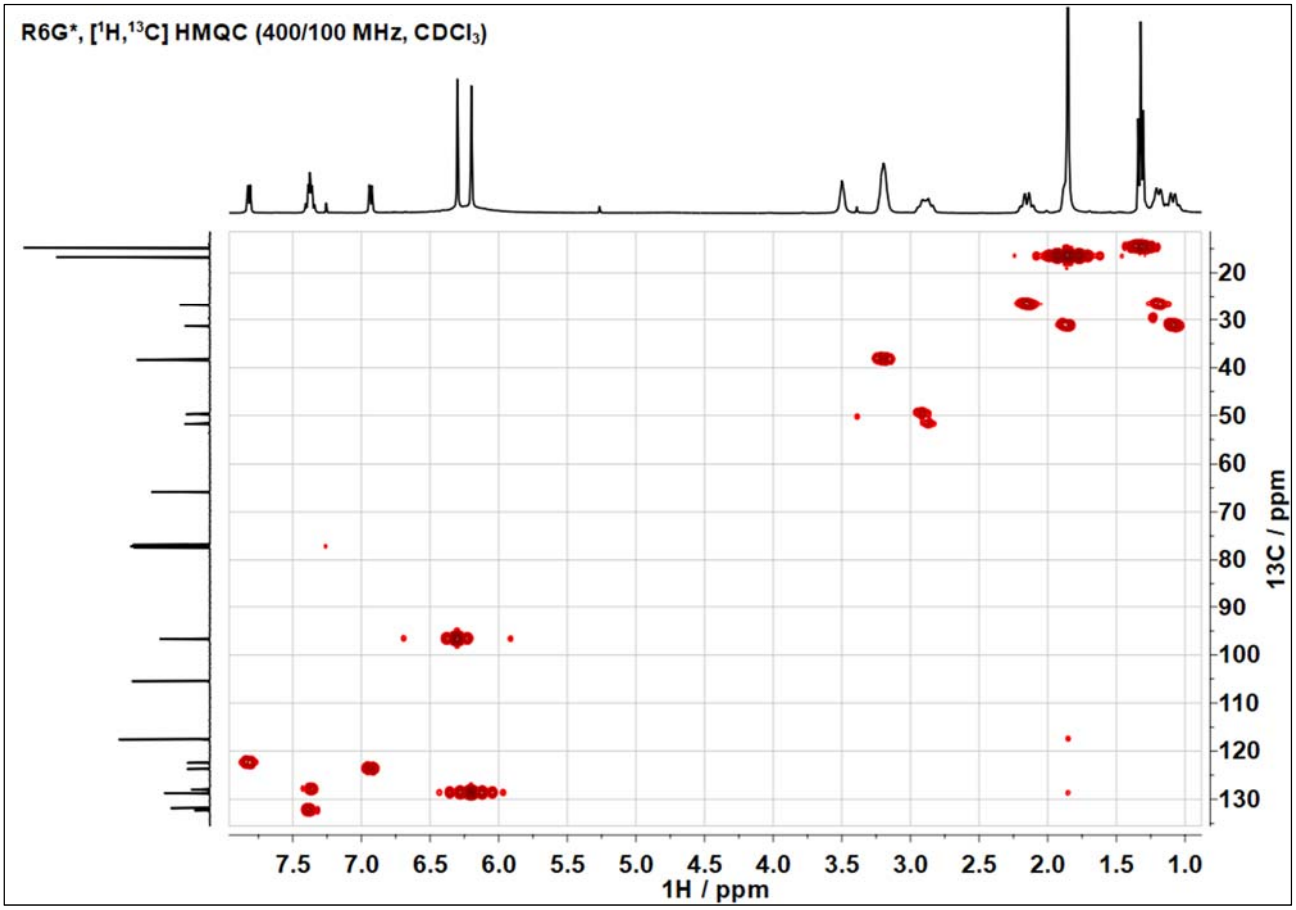
4.0
2.0
2.0
8.0
6.1
4.7

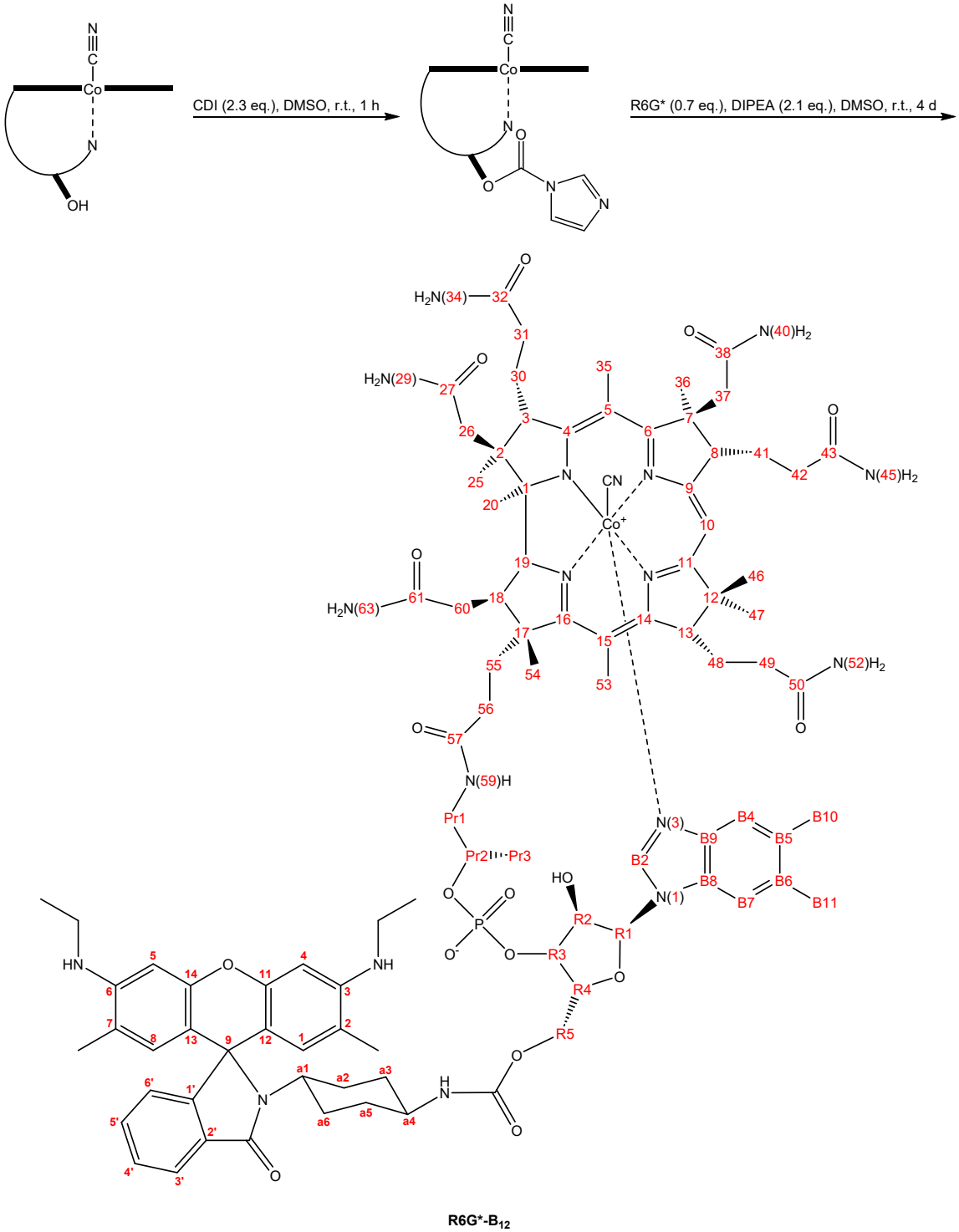

R6G*, [¹H,¹H] COSY (400 MHz, CDCl3)
8.0
7.5
7.0
6.5
6.0
3.5
3.0
2.5
2.0
1.5
1.0
1H / ppm
1.0
1.5
2.0
2.5
3.0
3.5
6.0
6.5
7.0
7.5
8.0
1H / ppm


R6G*, ¹³C{¹H} (100 MHz, CDCl3)
167.6
153.6
151.6
147.4
132.3
131.8
128.7
127.9
123.7
122.4
117.6
105.5
96.7
65.8
51.8
49.7
38.4
31.3
26.8
16.7
14.8
170
155
140
125
110
65
55
45
35
25
15
13C / ppm



2nd Step. Synthesis of the rhodamine 6G–*trans*-1,4-diaminocyclohexane–vitamin B_{12} conjugate (**R6G*-B_{12}**)

The reaction was carried out following a modified literature procedure [28]. Vitamin B_{12} (0.090 g, 0.07 mmol) was dissolved in DMSO (5 mL) and treated with CDI (0.026 g, 0.16 mmol). After stirring at room temperature for 1 h, **R6G*** (0.026 g, 0.05 mmol) and DIPEA (20.96 µL, 0.150 mmol) were added and the mixture was stirred at room temperature in the dark for 4 d. Addition of a 10:1 diethyl ether/DMF mixture (200 mL) led to the precipitation of a dark pink residue which was washed with diethyl ether (3 × 10 mL) and dried under vacuum over P_2O_5. The crude product was then purified by column chromatography (eluent: MeOH/CH_3CN/H_2O 30:1:0.5), and dried under vacuum over P_2O_5, yielding the title compound as a dark pink solid (**R6G*-B_{12}**, MM($C_{96}H_{124}CoN_{18}O_{17}P$) = 1892.06 g mol^{-1}, 0.068 g, 71% yield).

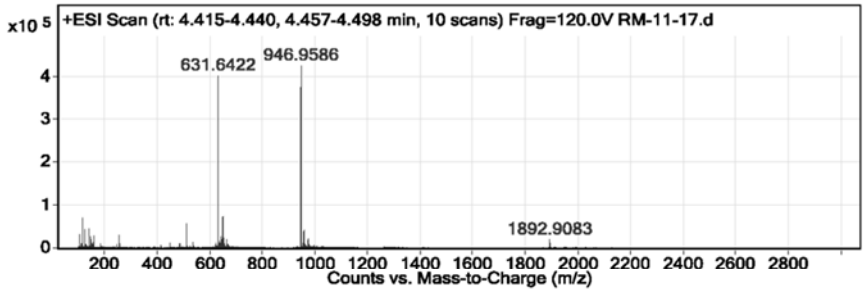


R6G*-B$_{12}$

HRMS (CH_3OH, *m/z*) calcd. for $[M+H]^+$: 1892.8564; found: 1892.9083; $[M+2H]^{2+}$: 946.9319; found: 946.9586; $[M+3H]^{3+}$: 631.6237; found: 631.6422. FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3410/3196 ($\nu_{a,s}(NH_2)_{amide}$ + $\nu(NH)_{amide}$ + $\nu(NH)_{amine}$ overlapped), 2139 ($\nu(C{\equiv}N)$), 1672 ($\nu(C{=}O)$/amide I + $(\nu(C{=}O)_{lactam}$ overlapped), 1622 ($\delta(NH_2)$/amide II), 1580 (corrin ring breathing mode), 1515 ($\delta(C{-}N{-}H)_{amine}$ + $\delta_{ip}(C{-}N{-}H)$/amide II + $\nu(C{=}C)$ overlapped), 1352 ($\nu(N{-}Ph)_{amine}$), 1271 ($\nu(C{-}O{-}C)_{xanthene}$), 1158/1147 ($\nu_{oop}(PO_2^-)$), 1070 ($\nu_{ip}(PO_2^-)$), 1001 ($\nu(P{-}O{-}C)$), 561 ($\delta(Co{-}C{\equiv}N)$), 492 ($\nu(Co{-}N)$), 412 ($\nu(Co{-}CN)$), 351/341 ($\nu(Co{-}N)$) cm^{-1}. ^1H NMR (400 MHz; CD_3OD; 298 K): δ

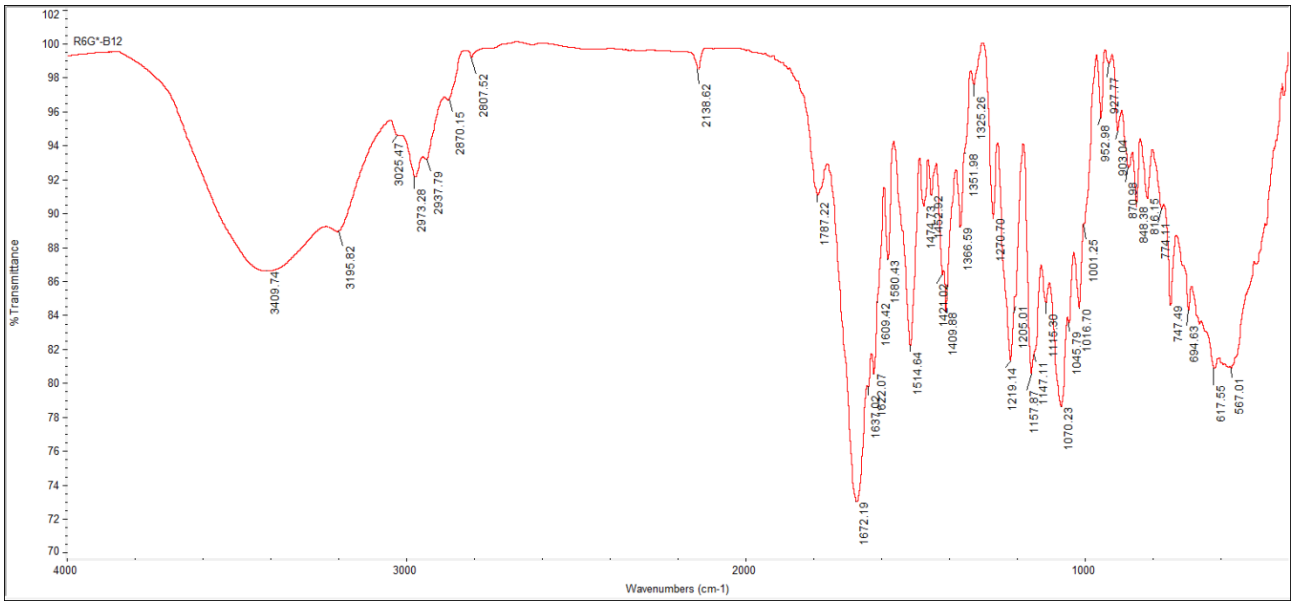
0.46 (3 H, s, $C^{20}\boldsymbol{H}_3$), 0.85-1.02 (2 H, m, $C^{a3,a5}\boldsymbol{H}_{ax}$), 1.04-1.16 (3 H, m, $C^{41}\boldsymbol{H}'$ + $C^{a2,a6}\boldsymbol{H}_{ax}$ overlapped), 1.17 (3 H, s, $C^{46}\boldsymbol{H}_3$), 1.22 (3 H, d, $C^{Pr3}\boldsymbol{H}_3$, $^3J_{H,H}$ = 6.3 Hz), 1.28-1.34 (1 H, m, $C^{42}\boldsymbol{H}'$), 1.30 (6 H, td, $C^{R6G}\boldsymbol{H}_{3(amine)}$, $^3J_{H,H}$ = 7.1 Hz; $^4J_{H,(N)H}$ = 2.4 Hz), 1.36 (3 H, s, $C^{25}\boldsymbol{H}_3$), 1.38 (3 H, s, $C^{54}\boldsymbol{H}_3$), 1.45 (3 H, s, $C^{47}\boldsymbol{H}_3$), 1.67-1.80 (4 H, m, $C^{42}\boldsymbol{H}''$ + $C^{55}\boldsymbol{H}'$ + $C^{a3,a5}\boldsymbol{H}_{eq}$ overlapped), 1.81-1.87 (2 H, m, $C^{48}\boldsymbol{H}_{2/diastereotopic}$), 1.88 (3 H, s, $C^{36}\boldsymbol{H}_3$), 1.89/1.90 (6 H, 2s, $C^{2,7R6G}C\boldsymbol{H}_3$, 1.93-1.98 (2 H, m, $C^{30}\boldsymbol{H}_{2/diastereotopic}$), 1.98-2.02 (1 H, m, $C^{41}\boldsymbol{H}''$), 2.01-2.10 (1 H, m, $C^{26}\boldsymbol{H}'$), 2.11-2.17 (1 H, m, $C^{56}\boldsymbol{H}'$), 2.20-2.24 (2 H, m, $C^{a2,a6}\boldsymbol{H}_{eq}$), 2.25 (3 H, s, $C^{B11}\boldsymbol{H}_3$), 2.28 (3 H, s, $C^{B10}\boldsymbol{H}_3$), 2.32-2.39 (2 H, m, $C^{37}\boldsymbol{H}_{2/diastereotopic}$), 2.44-2.54 (7 H, m, $C^{31}\boldsymbol{H}_{2/diastereotopic}$ + $C^{49}\boldsymbol{H}_{2/diastereotopic}$ + $C^{26}\boldsymbol{H}''$ + $C^{55}\boldsymbol{H}''$ + $C^{56}\boldsymbol{H}''$ overlapped), 2.56 (3 H, br s, $C^{35}\boldsymbol{H}_3$), 2.58 (3 H, br s, $C^{53}\boldsymbol{H}_3$), 2.60-2.65 (2 H, m, $C^{60}\boldsymbol{H}_{2/diastereotopic}$), 2.80-2.85 (1 H, m, $C^{18}\boldsymbol{H}$), 2.86-2.95 (2 H, m, $C^{Pr1}\boldsymbol{H}'$ + $C^{a1}\boldsymbol{H}$ overlapped), 3.22 (4 H, qd, $C^{R6G}\boldsymbol{H}_{2(amine)}$, $^3J_{H,H}$ = 7.1 Hz; $^3J_{H,(N)H}$ = 2.3 Hz), 3.25-3.30 (2 H, m, $C^{13}\boldsymbol{H}$ + $C^{a4}\boldsymbol{H}$ overlapped), 3.60-3.66 (2 H, m, $C^{8}\boldsymbol{H}$ + $C^{Pr1}\boldsymbol{H}''$ overlapped), 4.07-4.11 (2 H, m, $C^{R5}\boldsymbol{H}'$ + $C^{19}\boldsymbol{H}$ overlapped), 4.14-4.21 (2 H, m, $C^{R4}\boldsymbol{H}$ + $C^{R2}\boldsymbol{H}$ overlapped), 4.30-4.38 (1 H, m, $C^{Pr2}\boldsymbol{H}$), 4.52-4.54 (1 H, m, $C^{3}\boldsymbol{H}$), 4.58-4.65 (1 H, m, $C^{R5}\boldsymbol{H}''$), 4.74-4.79 (1 H, m, $C^{R3}\boldsymbol{H}$), 6.01 (1 H, s, $C^{10}\boldsymbol{H}$), 6.16 (3 H, br s, $C^{R1}\boldsymbol{H}$ + $C^{1,8R6G}\boldsymbol{H}$ overlapped), 6.35 (2 H, s, $C^{4,5R6G}\boldsymbol{H}$), 6.56 (1 H, s, $C^{B4}\boldsymbol{H}$), 6.94-7.00 (1 H, m, $C^{6'R6G}\boldsymbol{H}$), 7.12 (1 H, s, $C^{B2}\boldsymbol{H}$), 7.20 (1 H, s, $C^{B7}\boldsymbol{H}$), 7.48-7.53 (2 H, m, $C^{4'R6G}\boldsymbol{H}$ + $C^{5'R6G}\boldsymbol{H}$ overlapped), 7.83 (1 H, dd, $C^{3'R6G}\boldsymbol{H}$, $^3J_{H,H}$ = 6.0 Hz, $^4J_{H,H}$ = 2.6 Hz) ppm [28]. $^{13}C\{^1H\}$ NMR (100 MHz; CD_3OD; 298 K): δ14.7 ($\boldsymbol{C}^{R6G}H_{3(amine)}$), 16.2 ($\boldsymbol{C}^{53}H_3$), 16.4 ($\boldsymbol{C}^{35}H_3$), 17.1 ($C^{2,7R6G}\boldsymbol{C}H_3$), 17.2 ($\boldsymbol{C}^{25}H_3$), 17.5 ($\boldsymbol{C}^{54}H_3$), 19.9 ($\boldsymbol{C}^{36}H_3$), 20.0 ($\boldsymbol{C}^{Pr3}H_3$), 20.1 ($\boldsymbol{C}^{B11}H_3$), 20.2 ($\boldsymbol{C}^{47}H_3$), 20.3 ($\boldsymbol{C}^{20}H_3$), 20.9 ($\boldsymbol{C}^{B10}H_3$), 27.2 ($\boldsymbol{C}^{41}H_2$), 27.4 ($\boldsymbol{C}^{30}H_2$), 29.0 ($\boldsymbol{C}^{a2,a6}H_2$), 29.5 ($\boldsymbol{C}^{48}H_2$), 32.0 ($\boldsymbol{C}^{46}H_3$), 32.3 ($\boldsymbol{C}^{60}H_2$), 32.4 ($\boldsymbol{C}^{56}H_2$), 32.6 ($\boldsymbol{C}^{55}H_2$), 33.2 ($\boldsymbol{C}^{42}H_2$), 33.5 ($\boldsymbol{C}^{a3,a5}H_2$), 35.1 ($\boldsymbol{C}^{49}H_2$), 36.2 ($\boldsymbol{C}^{31}H_2$), 39.2 ($\boldsymbol{C}^{R6G}H_{2(amine)}$), 40.1 ($\boldsymbol{C}^{18}$H), 43.0 ($\boldsymbol{C}^{37}H_2$), 43.9 ($\boldsymbol{C}^{26}H_2$), 46.6 ($\boldsymbol{C}^{Pr1}H_2$), ~49 ($\boldsymbol{C}^{2}$ + $\boldsymbol{C}^{12}$ overlapped with the residual peak of CD_3OD), 50.4 ($\boldsymbol{C}^{a4}$H), 52.6 ($\boldsymbol{C}^{7}$), 54.3 ($\boldsymbol{C}^{a1}$H), 54.4 ($\boldsymbol{C}^{13}$H), 56.0 ($\boldsymbol{C}^{8}$H), 57.5 ($\boldsymbol{C}^{3}$H), 60.3 ($\boldsymbol{C}^{17}$), 64.0 ($\boldsymbol{C}^{R5}H_2$), 67.9 ($\boldsymbol{C}^{9}$), 70.5 ($\boldsymbol{C}^{R2}$H), 72.5 ($\boldsymbol{C}^{Pr2}$H), 75.2 ($\boldsymbol{C}^{R3}$H), 76.3 ($\boldsymbol{C}^{19}$H), 80.8 ($\boldsymbol{C}^{R4}$H), 86.4 ($\boldsymbol{C}^{1}$), 88.1 ($\boldsymbol{C}^{R1}$H), 95.3 ($\boldsymbol{C}^{10}$H), 97.4 ($\boldsymbol{C}^{4,5R6G}$H), 105.2 ($\boldsymbol{C}^{15}$), 105.9 ($\boldsymbol{C}^{12,13R6G}$), 108.2 ($\boldsymbol{C}^{5}$), 112.3 ($\boldsymbol{C}^{B7}$H), 112.7 ($\boldsymbol{C}$N), 117.9 ($\boldsymbol{C}^{B4}$H), 119.7 ($\boldsymbol{C}^{2,7R6G}$), 123.2 ($\boldsymbol{C}^{3'R6G}$H), 125.0 ($\boldsymbol{C}^{6'R6G}$H), 129.4 ($\boldsymbol{C}^{4'R6G}$H), 129.6 ($\boldsymbol{C}^{1,8R6G}$H), 131.3 ($\boldsymbol{C}^{B8}$), 132.6 ($\boldsymbol{C}^{2'R6G}$), 133.3 ($\boldsymbol{C}^{5'R6G}$H), 133.8 ($\boldsymbol{C}^{B5}$), 135.7 ($\boldsymbol{C}^{B6}$), 138.2 ($\boldsymbol{C}^{B9}$), 143.3 ($\boldsymbol{C}^{B2}$H), 149.4 ($\boldsymbol{C}^{3,6R6G}$), 153.3 ($\boldsymbol{C}^{11,14R6G}$), 154.9 ($\boldsymbol{C}^{1'}$), 166.9 ($\boldsymbol{C}^{14}$), 167.1 ($\boldsymbol{C}^{6}$), 169.3 ($\boldsymbol{C}^{R6G}$=O$_{(lactam)}$), 174.3 ($\boldsymbol{C}^{57}$), 174.6 ($\boldsymbol{C}^{38}$), 175.3 ($\boldsymbol{C}^{61}$), 175.6 ($\boldsymbol{C}^{27}$), 175.7 ($\boldsymbol{C}^{43}$), 176.5 ($\boldsymbol{C}^{32}$), 177.2 ($\boldsymbol{C}^{9}$), 177.4 ($\boldsymbol{C}^{R6G}$=O$_{(amide)}$), 177.5 ($\boldsymbol{C}^{50}$), 177.6 ($\boldsymbol{C}^{11}$), 180.1 ($\boldsymbol{C}^{16}$), 181.6 ($\boldsymbol{C}^{4}$) ppm [28]. $^{31}P\{^1H\}$ NMR (162 MHz; CD_3OD; 298 K): δ-0.2 ($\boldsymbol{P}O_4^-$) ppm.

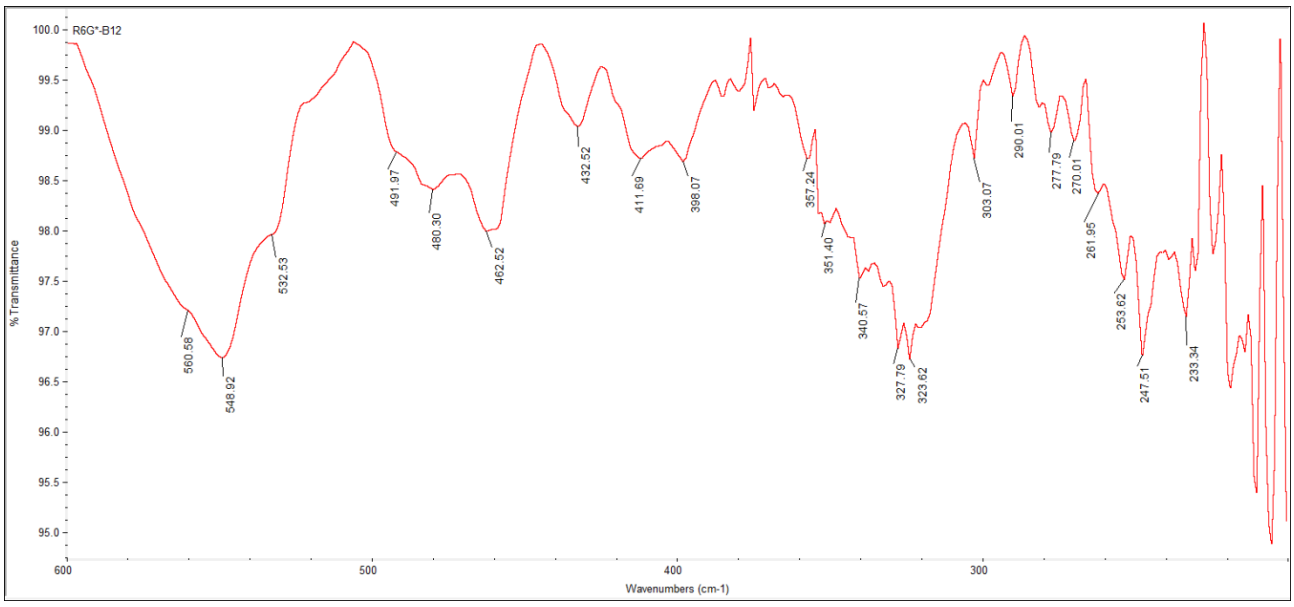
Spectrum Source	Fragmentor Voltage	Collision Energy	Ionization Mode
Peak (13) in "+ TIC Scan"	120	0	ESI

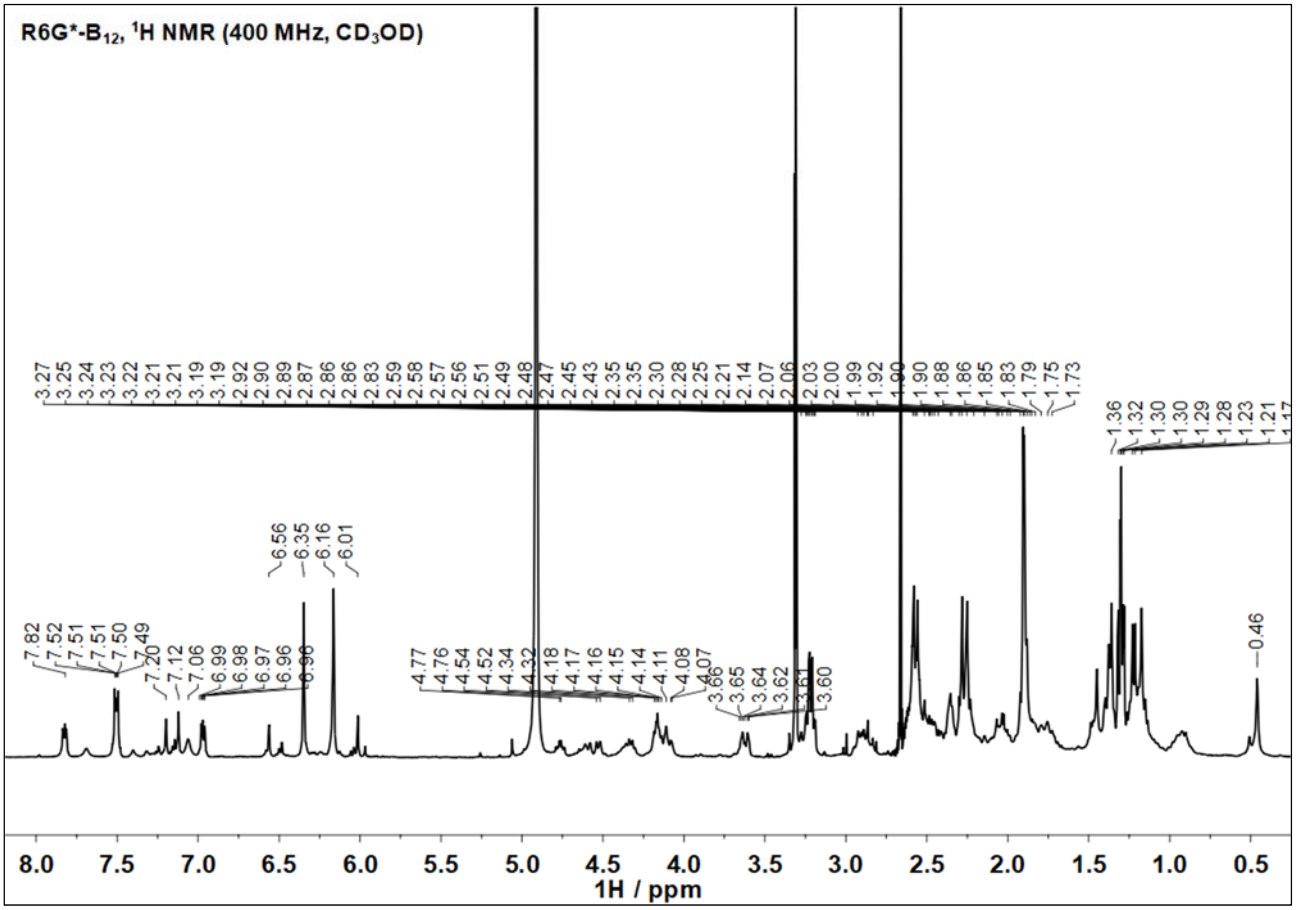


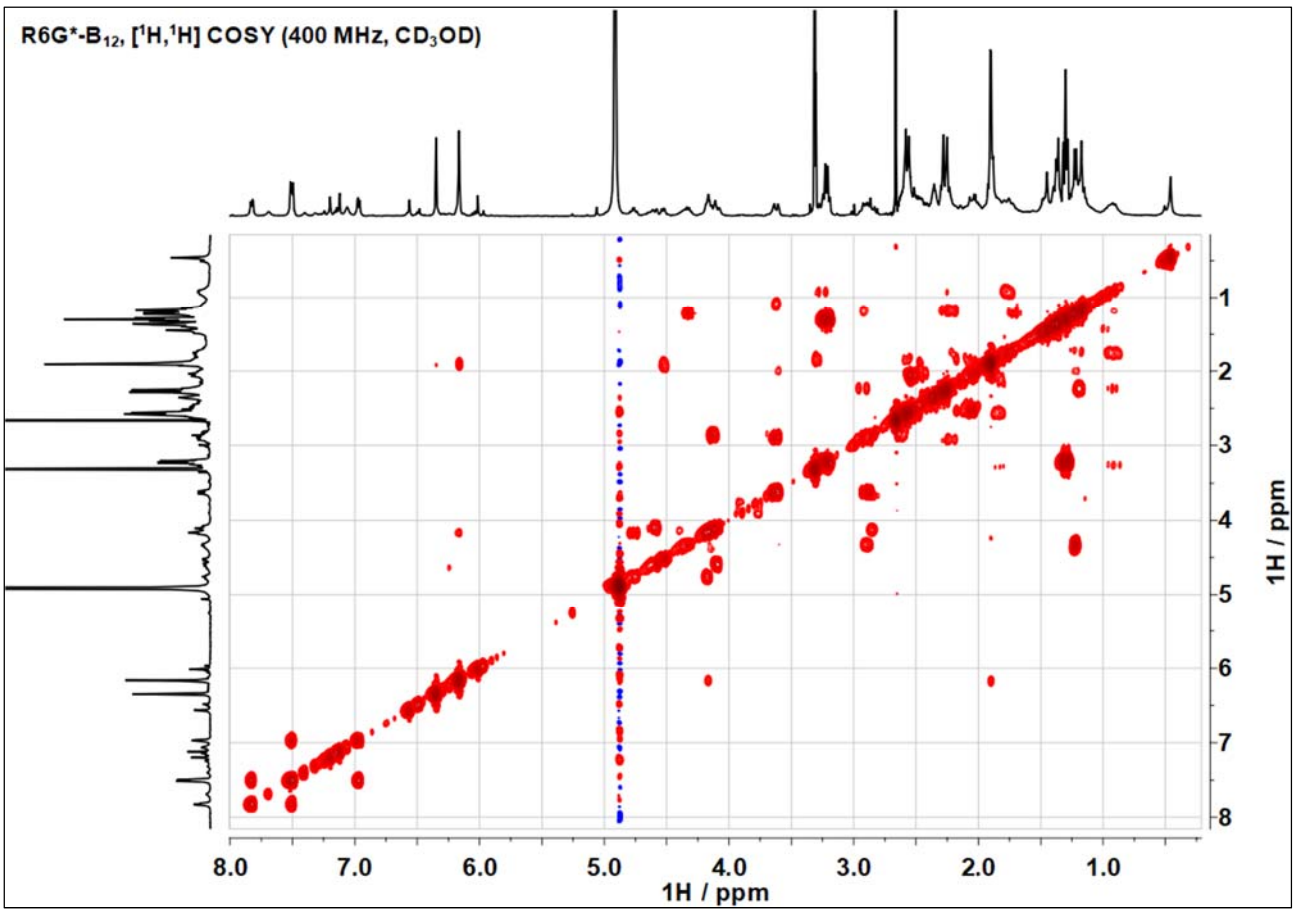
Peak List

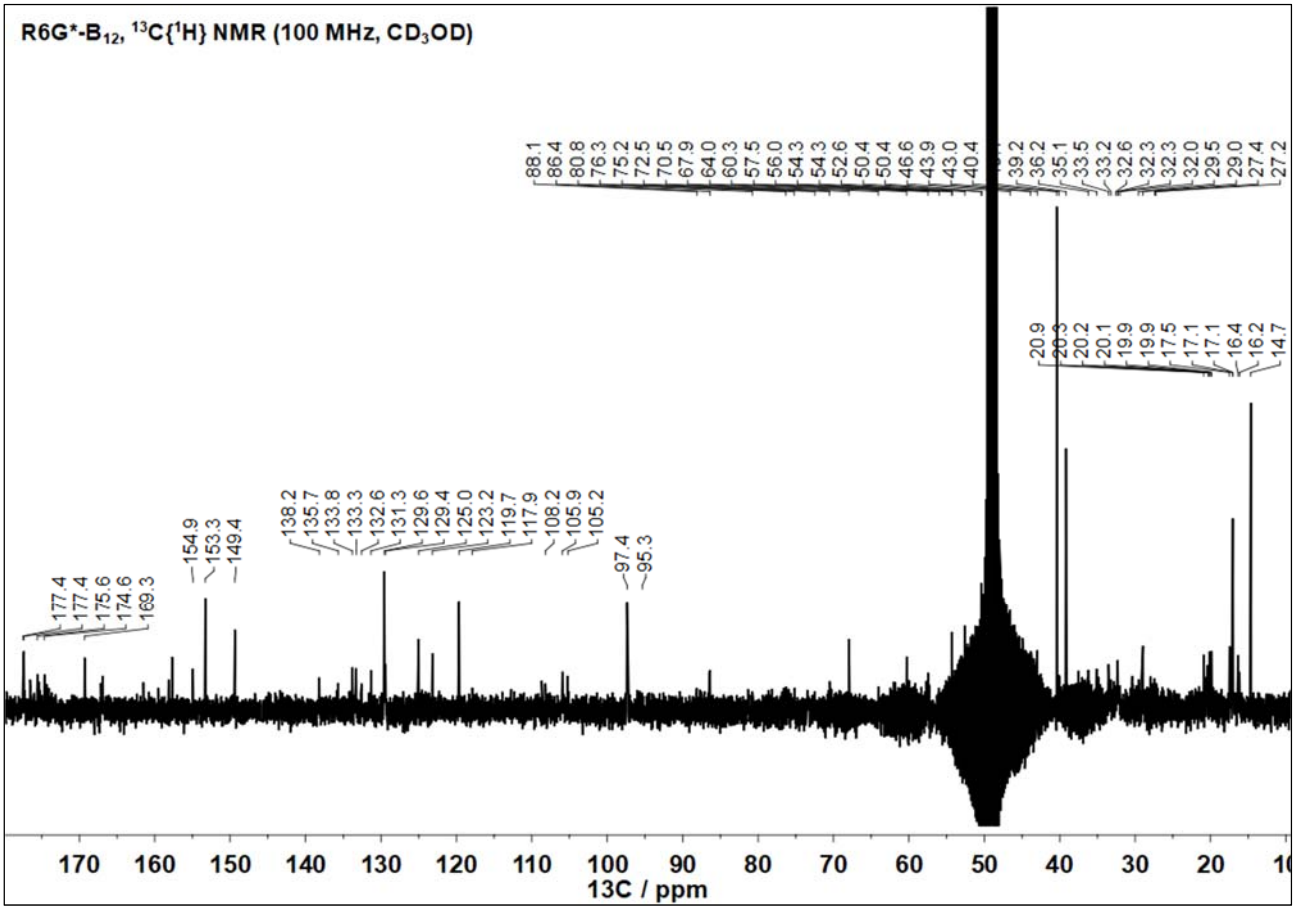
m/z	z	Abund
631.3078	1	357069.56
631.6422	1	401869.97
631.9769	1	231660
632.3112	1	91408.28
649.2822	1	71697.4
649.6167	1	73251.05
946.4569	2	374106.31
946.9586	2	425498.16
947.4602	2	247753.41
947.9619	2	97754.21






R6G*-B12, 1H NMR (400 MHz, CD3OD)
1H / ppm


R6G*-B12, [1H,1H] COSY (400 MHz, CD3OD)
1H / ppm
1H / ppm


R6G*-B12, 13C{1H} NMR (100 MHz, CD3OD)
88.1
86.4
80.8
76.3
75.2
72.5
70.5
67.9
64.0
60.3
57.5
56.0
54.3
54.3
52.6
50.4
50.4
46.6
43.9
43.0
40.4
39.2
36.2
35.1
33.5
33.2
32.6
32.3
32.3
32.0
29.5
29.0
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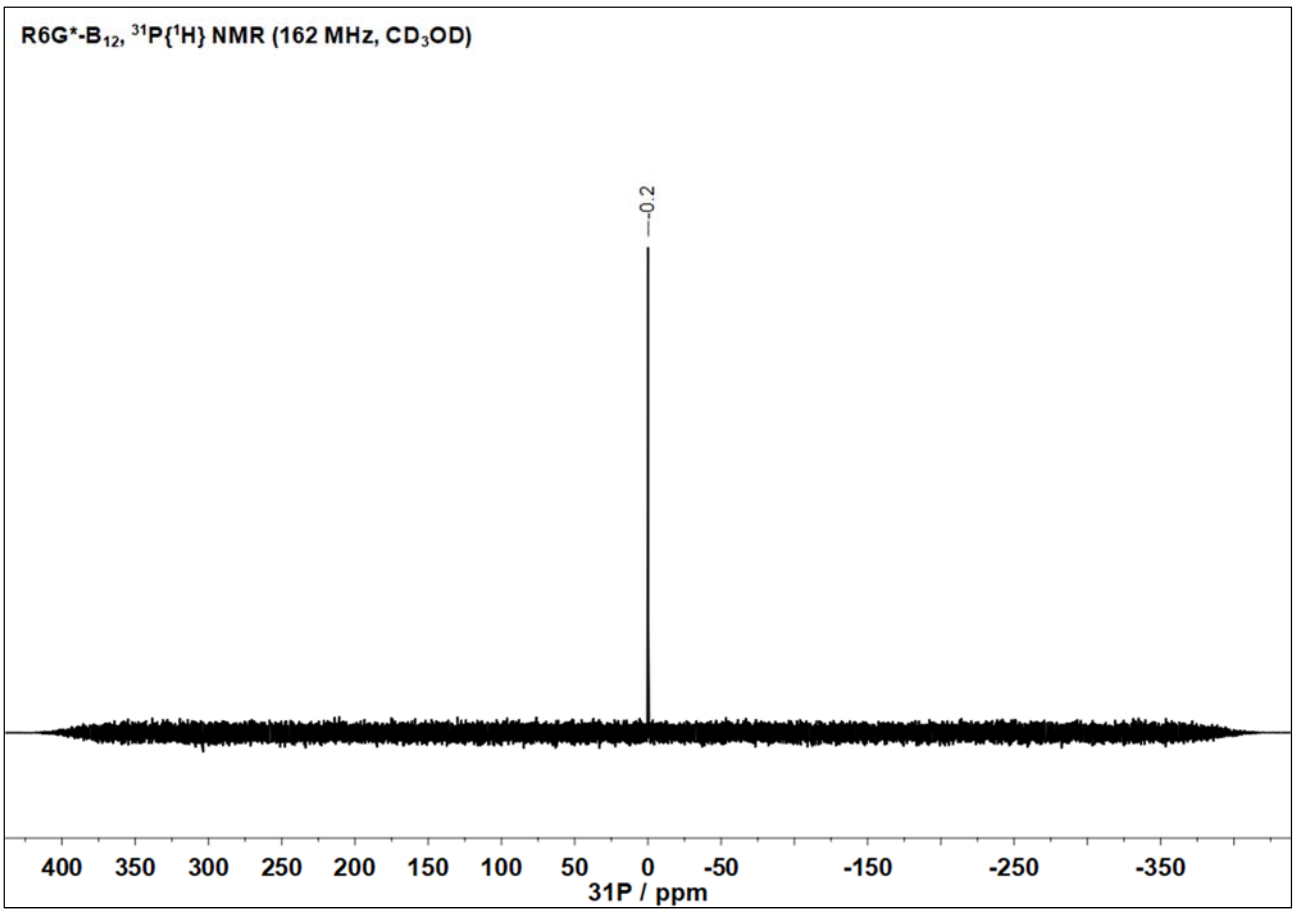
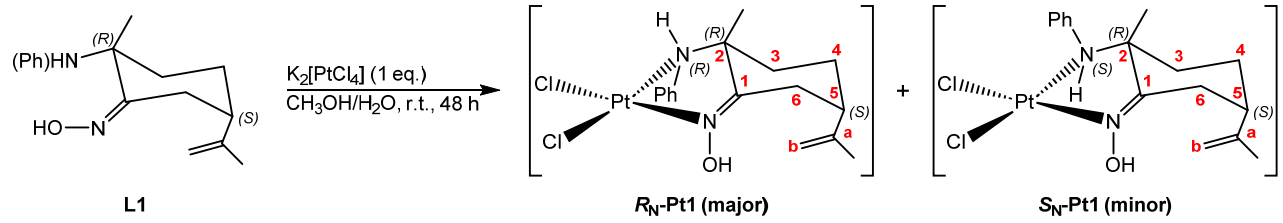
19.9
17.5
17.1
17.1
16.4
16.2
14.7
177.4
177.4
175.6
174.6
169.3
154.9
153.3
149.4
138.2
135.7
133.8
133.3
132.6
131.3
129.6
129.4
125.0
123.2
119.7
117.9
108.2
105.9
105.2
97.4
95.3
170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20
13C / ppm


R6G*-B12, 31P{1H} NMR (162 MHz, CD3OD)
-0.2
400 350 300 250 200 150 100 50 0 -50 -150 -250 -350
31P / ppm

Synthesis of the platinum(II) precursors

Dichloro[(2*R*,5*S*)-2-methyl-5-(1-methylethenyl)-2-(phenylamino-κ*N*)-cyclohexanone oxime-κ*N*(OH))]platinum(II) (Pt1)

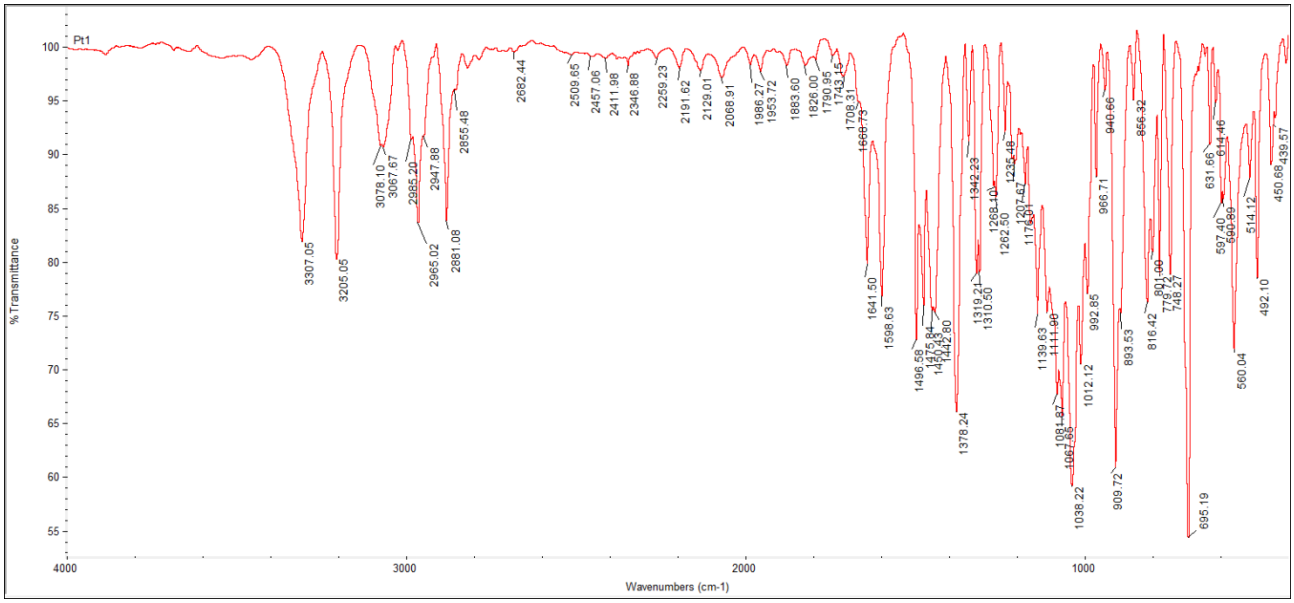
A water solution (2 mL) of $K_2[PtCl_4]$ (0.067 g, 0.16 mmol) was added dropwise to a methanol solution of **L1** (0.042 g, 0.16 mmol) and the mixture was stirred at room temperature for 48 h. The solvent was subsequently evaporated to dryness and the crude solid extracted with DCM (50 mL). The extract was then evaporated to dryness and dried under vacuum over P_2O_5, yielding a dark brown solid identified as a mixture of R_N and S_N epimers of the title compound. The pure major epimer (***R*$_N$-Pt1**) could be isolated by recrystallization from chloroform (MM($C_{16}H_{22}Cl_2N_2OPt$) = 524.35 g mol^{-1}, 0.075 g, 89% yield). However, upon dissolution in various solvents, an epimerization process involving the coordinated amino moiety occurred, leading to an R_N:S_N epimers ratio of *ca.* 2.5:1 in both $CDCl_3$ (within 24 h) and CD_3OD (within a few minutes), as estimated by ^{1}H NMR spectroscopy. Epimerization was also evaluated in both solvents at increasing temperatures (up to 70°C), which did not significantly affect the final ratio of the ***R*$_N$-Pt1** and ***S*$_N$-Pt1** epimers in solution.

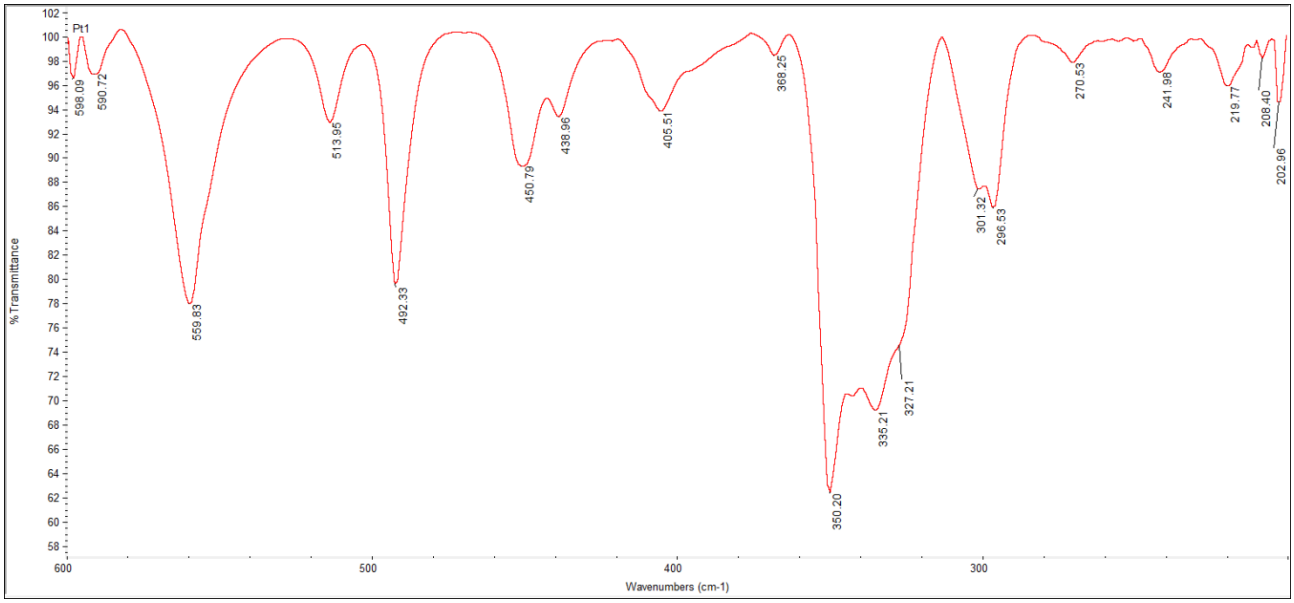


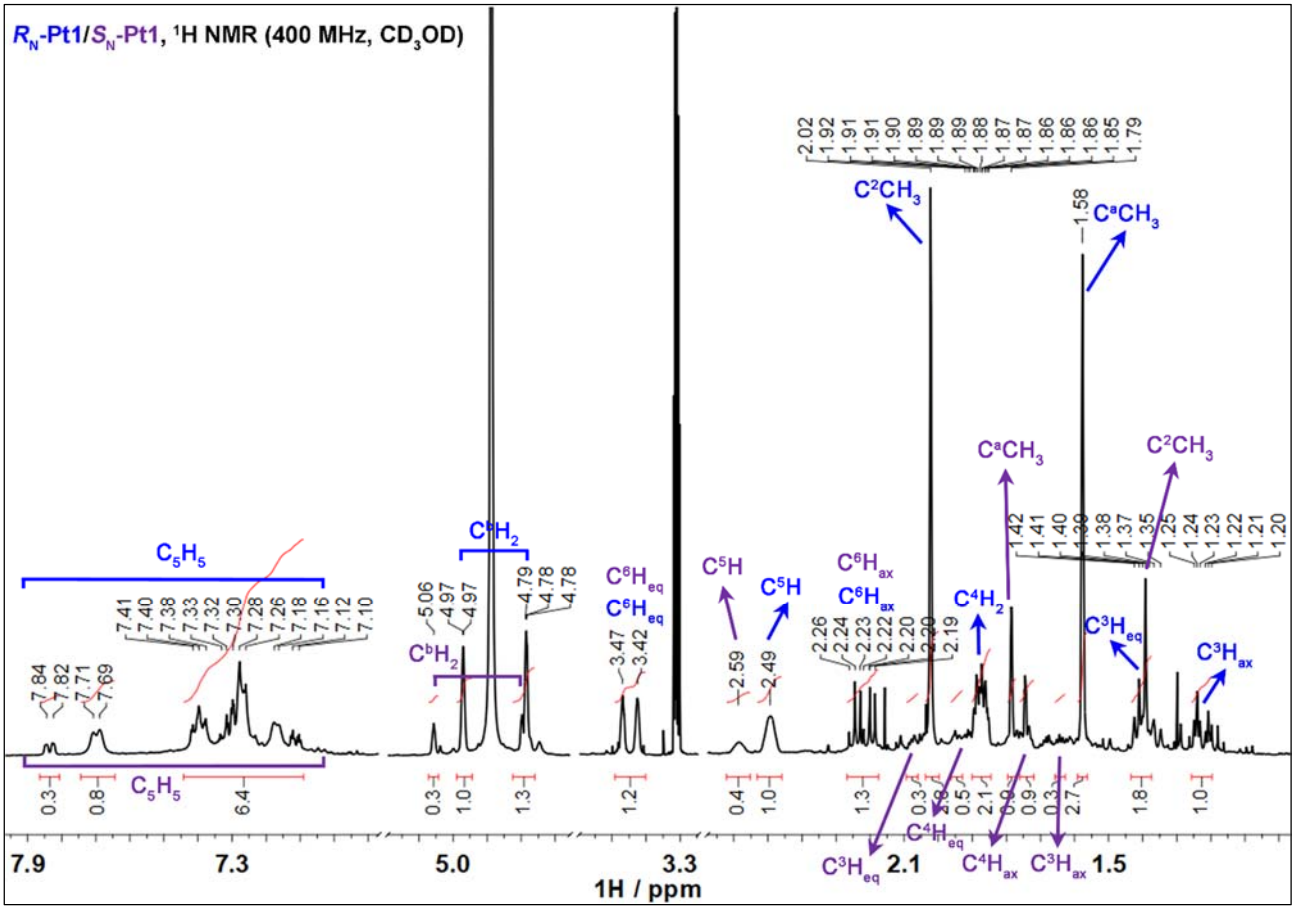
L1 **R_N-Pt1 (major)** **S_N-Pt1 (minor)**

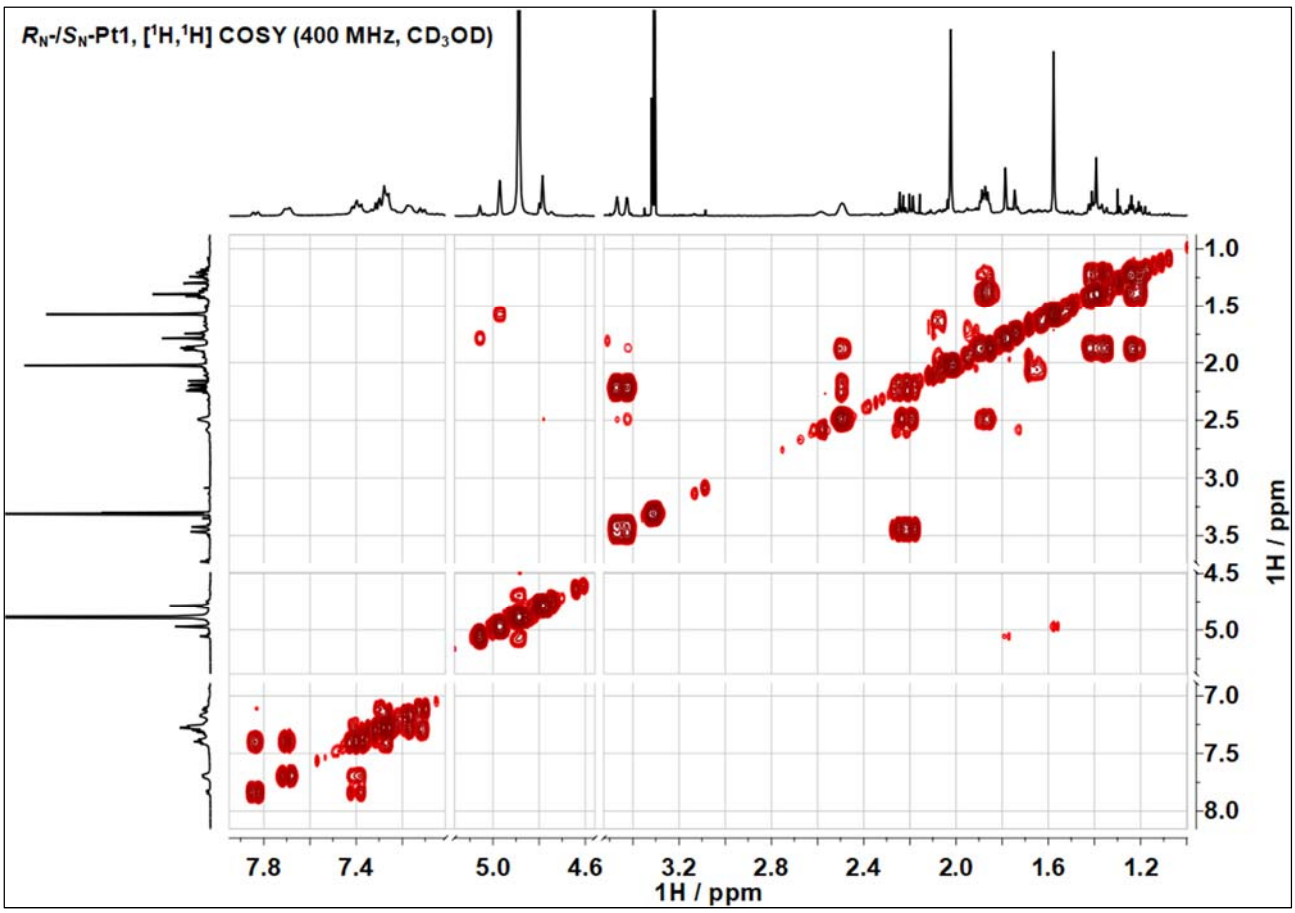
Anal. (%) calcd for $C_{16}H_{22}Cl_2N_2OPt{\cdot}2H_2O$: C, 34.29; H, 4.68; N, 5.00; found: C, 34.57; H, 4.08; N, 5.07. FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3307 (ν(OH)$_{oxime}$), 3205 (ν(NH)$_{amine}$), 1642 (ν(C=C)$_{alkene}$), 1599 (ν(C=N)$_{oxime}$), 1236 (ν(Ph–N)), 910 (ν(N–OH)$_{oxime}$), 451 (ν(Pt–NOH)), 350 (ν(Pt–NPh)), 335/327 ($\nu_{as,s}$(PtCl$_2$)) cm^{-1} [40,48,49]. ^{1}H NMR (400 MHz; CD_3OD; 298 K): δ 1.22 (1 H, m, C^3***H***$_{ax}$ R_N), 1.39 (1.2 H, s, C^{2}C***H***$_3$ S_N), 1.35-1.42 (1 H, m, C^3***H***$_{eq}$ R_N), 1.58 (3 H, br t, C^{a}C***H***$_3$ R_N), 1.61-1.70 (0.4 H, m, C^3***H***$_{ax}$ S_N), 1.73-1.77 (0.4 H, m, C^4***H***$_{ax}$ S_N), 1.79 (1.2 H, br t, C^{a}C***H***$_3$ S_N), 1.85-1.90 (2 H, m, C^4***H***$_{2/diastereotopic}$ R_N), 1.91-1.96 (0.4 H, m, C^4***H***$_{eq}$ S_N), 2.02 (3 H, br d, C^{2}C***H***$_3$ R_N), 2.04-2.12 (0.4 H, m, C^3***H***$_{eq}$ S_N), 2.19-2.26 (1.4 H, m, C^6***H***$_{ax}$ R_N + S_N overlapped), 2.49 (1 H, br s, C^5***H*** R_N), 2.59 (0.4 H, br s, C^5***H*** S_N), 3.45 (1.4 H, m, C^6***H***$_{eq}$ R_N + S_N overlapped), 4.85 (2 H, br dd, C^b***H***$_{2/diastereotopic}$ R_N), 4.93 (0.8 H, br dd, C^b***H***$_{2/diastereotopic}$ S_N), 7.10-7.84 (7 H, m, C$_5$***H***$_{5/Ph}$ R_N + S_N overlapped) ppm. ^{13}C{^{1}H} NMR (100 MHz; CD_3OD; 298 K): δ 21.7 (C^a***C***H$_3$ R_N), 22.2 (C^a***C***H$_3$ S_N), 23.16 (C^2***C***H$_3$ S_N), 25.1 (***C***^{4}H$_2$ S_N), 25.7 (***C***^{4}H$_2$ R_N), 26.7 (C^2***C***H$_3$ R_N), 29.9 (***C***^{6}H$_2$ S_N), 30.0 (***C***^{6}H$_2$ R_N), 31.5 (***C***^{3}H$_2$ R_N), 32.4 (***C***^{3}H$_2$ S_N), 39.3 (***C***^{5}H R_N), 40.1 (***C***^{5}H S_N), 75.1 (***C***^{2} S_N), 75.7 (***C***^{2} R_N), 112.8 (***C***^{b}H$_2$ S_N), 112.9 (***C***^{b}H$_2$ R_N), 124.3/126.1/127.5/128.6/128.7/128.9/129.2/129.8/131.1/132.5 (***C***$_5$H$_{5/Ph}$ R_N + S_N overlapped), 143.6 (*i*-***C*** S_N), 143.7 (*i*-***C*** R_N), 146.3 (***C***^{a} S_N), 147.0 (***C***^{a} R_N), 170.6 (***C***^{1} S_N), 171.6 (***C***^{1} R_N) ppm

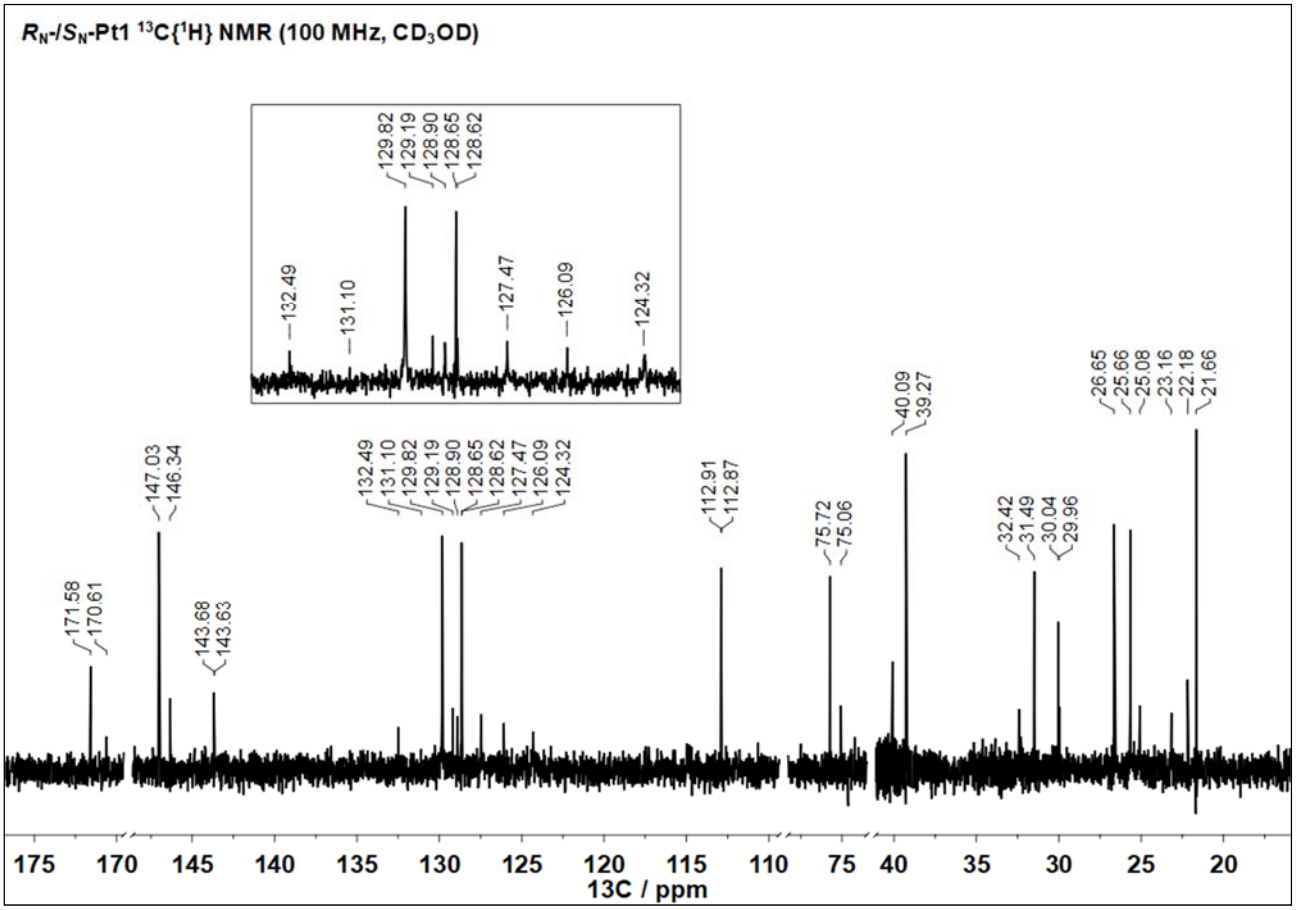
[48,102]. [^{1}H,^{13}C] HMBC (100.6 MHz; $CDCl_3$; 293 K): δ 75.1 (***C***^{2} S_N), 75.7 (***C***^{2} R_N), 170.6 (***C***^{1} S_N), 171.6 (***C***^{1} R_N) ppm. [^{1}H,^{15}N] HMBC (40.5 MHz; $CDCl_3$; 293 K): δ 51.2 (C^{2}***N***(Ph)H R_N, S_N not detected), 171.6 (C^{1}***N***OH R_N, S_N not detected) ppm.

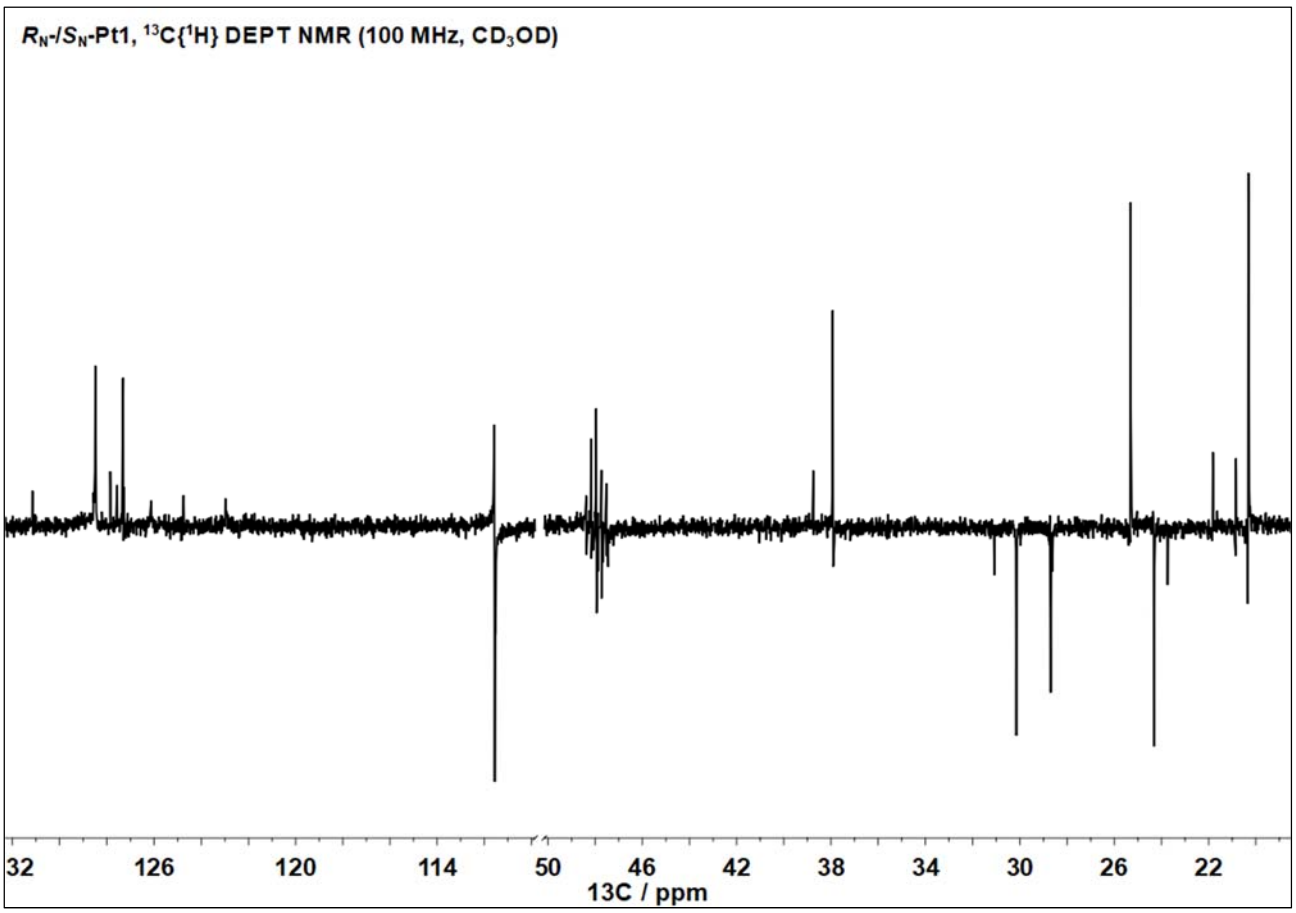


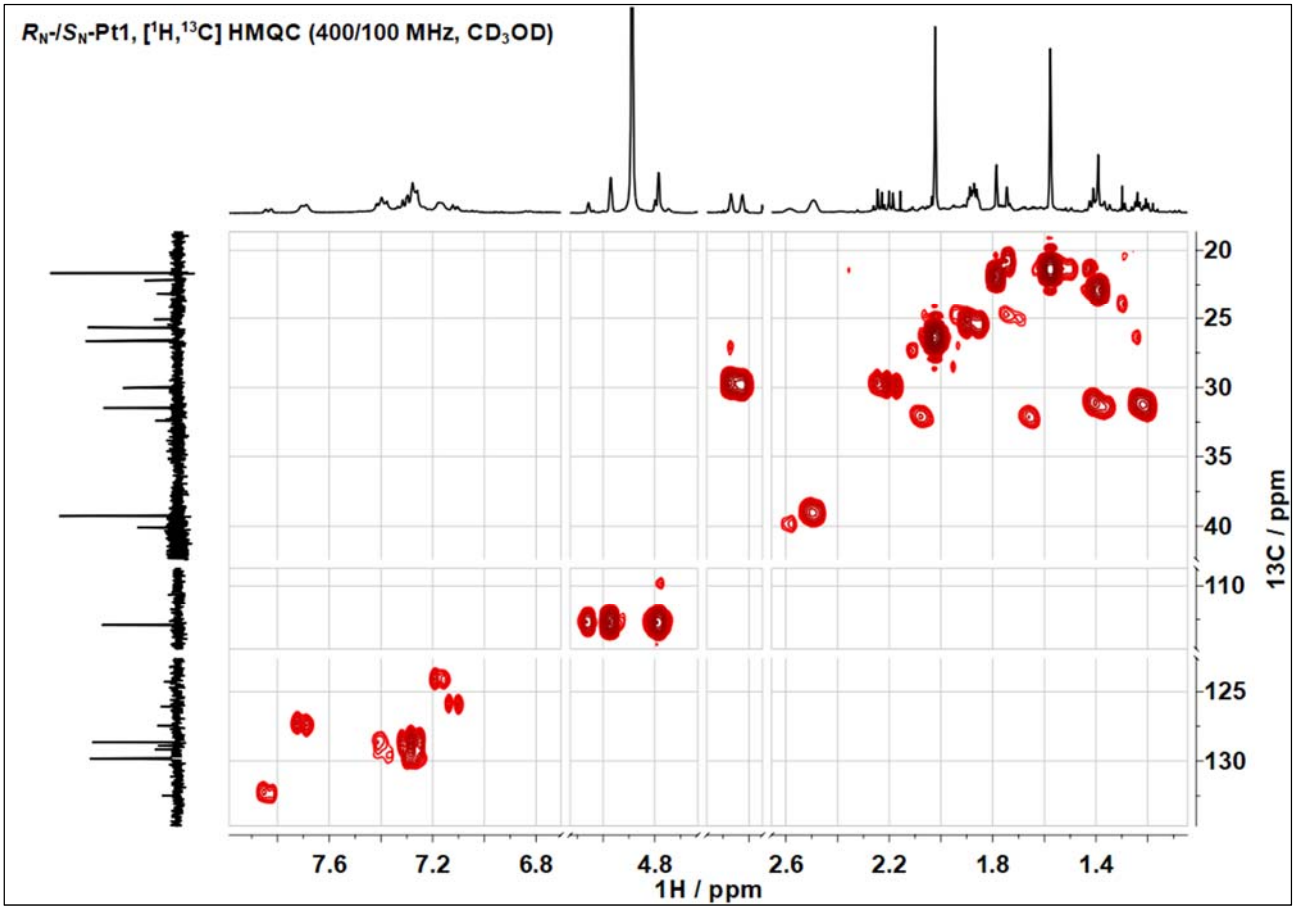



R_N-Pt1/S_N-Pt1, ^{1}H NMR (400 MHz, CD_3OD)
C_5H_5
C^bH_2
C^6H_{eq}
C^6H_{eq}
C^5H
C^5H
C^6H_{ax}
C^6H_{ax}
C^4H_2
C^aCH_3
C^2CH_3
C^2CH_3
C^aCH_3
C^3H_{eq}
C^3H_{ax}
C^4H_{eq}
C^3H_{eq}
C^4H_{ax}
C^3H_{ax}
1H / ppm


R_N-/S_N-Pt1, [^{1}H,^{1}H] COSY (400 MHz, CD_3OD)
1H / ppm
1H / ppm

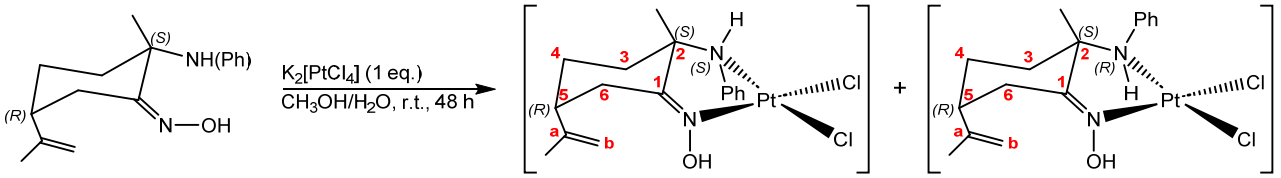

R_N-/S_N-Pt1 $^{13}C\{^1H\}$ NMR (100 MHz, CD_3OD)
132.49
131.10
129.82
129.19
128.90
128.65
128.62
127.47
126.09
124.32
171.58
170.61
147.03
146.34
143.68
143.63
112.91
112.87
75.72
75.06
40.09
39.27
32.42
31.49
30.04
29.96
26.65
25.66
25.08
23.16
22.18
21.66
175 170 145 140 135 130 125 120 115 110 75 40 35 30 25 20
13C / ppm


R_N-/S_N-Pt1, $^{13}C\{^1H\}$ DEPT NMR (100 MHz, CD_3OD)
32 126 120 114 50 46 42 38 34 30 26 22
13C / ppm

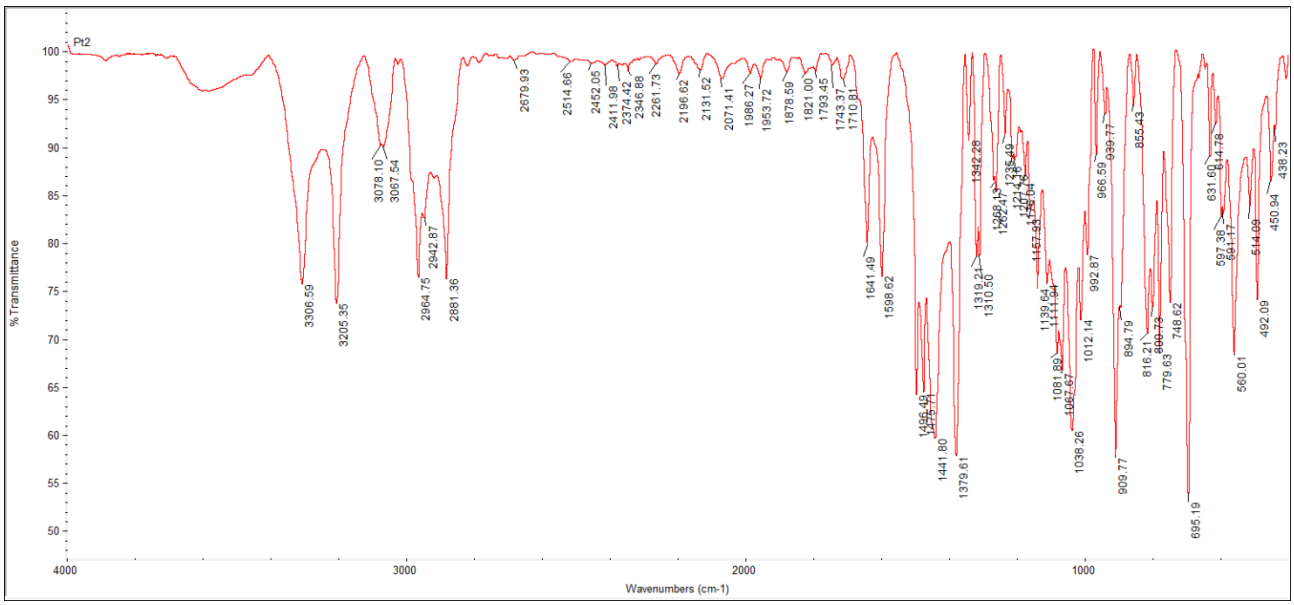


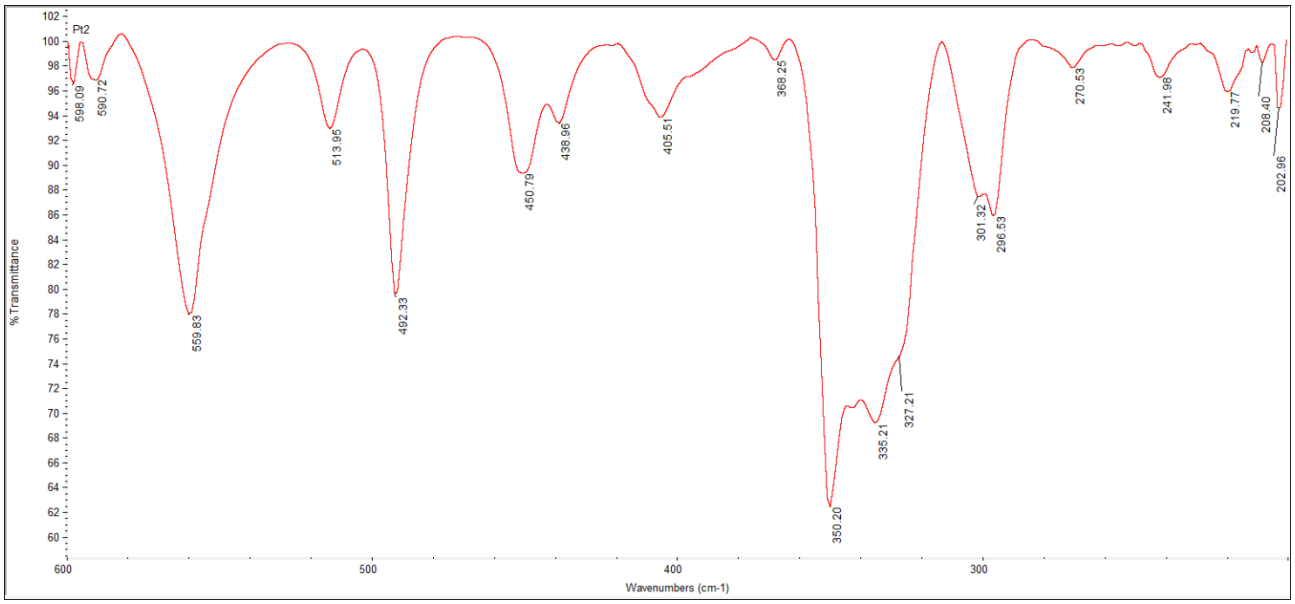
Dichloro[(2*S*,5*R*)-2-methyl-5-(1-methylethenyl)-2-(phenylamino-κ*N*)-cyclohexanone oxime-κ*N*(OH))]platinum(II) (Pt2)

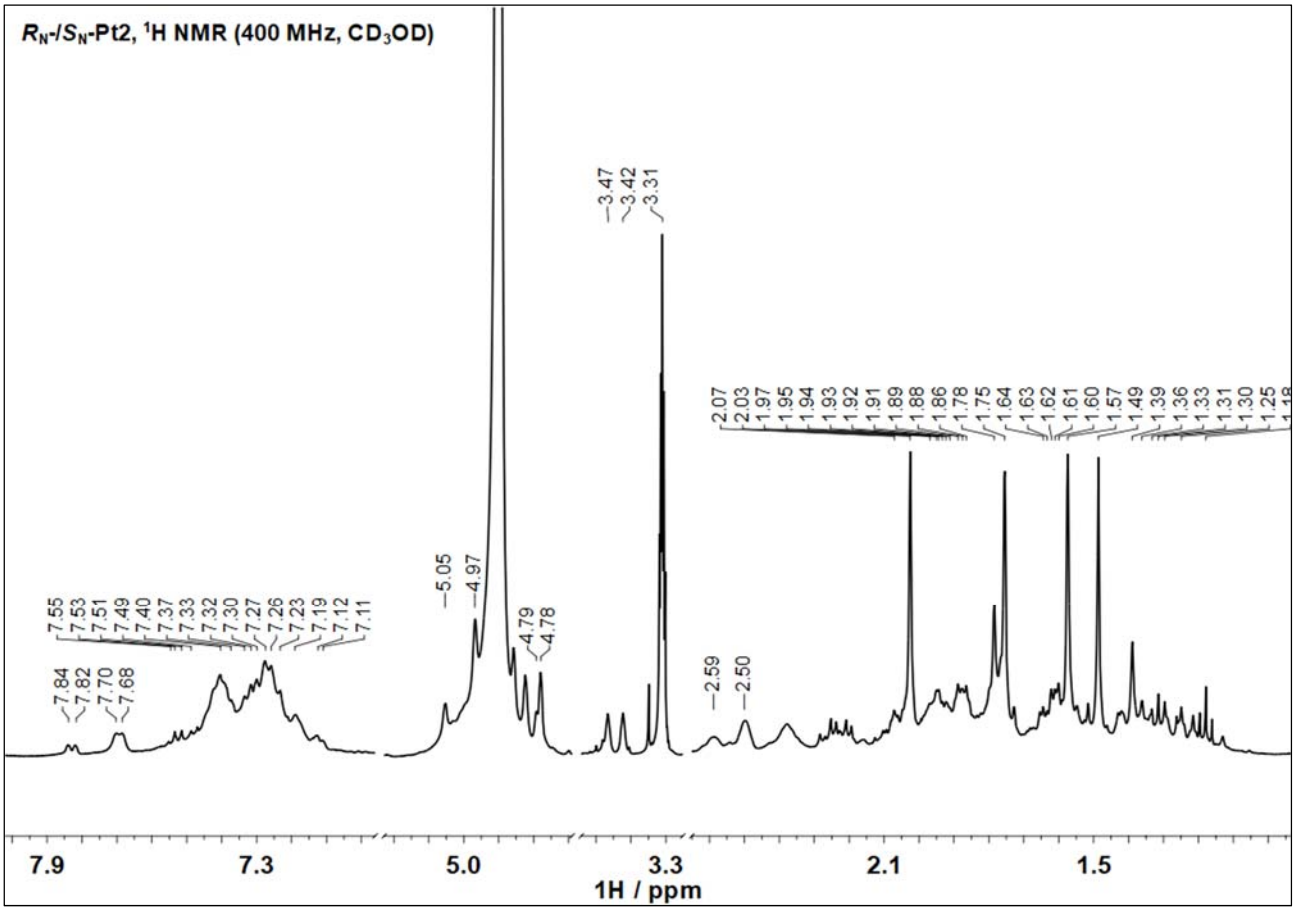
A water solution (2 mL) of $K_2[PtCl_4]$ (0.067 g, 0.16 mmol) was added dropwise to a methanol solution of **L2** (0.042 g, 0.16 mmol) and the mixture was stirred at room temperature for 48 h. The solvent was subsequently evaporated to dryness and the crude solid extracted with DCM (50 mL). The extract was then evaporated to dryness and dried under vacuum over P_2O_5, yielding a dark brown solid identified as a mixture of S_N and R_N epimers of the title compound. The pure major epimer (**S_N-Pt2**) could be isolated by recrystallization from chloroform (MM($C_{16}H_{22}Cl_2N_2OPt$) = 524.35 g mol^{-1}, 0.079 g, 94% yield). However, upon dissolution in various solvents, an epimerization process involving the coordinated amino moiety occurred, leading to an R_N:S_N epimers ratio of *ca.* 2.5:1 in both $CDCl_3$ (within 24 h) and CD_3OD (within a few minutes), as estimated by ^{1}H NMR spectroscopy. Epimerization was also evaluated in both solvents at increasing temperatures (up to 70°C), which did not significantly affect the final ratio of the **S_N-Pt2** and **R_N-Pt2** epimers in solution with respect to dissolution at room temperature.

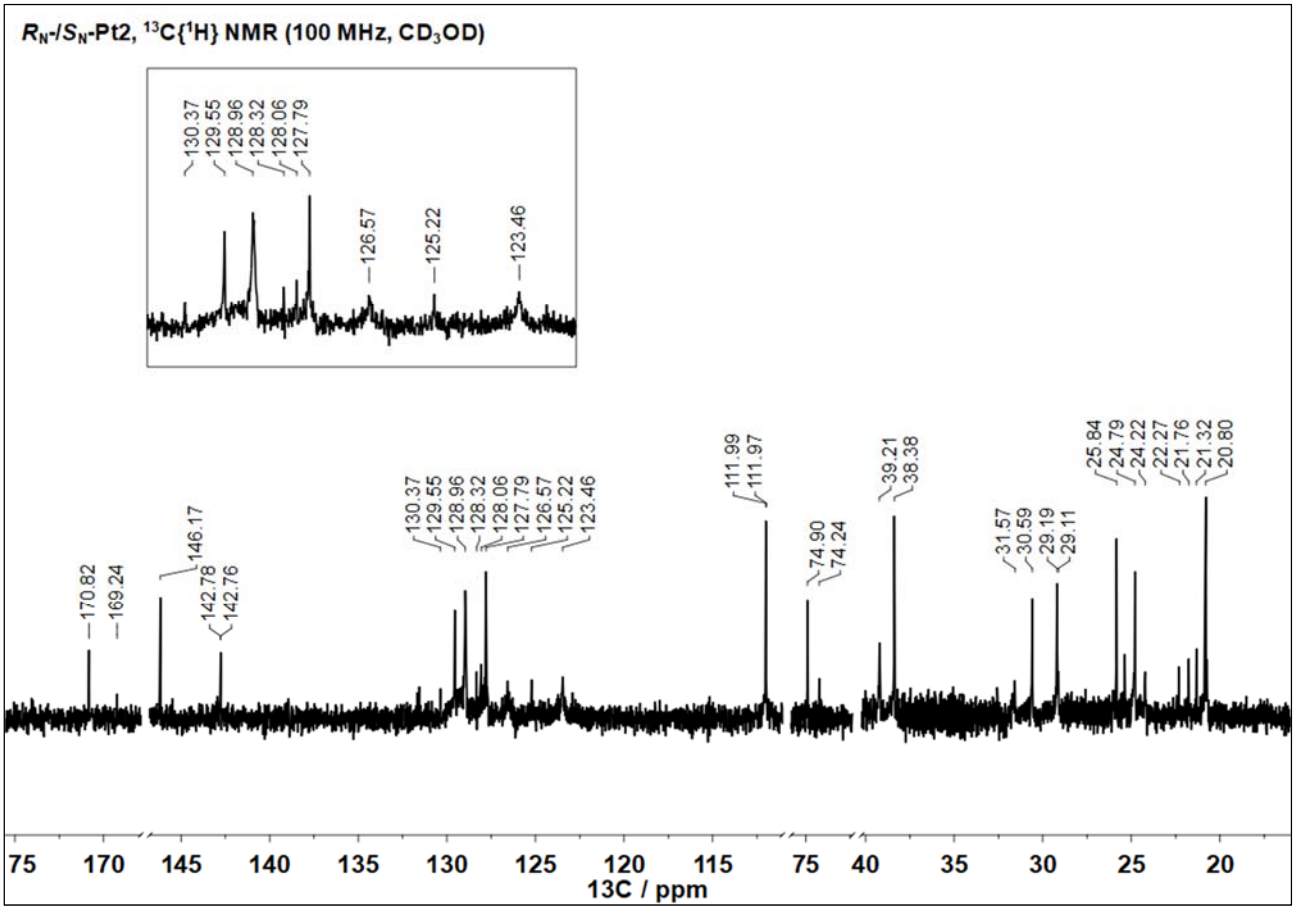


Anal. (%) calcd for $C_{16}H_{22}Cl_2N_2OPt{\cdot}2H_2O$: C, 34.29; H, 4.68; N, 5.00; found: C, 34.65; H, 4.04; N, 5.03. FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3307 (ν(OH)$_{oxime}$), 3205 (ν(NH)$_{amine}$), 1642 (ν(C=C)$_{alkene}$), 1599 (ν(C=N)$_{oxime}$), 1236 (ν(Ph–N)), 910 (ν(N–OH)$_{oxime}$), 451 (ν(Pt–NOH)), 350 (ν(Pt–NPh)), 335/327 ($\nu_{as,s}$(PtCl$_2$)) cm^{-1}[40,48,49]. ^{1}H NMR (400 MHz; CD$_3$OD; 298 K): δ 1.16-1.29 (1 H, m, C^3***H***$_{ax}$ *S*$_N$), 1.39 (0.9 H, s, C^{2}C***H***$_3$ *R*$_N$), 1.30-1.43 (1 H, m, C^3***H***$_{eq}$ *S*$_N$), 1.57 (3 H, br t, C^{a}C***H***$_3$ *S*$_N$), 1.59-1.65 (0.3 H, m, C^3***H***$_{ax}$ *R*$_N$), 1.73-1.76 (0.3 H, m, C^4***H***$_{ax}$ *R*$_N$), 1.78 (0.9 H, br t, C^{a}C***H***$_3$ *R*$_N$), 1.86-1.95 (2 H, m, C^4***H***$_{2/diastereotopic}$ *S*$_N$), 1.97-2.01 (0.3 H, m, C^4***H***$_{eq}$ *R*$_N$), 2.03 (3 H, br d, C^{2}C***H***$_3$ *S*$_N$), 2.04-2.10 (0.3 H, m, C^3***H***$_{eq}$ *R*$_N$), 2.19-2.28 (1.3 H, m, C^6***H***$_{ax}$ *S*$_N$ + *R*$_N$ overlapped), 2.50 (1 H, br s, C^5***H*** *S*$_N$), 2.59 (0.4 H, br s, C^5***H*** *R*$_N$), 3.44 (1.3 H, m, C^6***H***$_{eq}$ *S*$_N$ + *R*$_N$ overlapped), 4.88 (2 H, br dd, C^b***H***$_{2/diastereotopic}$ *S*$_N$), 4.93 (0.6 H, br dd, C^b***H***$_{2/diastereotopic}$ *R*$_N$), 7.11-7.84 (6.5 H, m, C$_5$***H***$_{5/Ph}$ *S*$_N$ + *R*$_N$ overlapped) ppm. ^{13}C{^{1}H} NMR (100 MHz; CD$_3$OD; 298 K): δ 20.8 (C^a***C***H$_3$ *S*$_N$), 21.3 (C^a***C***H$_3$ *R*$_N$), 22.3 (C^2***C***H$_3$ *R*$_N$), 24.2 (***C***^{4}H$_2$ *R*$_N$), 24.8 (***C***^{4}H$_2$ *S*$_N$), 25.8 (C^2***C***H$_3$ *S*$_N$), 29.1 (***C***^{6}H$_2$ *R*$_N$), 29.2 (***C***^{6}H$_2$ *S*$_N$), 30.6 (***C***^{3}H$_2$ *S*$_N$), 31.6 (***C***^{3}H$_2$ *R*$_N$), 38.4 (***C***^{5}H *S*$_N$), 39.2 (***C***^{5}H *R*$_N$), 74.2 (***C***^{2} *R*$_N$), 74.9 (***C***^{2} *S*$_N$), 111.9 (***C***^{b}H$_2$ *R*$_N$), 112.0 (***C***^{b}H$_2$ *S*$_N$), 123.5/125.2/126.6/127.8/128.1/128.3/129.0/129.6/130.4 (***C***$_5$H$_{5/Ph}$ *S*$_N$ + *R*$_N$ overlapped), 142.7 (*i*-***C*** *S*$_N$), 142.9 (*i*-***C*** *R*$_N$), 145.5 (***C***^{a} *R*$_N$), 146.2 (***C***^{a} *S*$_N$), 169.2 (***C***^{1} *R*$_N$), 170.8 (***C***^{1} *S*$_N$) ppm [40,49]. [^{1}H,^{13}C] HMBC (100.6 MHz; CDCl$_3$; 293 K): δ 75.1 (***C***^{2} *R*$_N$), 75.7 (***C***^{2} *S*$_N$), 170.6 (***C***^{1} *R*$_N$), 171.6 (***C***^{1} *S*$_N$) ppm. [^{1}H,^{15}N] HMBC (40.5 MHz; CDCl$_3$; 293 K): δ 51.2 (C^2***N***(Ph)H *S*$_N$, *R*$_N$ not detected), 171.6 (C^1***N***OH *S*$_N$, *R*$_N$ not detected) ppm.




Pt2
% Transmittance
Wavenumbers (cm-1)
598.09
590.72
559.83
513.95
492.33
450.79
438.96
405.51
368.25
350.20
335.21
327.21
301.32
296.53
270.53
241.98
219.77
208.40
202.96

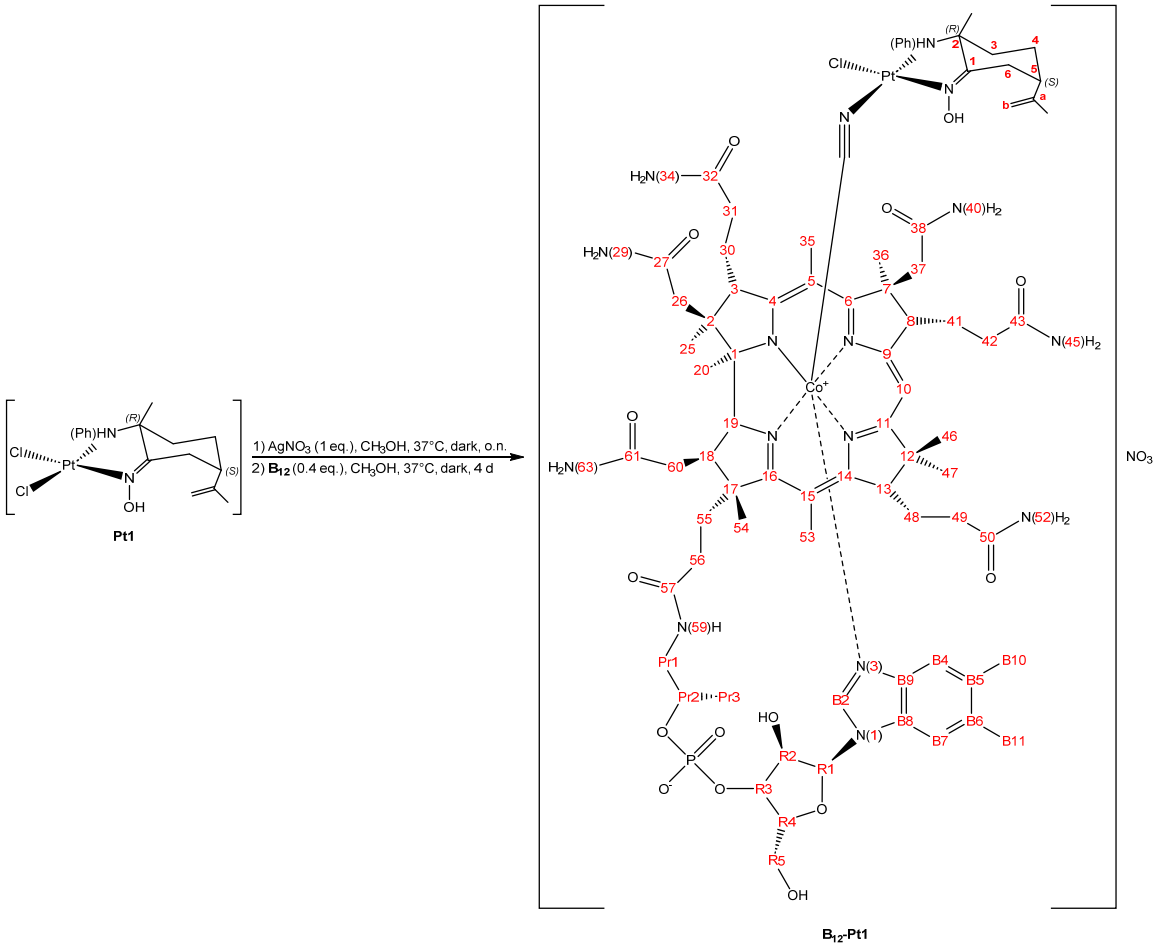

R_N-/S_N-Pt2, ^{1}H NMR (400 MHz, CD_3OD)
7.55
7.53
7.51
7.49
7.40
7.37
7.33
7.32
7.30
7.27
7.26
7.23
7.19
7.12
7.11
7.84
7.82
7.70
7.68
5.05
4.97
4.79
4.78
3.47
3.42
3.31
2.59
2.50
2.07
2.03
1.97
1.95
1.94
1.93
1.92
1.91
1.89
1.88
1.86
1.78
1.75
1.64
1.63
1.62
1.61
1.60
1.57
1.49
1.39
1.36
1.33
1.31
1.30
1.25
7.9
7.3
5.0
3.3
2.1
1.5
1H / ppm


R_N-/S_N-Pt2, $^{13}C\{^1H\}$ NMR (100 MHz, CD_3OD)
130.37
129.55
128.96
128.32
128.06
127.79
126.57
125.22
123.46
170.82
169.24
146.17
142.78
142.76
130.37
129.55
128.96
128.32
128.06
127.79
126.57
125.22
123.46
111.99
111.97
74.90
74.24
39.21
38.38
31.57
30.59
29.19
29.11
25.84
24.79
24.22
22.27
21.76
21.32
20.80
75 170 145 140 135 130 125 120 115 75 40 35 30 25 20
13C / ppm

Synthesis of the vitamin B_{12}–platinum(II) conjugates

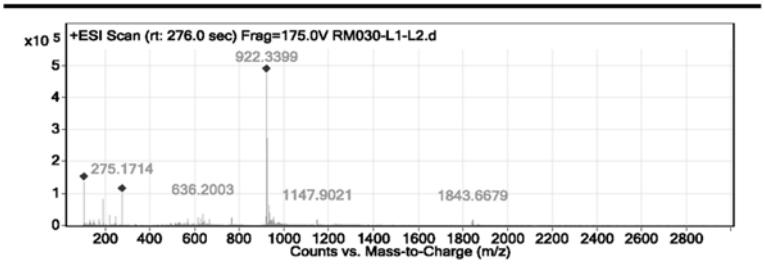
B_{12}-Pt1

A water solution (0.5 mL) of $AgNO_3$ (0.019 g, 0.11 mmol) was added dropwise to a methanol solution (6 mL) of **Pt1** (0.058 g, 0.11 mmol) and the mixture was stirred at 37°C overnight in the dark. The off-white insoluble residue (AgCl) was filtered off and the resulting clear pale yellow filtrate was added dropwise to a methanol solution (20 mL) of vitamin B_{12} (0.050 g, 0.04 mmol). The mixture was stirred at 37°C for 4 d in the dark and the solvent was subsequently evaporated to dryness. The crude product was then purified by semi-preparative reversed-phase HPLC (column: Kromasil 100-5-C18, 10×250 mm, 5 µm particle size) over 40 min at a flow rate of 3.0 mL min^{-1} (solvent A: H_2O + 0.1% TFA; solvent B: CH_3CN + 0.1% TFA; 0-6 min: isocratic flow of 90:10 parts A:B; 6-30 min linear gradient to 50:50 parts A:B; 30-35 min: linear gradient to 90:10 parts A:B; 35-40 min: isocratic flow of 90:10 parts A:B). The desired compound was eluted at a retention time of 23 min. The solvent was then evaporated to dryness and the residue was dried under vacuum over P_2O_5, yielding a dark purple solid identified as a mixture of R_N and S_N epimers (R_N:S_N ratio of *ca.* 2.1:1 in CD_3OD as estimated by ^1H NMR spectroscopy) of the title compound (MM($C_{79}H_{110}ClCoN_{17}O_{18}PPt$) = 1906.29 g mol^{-1}, 0.046 g, 60% yield).



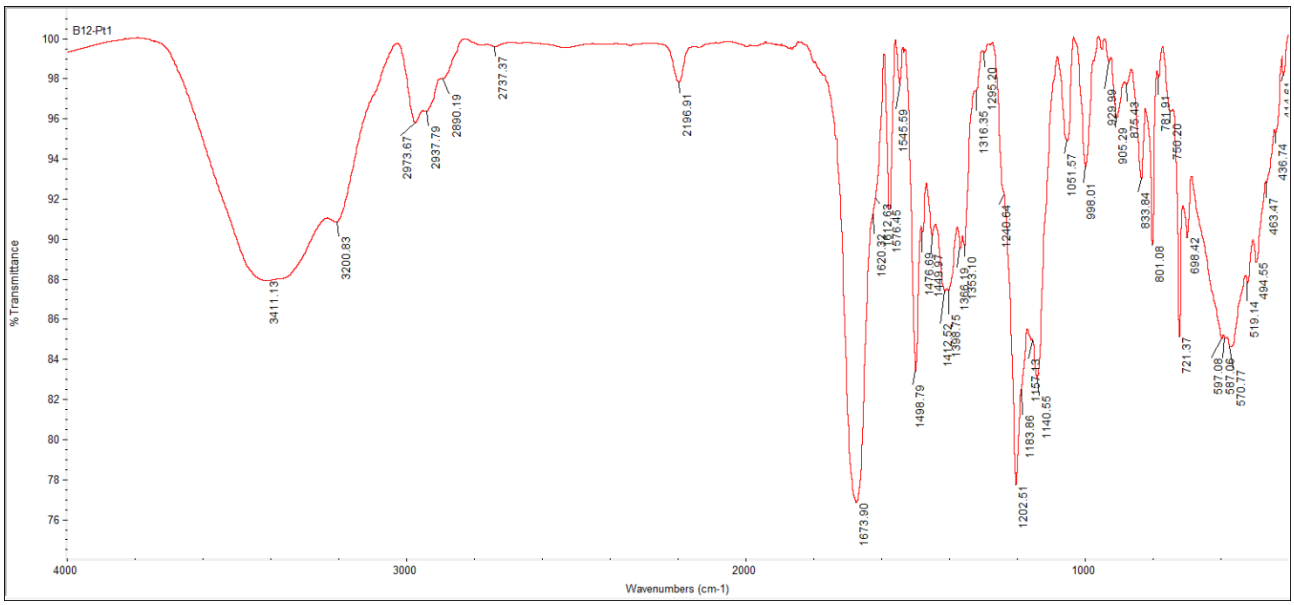
HRMS (CH_3OH, *m/z*) calcd. for $[M\text{-}NO_3]^+$: 1843.6750; found: 1843.6679; $[M\text{-}NO_3\text{+}H]^{2+}$: 922.3411; found: 922.3399. FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3411/3201 ($\nu_{a,s}(NH_2)_{amide}$ + $\nu(NH)_{amide}$ + $\nu(OH)_{ribose}$ + $\nu(OH)_{oxime}$ + $\nu(NH)_{amine}$ overlapped), 2197 (ν(C≡N)) [57,58], 1674 (ν(C=O)/amide I + $\nu(C=C)_{alkene}$ overlapped), 1620 (δ(NH_2)/amide II), 1613 ($\nu(C=N)_{oxime}$), 1577/1546 (corrin ring breathing mode), 1499/1477 (ν(C=N) + ν(C=C) overlapped), 1399 ($\nu_a(NO_3^-)$ [59], 1240 (ν(Ph–N)), 1157/1141 ($\nu_{oop}(PO_2^-)$), 998 (ν(P–O–C)), 905 ($\nu(N–OH)_{oxime}$), 834 ($w(NO_3^-)$ [59], 573 (δ(Co–C≡N)), 490 (ν(Co–N)), 454 (ν(Pt–NOH)), 435 (ν(Pt–NC), 396 (ν(Co–CN)), 342 (ν(Pt–NPh) + ν(Co–N) overlapped), 337 (ν(Pt–Cl)) cm^{-1}. ^1H NMR (500 MHz; CD_3OD; 298 K): δ 0.45 (3 H, s, $C^{20}\boldsymbol{H}_3$), 0.98-1.07 (1 H, m, $C^{41}\boldsymbol{H}'$), 1.19-1.31 (2 H, m, $C^{3Pt}\boldsymbol{H}_{2/diastereotopic}$ R_N), 1.25 (3 H, s, $C^{46}\boldsymbol{H}_3$), 1.26 (3 H, shoulder, $C^{Pr3}\boldsymbol{H}_3$), 1.28-1.34 (1 H, m, $C^{42}\boldsymbol{H}'$), 1.36 (3 H, s, $C^{25}\boldsymbol{H}_3$), 1.37 (3 H, s, $C^{54}\boldsymbol{H}_3$), 1.45 (3 H, s, $C^{47}\boldsymbol{H}_3$), 1.53 (3 H, br t, $C^{aPt}C\boldsymbol{H}_3$ R_N), 1.61-1.66 (1 H, m, $C^{42}\boldsymbol{H}''$), 1.74 (1.4 H, br t, $C^{aPt}C\boldsymbol{H}_3$ S_N), 1.79-1.88 (3 H, m, $C^{55}\boldsymbol{H}'$ + $C^{4Pt}\boldsymbol{H}_{2/diastereotopic}$ R_N overlapped), 1.83-2.05 (5 H, m, $C^{48}\boldsymbol{H}_{2/diastereotopic}$ + $C^{41}\boldsymbol{H}''$ + $C^{30}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 1.85 (3 H, s, $C^{36}\boldsymbol{H}_3$), 1.93 (1.4 H, s, $C^{2Pt}C\boldsymbol{H}_3$ S_N), 1.94 (3 H, s, $C^{2Pt}C\boldsymbol{H}_3$ R_N), 2.04-2.15 (1 H, m, $C^{56}\boldsymbol{H}'$), 2.17-2.24 (1 H, m, $C^{6Pt}\boldsymbol{H}_{ax}$ R_N), 2.29 (3 H, s, $C^{B11}\boldsymbol{H}_3$), 2.32 (3 H, s, $C^{B10}\boldsymbol{H}_3$), 2.33-2.41 (3 H, m, $C^{26}\boldsymbol{H}'$ + $C^{37}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 2.42-2.56 (5 H, m, $C^{5Pt}\boldsymbol{H}$ R_N + $C^{31}\boldsymbol{H}_{2/diastereotopic}$ + $C^{49}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 2.56-2.63 (3 H, m, $C^{26}\boldsymbol{H}''$ + $C^{55}\boldsymbol{H}''$ + $C^{56}\boldsymbol{H}''$ overlapped), 2.58 (3 H, s, $C^{53}\boldsymbol{H}_3$), 2.65 (3 H, s, $C^{35}\boldsymbol{H}_3$), 2.68-2.78 (2 H, m, $C^{60}\boldsymbol{H}_{2/diastereotopic}$), 2.80-2.91 (2 H, m, $C^{Pr1}\boldsymbol{H}'$ + $C^{18}\boldsymbol{H}$ overlapped), 3.30-3.35 (1 H, m, $C^{13}\boldsymbol{H}$), 3.44-3.49 (1 H, m, $C^{6Pt}\boldsymbol{H}_{eq}$ R_N), 3.50-3.54 (1 H, m, $C^{8}\boldsymbol{H}$), 3.65-3.67 (1 H, m, $C^{Pr1}\boldsymbol{H}''$), 3.74-3.92 (2 H, m, $C^{R5}\boldsymbol{H}_{2/diastereotopic}$), 4.05-4.10 (1 H, m, $C^{R4}\boldsymbol{H}$), 4.15-4.17 (1 H, m, $C^{19}\boldsymbol{H}$), 4.18-4.21 (1 H, m, $C^{R2}\boldsymbol{H}$), 4.30-4.36 (1 H, m, $C^{Pr2}\boldsymbol{H}$), 4.59 (2 H, m, $C^{bPt}\boldsymbol{H}_{2/diastereotopic}$ R_N), 4.64-4.71 (1 H, m, $C^{R3}\boldsymbol{H}$), 4.72-4.76 (1 H, m, $C^{3}\boldsymbol{H}$), 5.97 (1 H, s, $C^{10}\boldsymbol{H}$), 6.28 (1 H, br s, $C^{R1}\boldsymbol{H}$), 6.60 (1 H, br s, $C^{B4}\boldsymbol{H}$), 6.95-7.81 (7 H, m, $C_5\boldsymbol{H}_{5/PhPt}$ R_N + S_N overlapped), 7.12 (1 H, s, $C^{B2}\boldsymbol{H}$), 7.29 (1 H, s, $C^{B7}\boldsymbol{H}$) ppm. $^{13}C\{^1H\}$ NMR (125 MHz; CD_3OD; 298 K): δ 16.1 ($\boldsymbol{C}^{53}H_3$), 16.6 ($\boldsymbol{C}^{35}H_3$), 17.5 ($\boldsymbol{C}^{25}H_3$), 17.6 ($\boldsymbol{C}^{54}H_3$), 20.1 ($\boldsymbol{C}^{Pr3}H_3$), 20.2 ($\boldsymbol{C}^{B11}H_3$), 20.3 ($\boldsymbol{C}^{20}H_3$), 20.6 ($\boldsymbol{C}^{47}H_3$), 21.0 ($\boldsymbol{C}^{B10}H_3$), 21.6 ($C^{aPt}\boldsymbol{C}H_3$ R_N), 21.7 ($\boldsymbol{C}^{36}H_3$), 21.9 ($C^{2Pt}\boldsymbol{C}H_3$ S_N), 22.1 ($C^{aPt}\boldsymbol{C}H_3$ S_N), 25.5 ($\boldsymbol{C}^{4Pt}H_2$ R_N), 27.3 ($C^{2Pt}\boldsymbol{C}H_3$ R_N), 27.4 ($\boldsymbol{C}^{41}H_2$), 27.9 ($\boldsymbol{C}^{30}H_2$), 29.4 ($\boldsymbol{C}^{48}H_2$), 30.4 ($\boldsymbol{C}^{6Pt}H_2$ R_N), 31.5 ($\boldsymbol{C}^{3Pt}H_2$ R_N), 32.2 ($\boldsymbol{C}^{60}H_2$), 32.7 ($\boldsymbol{C}^{46}H_3$), 32.8 ($\boldsymbol{C}^{42}H_2$), 33.1 ($\boldsymbol{C}^{56}H_2$), 33.5 ($\boldsymbol{C}^{55}H_2$), 35.3 ($\boldsymbol{C}^{49}H_2$), 36.3 ($\boldsymbol{C}^{31}H_2$), 39.5 ($\boldsymbol{C}^{5Pt}H$ R_N), 40.3 ($\boldsymbol{C}^{18}H$), 43.7 ($\boldsymbol{C}^{37}H_2$), 46.5 ($\boldsymbol{C}^{26}H_2$), 46.8 ($\boldsymbol{C}^{Pr1}H_2$), ~49 ($\boldsymbol{C}^2$ + $\boldsymbol{C}^{12}$ overlapped with the residual peak of CD_3OD), 51.6 ($\boldsymbol{C}^7$), 55.1 ($\boldsymbol{C}^{13}H$), 57.1 ($\boldsymbol{C}^8H$), 57.5 ($\boldsymbol{C}^3H$), 60.5 ($\boldsymbol{C}^{17}$), 62.6 ($\boldsymbol{C}^{R5}H_2$), 70.7 ($\boldsymbol{C}^{R2}H$), 73.7 ($\boldsymbol{C}^{Pr2}H$), 75.5 ($\boldsymbol{C}^{R3}H$), 75.5 ($\boldsymbol{C}^{2Pt}$ R_N), 76.6 ($\boldsymbol{C}^{19}H$), 83.8 ($\boldsymbol{C}^{R4}H$), 86.5 ($\boldsymbol{C}^1$), 88.2 ($\boldsymbol{C}^{R1}H$), 95.4 ($\boldsymbol{C}^{10}H$), 105.0 ($\boldsymbol{C}^{15}$), 108.9 ($\boldsymbol{C}^5$), 112.8 ($\boldsymbol{C}^{B7}H$), 112.9 ($\boldsymbol{C}^{bPt}H_2$ R_N), 117.6 ($\boldsymbol{C}^{B4}H$), 124.3 ($\boldsymbol{C}$N) [28,29], 129.1/130.0 ($\boldsymbol{C}_5H_{5/PhPt}$ R_N), 131.4 ($\boldsymbol{C}^{B8}$), 134.1 ($\boldsymbol{C}^{B5}$), 135.9 ($\boldsymbol{C}^{B6}$), 138.1 ($\boldsymbol{C}^{B9}$), 143.0 (*i*-$\boldsymbol{C}^{Pt}$ R_N), 143.3 ($\boldsymbol{C}^{B2}H$), 147.0 ($\boldsymbol{C}^{aPt}$ R_N), 166.8 ($\boldsymbol{C}^{14}$), 167.4 ($\boldsymbol{C}^6$), 174.2 ($\boldsymbol{C}^{57}$), 174.7 ($\boldsymbol{C}^{38}$), 175.2 ($\boldsymbol{C}^{61}$), 175.6 ($\boldsymbol{C}^{27}$), 176.2 ($\boldsymbol{C}^{43}$), 176.7 ($\boldsymbol{C}^{32}$), 177.3 ($\boldsymbol{C}^9$), 177.5 ($\boldsymbol{C}^{50}$),

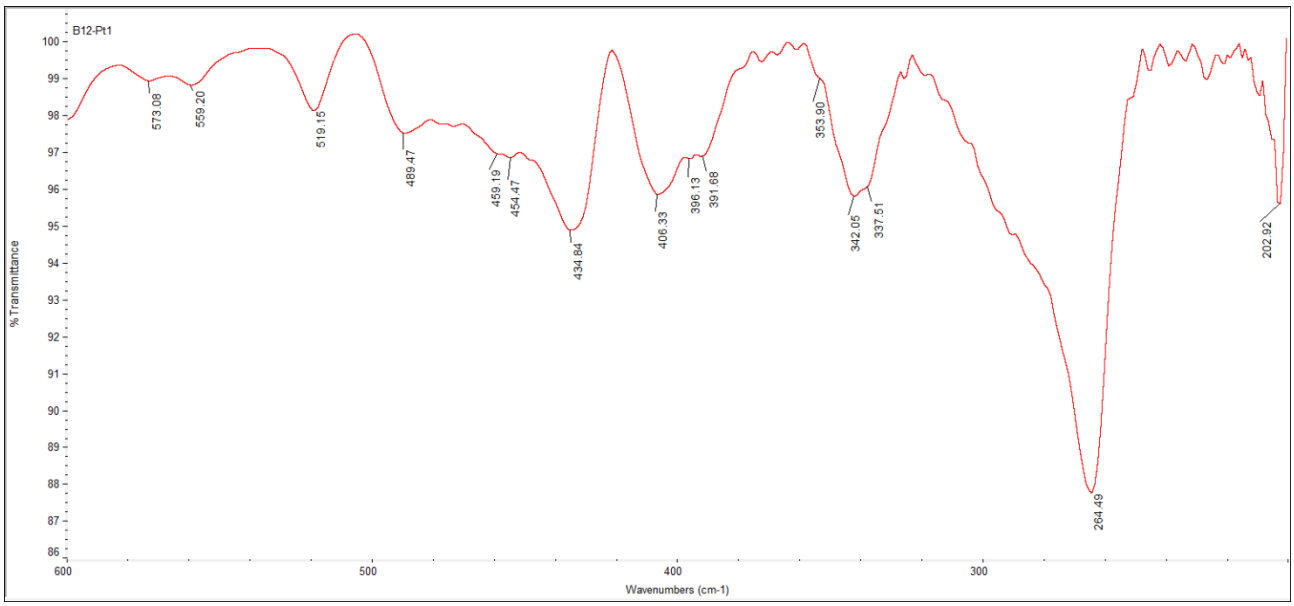
177.6 (***C***^{11}), 178.3 (***C***^{1Pt} R_N), 180.0 (***C***^{16}), 182.5 (***C***^{4}) ppm. $^{31}P\{^{1}H\}$ NMR (162 MHz; CD_3OD; 298 K): δ -0.1 (***P***O_4^-) ppm.

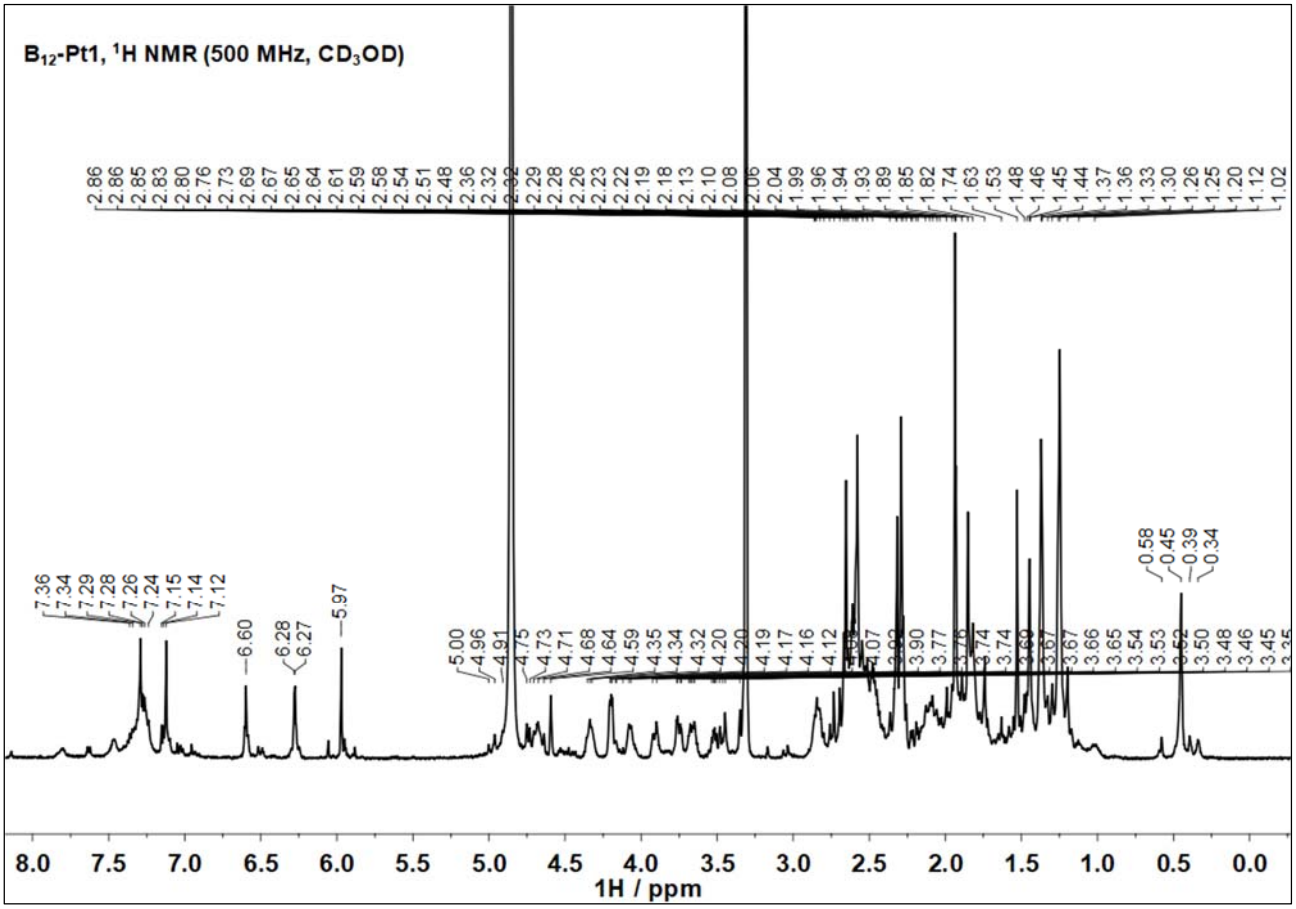


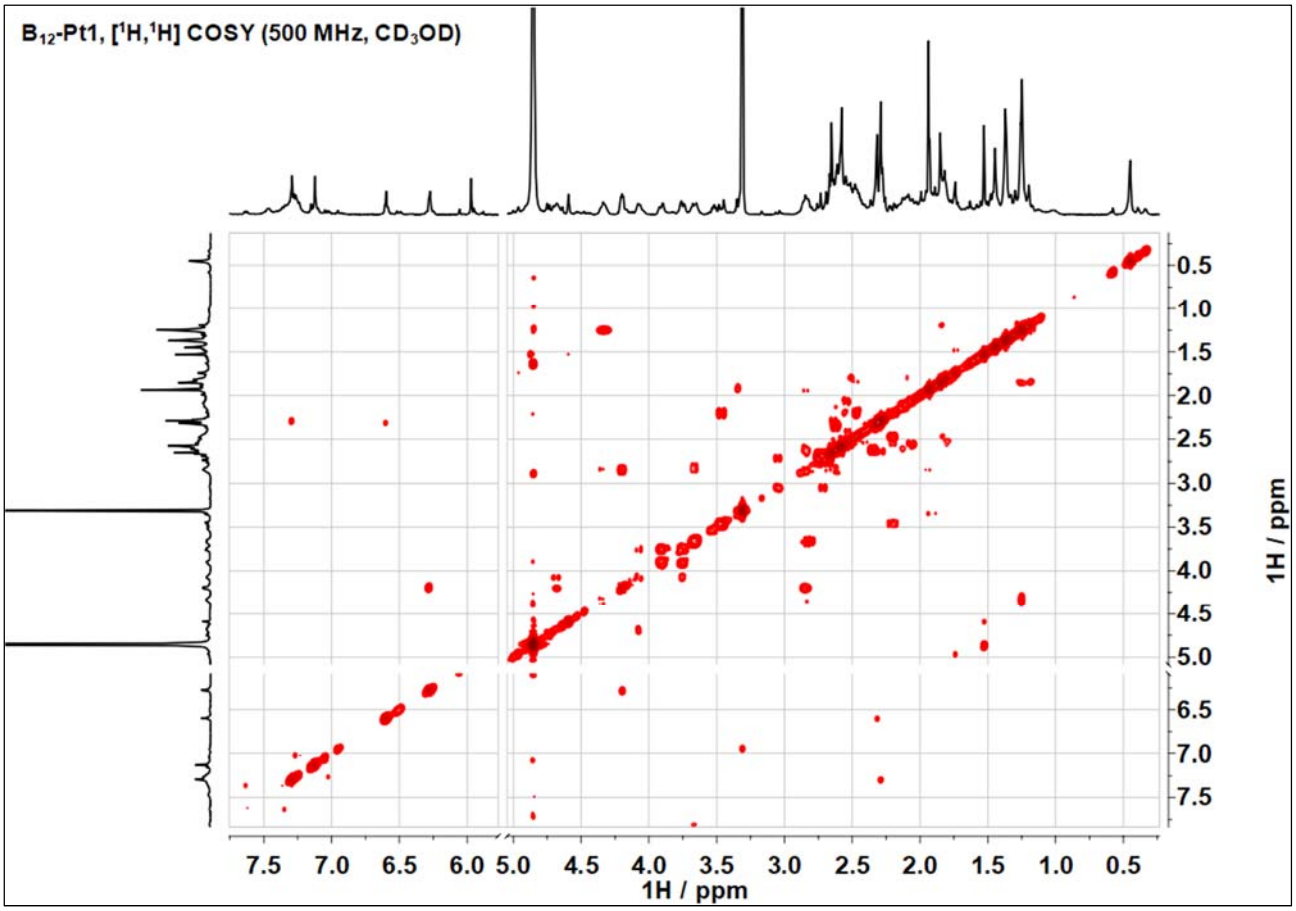
Peak List

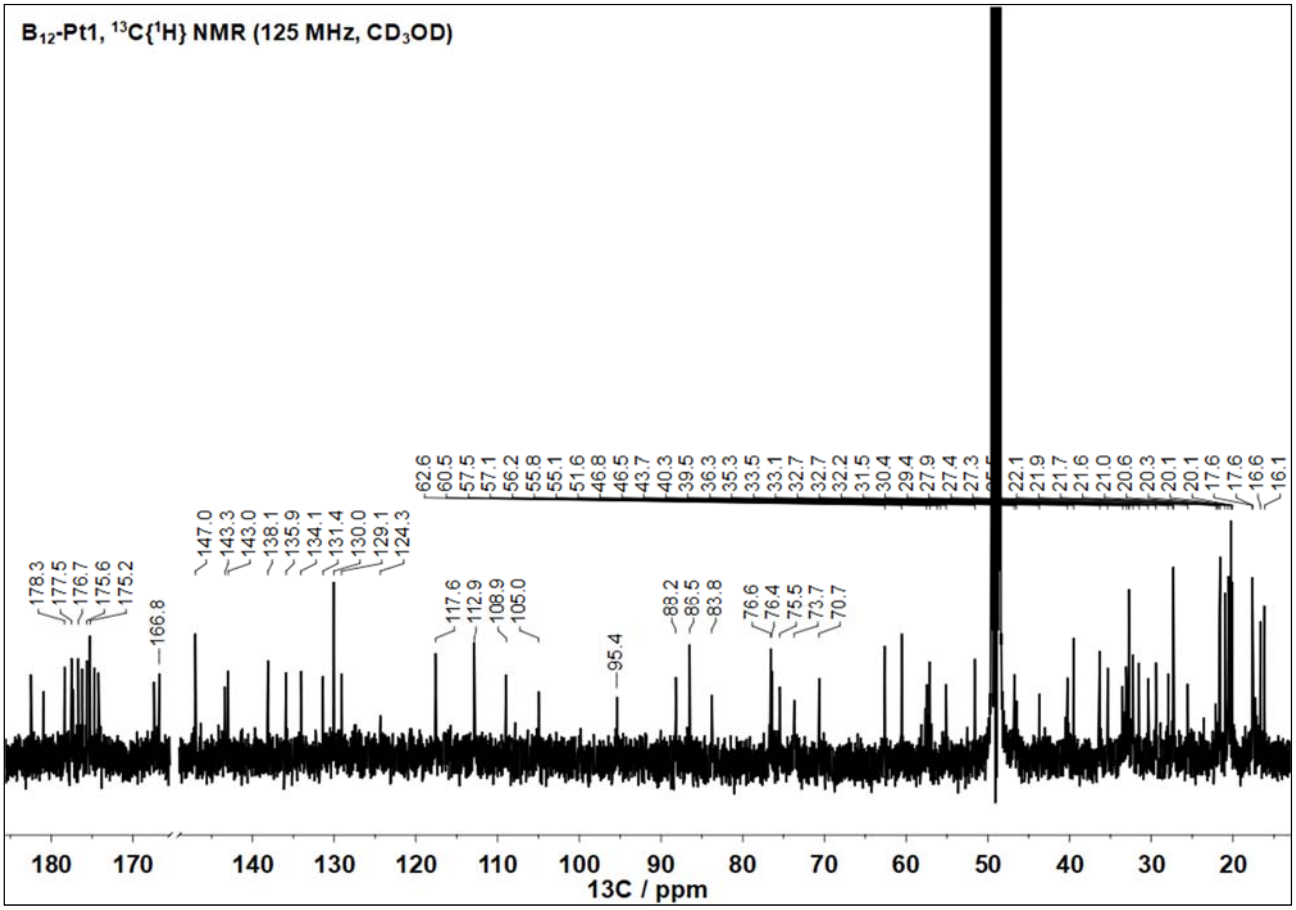
m/z	z	Abund
103.9558		136349.75
191.1026		82107.95
275.1714	1	100384.01
921.3385	2	212916
921.8398	2	376535.56
922.3399	2	473787.16
922.8404	2	372524.44
923.3413	2	271810.53
923.8423	2	154147.2
924.343	2	84339.22

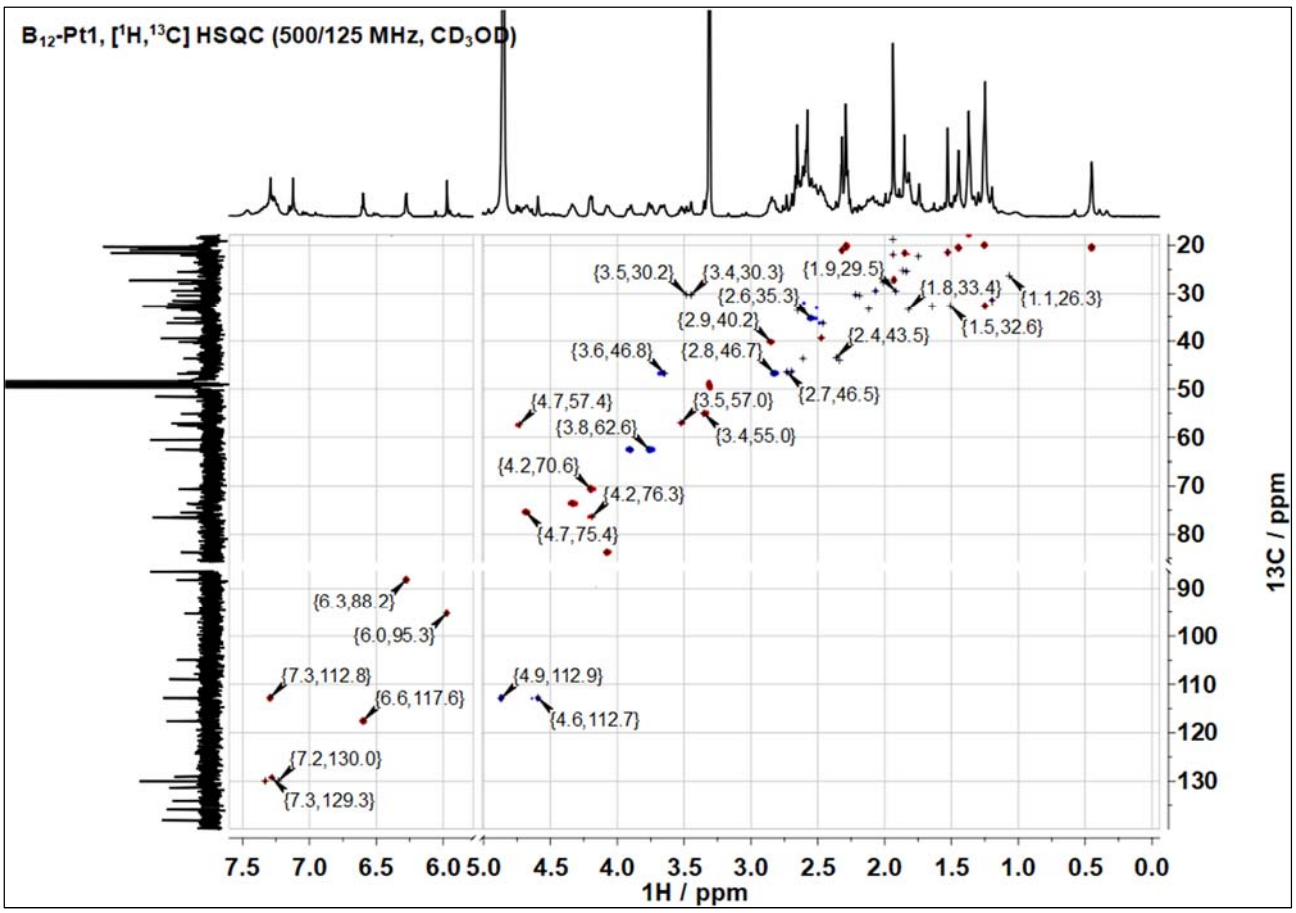


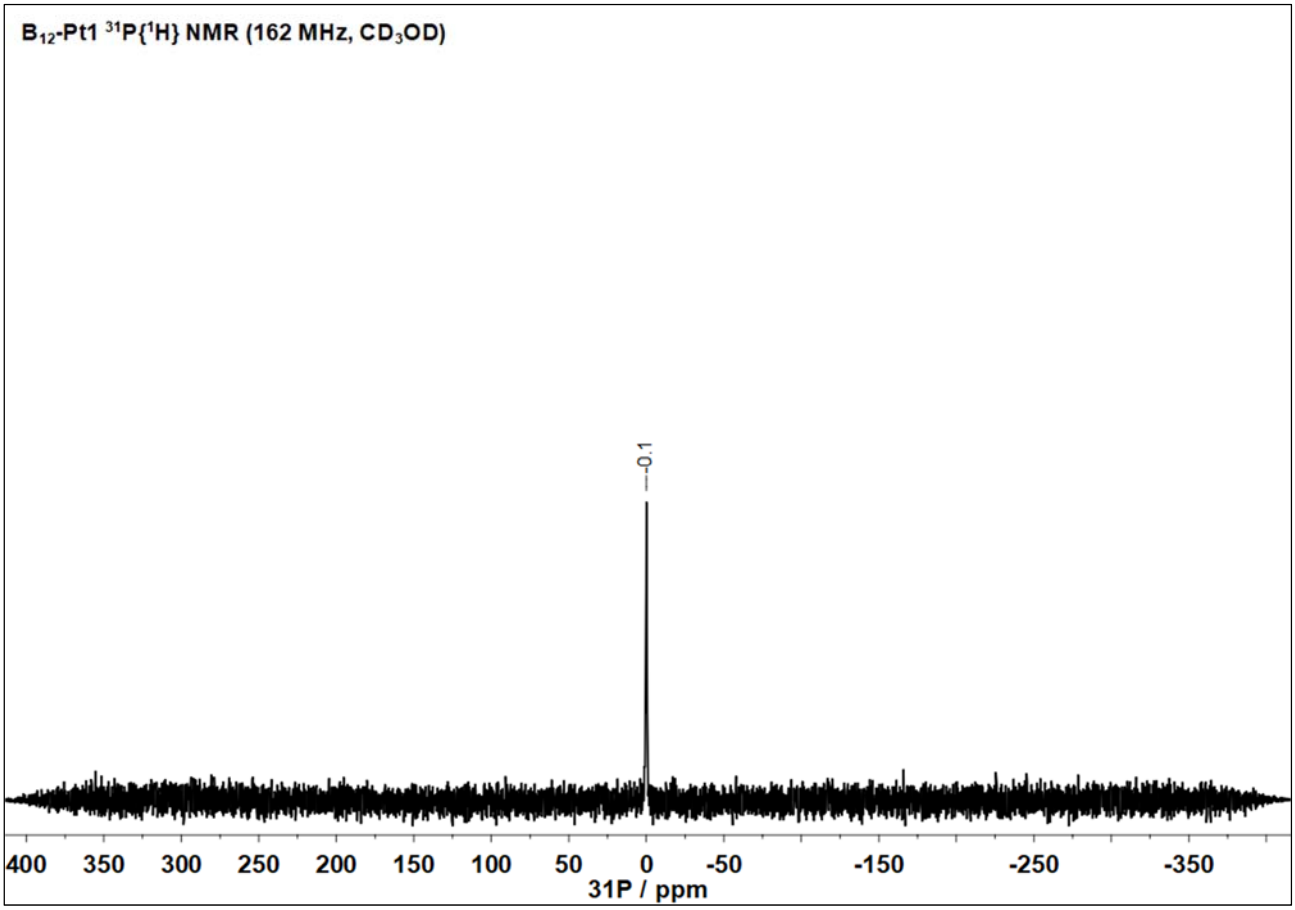



B12-Pt1, 1H NMR (500 MHz, CD3OD)
1H / ppm


B12-Pt1, [1H,1H] COSY (500 MHz, CD3OD)
1H / ppm
1H / ppm

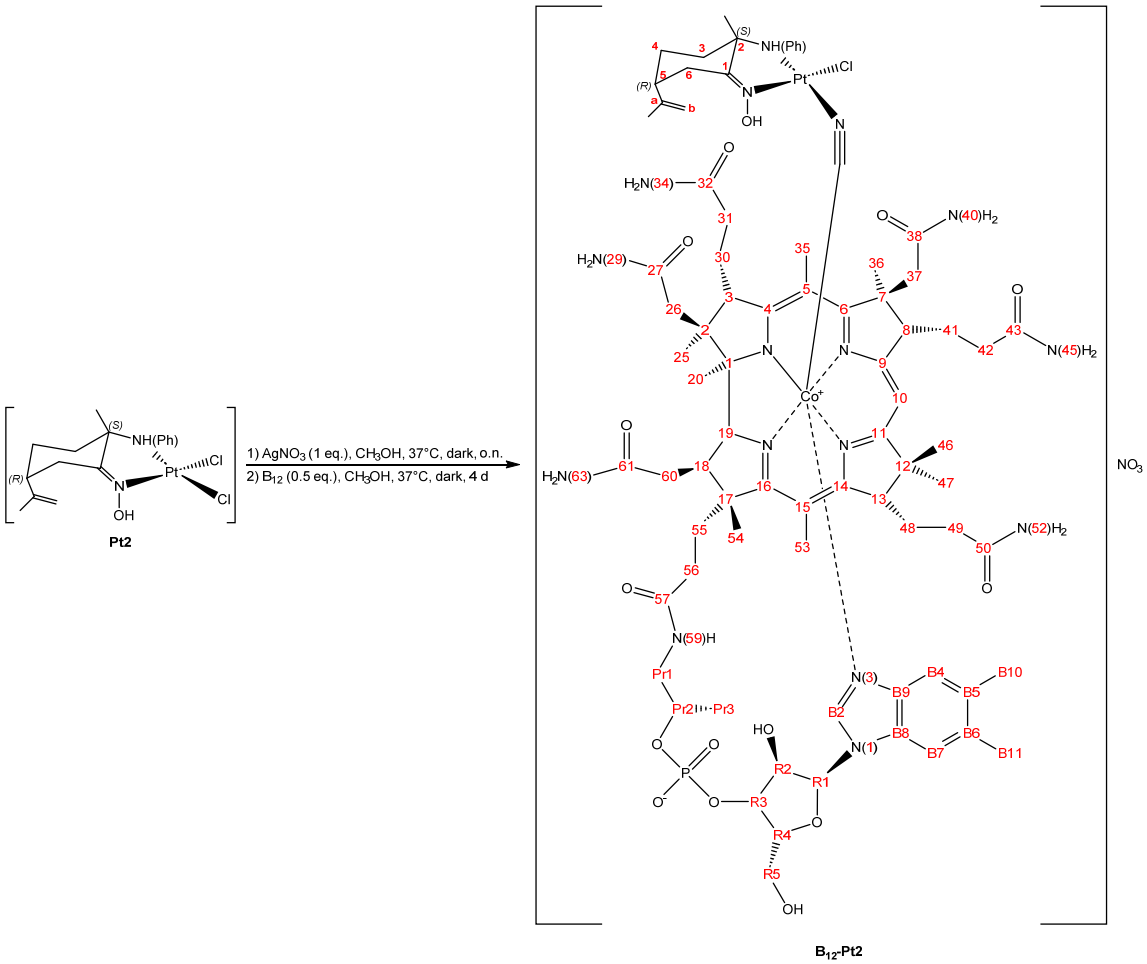

B12-Pt1, 13C{1H} NMR (125 MHz, CD3OD)
178.3
177.5
176.7
175.6
175.2
166.8
147.0
143.3
143.0
138.1
135.9
134.1
131.4
130.0
129.1
124.3
117.6
112.9
108.9
105.0
95.4
88.2
86.5
83.8
76.6
76.4
75.5
73.7
70.7
62.6
60.5
57.5
57.1
56.2
55.8
55.1
51.6
46.8
46.5
43.7
40.3
39.5
36.3
35.3
33.5
33.1
32.7
32.7
32.2
31.5
30.4
29.4
27.9
27.4
27.3
22.1
21.9
21.7
21.6
21.0
20.6
20.3
20.1
20.1
17.6
17.6
16.6
16.1
180
170
140
130
120
110
100
90
80
70
60
50
40
30
20
13C / ppm


B12-Pt1, [1H,13C] HSQC (500/125 MHz, CD3OD)
{3.5,30.2}
{3.4,30.3}
{1.9,29.5}
{2.6,35.3}
{1.8,33.4}
{1.1,26.3}
{2.9,40.2}
{1.5,32.6}
{2.4,43.5}
{3.6,46.8}
{2.8,46.7}
{2.7,46.5}
{4.7,57.4}
{3.5,57.0}
{3.8,62.6}
{3.4,55.0}
{4.2,70.6}
{4.2,76.3}
{4.7,75.4}
{6.3,88.2}
{6.0,95.3}
{7.3,112.8}
{6.6,117.6}
{4.9,112.9}
{4.6,112.7}
{7.2,130.0}
{7.3,129.3}
20
30
40
50
60
70
80
90
100
110
120
130
13C / ppm
7.5
7.0
6.5
6.0
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
1H / ppm



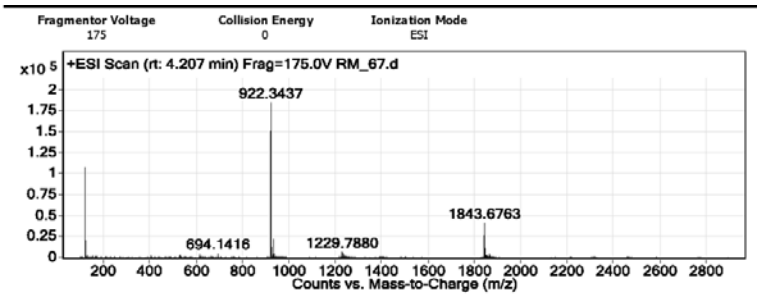
B_{12}-Pt2

A water solution (1 mL) of $AgNO_3$ (0.050 g, 0.29 mmol) was added dropwise to a methanol solution (6 mL) of **Pt2** (0.155 g, 0.29 mmol) and the mixture was stirred at 37°C overnight in the dark. The off-white insoluble residue (AgCl) was filtered off and the resulting clear pale yellow filtrate was added dropwise to a methanol solution (25 mL) of vitamin B_{12} (0.200 g, 0.15 mmol). The mixture was stirred at 37°C for 4 d in the dark and the solvent was subsequently evaporated to dryness. The crude product was then purified by semi-preparative reversed-phase HPLC (column: Kromasil 100-5-C18, 10×250 mm, 5 µm particle size) over 40 min at a flow rate of 3.0 mL min^{-1} (solvent A: H_2O + 0.1% TFA; solvent B: CH_3CN + 0.1% TFA; 0-6 min: isocratic flow of 90:10 parts A:B; 6-30 min linear gradient to 50:50 parts A:B; 30-35 min: linear gradient to 90:10 parts A:B; 35-40 min: isocratic flow of 90:10 parts A:B). The desired compound was eluted at a retention time of 23 min. The solvent was then evaporated to dryness and the residue was dried under vacuum over P_2O_5, yielding a dark purple solid identified as a mixture of S_N and R_N epimers (S_N:R_N ratio of *ca.* 2.5:1 in CD_3OD as estimated by ^1H NMR spectroscopy) of the title compound (MM($C_{79}H_{110}ClCoN_{17}O_{18}PPt$) = 1906.29 g mol^{-1}, 0.149 g, 52% yield).



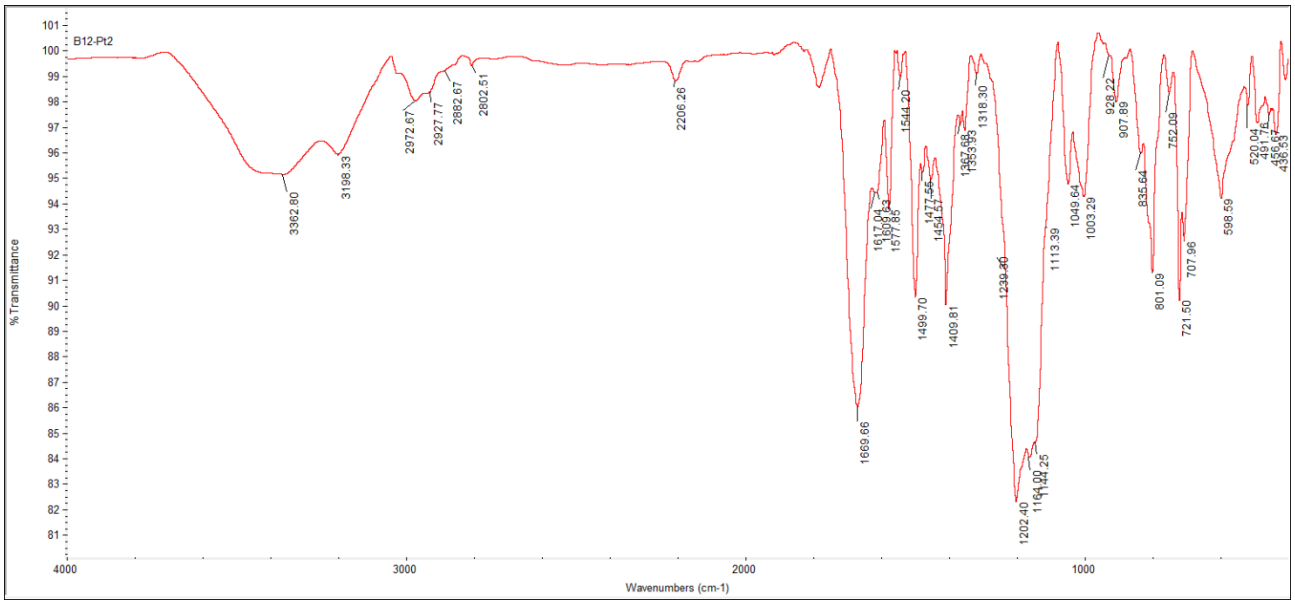
HRMS (CH_3OH, *m/z*) calcd. for $[M\text{-}NO_3]^+$: 1843.6750; found: 1843.6763; $[M\text{-}NO_3\text{+}H]^{2+}$: 922.3411; found: 922.3437. FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3363/3198 ($\nu_{a,s}(NH_2)_{amide}$ + $\nu(NH)_{amide}$ + $\nu(OH)_{ribose}$ + $\nu(OH)_{oxime}$ + $\nu(NH)_{amine}$ overlapped), 2206 ($\nu(C{\equiv}N)$) [57,58], 1670 ($\nu(C{=}O)$/amide I + $\nu(C{=}C)_{alkene}$ overlapped), 1617 ($\delta(NH_2)$/amide II), 1610 ($\nu(C{=}N)_{oxime}$), 1578/1544 (corrin ring breathing mode), 1500/1478 ($\nu(C{=}N)$ + $\nu(C{=}C)$ overlapped), 1410 ($\nu_a(NO_3^-)$ [59], 1239 ($\nu(Ph{-}N)$), 1164/1144 ($\nu_{oop}(PO_2^-)$), 1003 ($\nu(P{-}O{-}C)$), 908 ($\nu(N{-}OH)_{oxime}$), 836 ($w(NO_3^-)$ [59], 574 ($\delta(Co{-}C{\equiv}N)$), 494 ($\nu(Co{-}N)$), 454 ($\nu(Pt{-}NOH)$), 434 ($\nu(Pt{-}NC)$, 395 ($\nu(Co{-}CN)$), 345 ($\nu(Pt{-}NPh)$ + $\nu(Co{-}N)$ overlapped), 340 ($\nu(Pt{-}Cl)$) cm^{-1}. ^1H NMR (400 MHz; CD_3OD; 298 K): δ 0.44 (3 H, s, $C^{20}\boldsymbol{H}_3$), 0.96-1.08 (1 H, m, $C^{41}\boldsymbol{H}'$), 1.14-1.31 (2 H, m, $C^{3Pt}\boldsymbol{H}_{2/diastereotopic}$ *S*N), 1.24 (3 H, s, $C^{46}\boldsymbol{H}_3$), 1.26 (3 H, d, $C^{Pr3}\boldsymbol{H}_3$, $^3J_{H,H}$ = 6.4 Hz), 1.27-1.35 (1 H, m, $C^{42}\boldsymbol{H}'$), 1.36 (3 H, s, $C^{25}\boldsymbol{H}_3$), 1.38 (3 H, s, $C^{54}\boldsymbol{H}_3$), 1.44 (3 H, s, $C^{47}\boldsymbol{H}_3$), 1.52 (3 H, m, $C^{aPt}C\boldsymbol{H}_3$ *S*N), 1.59-1.65 (1 H, m, $C^{42}\boldsymbol{H}''$), 1.73 (1.2 H, m, $C^{aPt}C\boldsymbol{H}_3$ *R*N), 1.77-1.90 (3 H, m, $C^{55}\boldsymbol{H}'$ + $C^{4Pt}\boldsymbol{H}_{2/diastereotopic}$ *S*N overlapped), 1.88-2.04 (5 H, m, $C^{48}\boldsymbol{H}_{2/diastereotopic}$ + $C^{41}\boldsymbol{H}''$ + $C^{30}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 1.85 (3 H, s, $C^{36}\boldsymbol{H}_3$), 1.94 (1.3 H, s, $C^{2Pt}C\boldsymbol{H}_3$ *R*N), 1.95 (3 H, s, $C^{2Pt}C\boldsymbol{H}_3$ *S*N), 2.04-2.14 (1 H, m, $C^{56}\boldsymbol{H}'$), 2.16-2.25 (1 H, m, $C^{6Pt}\boldsymbol{H}_{ax}$ *S*N), 2.28 (3 H, s, $C^{B11}\boldsymbol{H}_3$), 2.31 (3 H, s, $C^{B10}\boldsymbol{H}_3$), 2.33-2.42 (3 H, m, $C^{26}\boldsymbol{H}'$ + $C^{37}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 2.42-2.56 (5 H, m, $C^{5Pt}\boldsymbol{H}$ *S*N + $C^{31}\boldsymbol{H}_{2/diastereotopic}$ + $C^{49}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 2.54-2.63 (3 H, m, $C^{26}\boldsymbol{H}''$ + $C^{55}\boldsymbol{H}''$ + $C^{56}\boldsymbol{H}''$

overlapped), 2.58 (3 H, s, $C^{53}\boldsymbol{H}_3$), 2.65 (3 H, s, $C^{35}\boldsymbol{H}_3$), 2.68-2.78 (2 H, m, $C^{60}\boldsymbol{H}_{2/\text{diastereotopic}}$), 2.79-2.90 (2 H, m, $C^{Pr1}\boldsymbol{H}'$ + $C^{18}\boldsymbol{H}$ overlapped), 3.30-3.35 (1 H, m, $C^{13}\boldsymbol{H}$), 3.44-3.48 (1 H, m, $C^{6Pt}\boldsymbol{H}_{eq}$ S_N), 3.51-3.54 (1 H, m, $C^{8}\boldsymbol{H}$), 3.65-3.68 (1 H, m, $C^{Pr1}\boldsymbol{H}''$), 3.72-3.95 (2 H, m, $C^{R5}\boldsymbol{H}_{2/\text{diastereotopic}}$), 4.09-4.14 (1 H, m, $C^{R4}\boldsymbol{H}$), 4.20 (1 H, d, $C^{19}\boldsymbol{H}$, $^3J_{H,H}$ = 11.2 Hz), 4.21-4.23 (1 H, m, $C^{R2}\boldsymbol{H}$), 4.33-4.47 (1 H, m, $C^{Pr2}\boldsymbol{H}$), 4.59-4.86 (2 H, m, $C^{bPt}\boldsymbol{H}_{2/\text{diastereotopic}}$ S_N), 4.64-4.71 (1 H, m, $C^{R3}\boldsymbol{H}$), 4.72-4.76 (1 H, m, $C^{3}\boldsymbol{H}$), 5.99 (1 H, s, $C^{10}\boldsymbol{H}$), 6.29 (1 H, d, $C^{R1}\boldsymbol{H}$, $^3J_{H,H}$ = 2.8 Hz), 6.60 (1 H, br s, $C^{B4}\boldsymbol{H}$), 6.90-7.83 (7 H, m, $C_5\boldsymbol{H}_{5/PhPt}$ S_N + R_N overlapped), 7.12 (1 H, s, $C^{B2}\boldsymbol{H}$), 7.28 (1 H, s, $C^{B7}\boldsymbol{H}$) ppm. $^{13}C\{^1H\}$ NMR (100 MHz; CD_3OD; 298 K): δ 16.1 ($\boldsymbol{C}^{53}H_3$), 16.6 ($\boldsymbol{C}^{35}H_3$), 17.6 ($\boldsymbol{C}^{25}H_3$), 17.7 ($\boldsymbol{C}^{54}H_3$), 20.1 ($\boldsymbol{C}^{Pr3}H_3$ + $\boldsymbol{C}^{B11}H_3$ overlapped), 20.3 ($\boldsymbol{C}^{20}H_3$), 20.5 ($\boldsymbol{C}^{47}H_3$), 21.0 ($\boldsymbol{C}^{B10}H_3$), 21.6 ($C^{aPt}\boldsymbol{C}H_3$ S_N), 21.7 ($\boldsymbol{C}^{36}H_3$), 21.8 ($C^{2Pt}\boldsymbol{C}H_3$ R_N), 22.1 ($C^{aPt}\boldsymbol{C}H_3$ R_N), 25.5 ($\boldsymbol{C}^{4Pt}H_2$ S_N), 27.3 ($C^{2Pt}\boldsymbol{C}H_3$ S_N + $\boldsymbol{C}^{41}H_2$ overlapped), 28.0 ($\boldsymbol{C}^{30}H_2$), 29.4 ($\boldsymbol{C}^{48}H_2$), 30.4 ($\boldsymbol{C}^{6Pt}H_2$ S_N), 31.5 ($\boldsymbol{C}^{3Pt}H_2$ S_N), 32.2 ($\boldsymbol{C}^{60}H_2$), 32.7 ($\boldsymbol{C}^{46}H_3$), 32.9 ($\boldsymbol{C}^{42}H_2$), 33.4 ($\boldsymbol{C}^{56}H_2$), 33.6 ($\boldsymbol{C}^{55}H_2$), 35.3 ($\boldsymbol{C}^{49}H_2$), 36.3 ($\boldsymbol{C}^{31}H_2$), 39.5 ($\boldsymbol{C}^{5Pt}H$ S_N), 40.2 ($\boldsymbol{C}^{18}H$), 43.5 ($\boldsymbol{C}^{37}H_2$), 46.4 ($\boldsymbol{C}^{26}H_2$), 46.6 ($\boldsymbol{C}^{Pr1}H_2$), ~49 ($\boldsymbol{C}^2$ + $\boldsymbol{C}^{12}$ overlapped with the residual peak of CD_3OD), 51.6 ($\boldsymbol{C}^7$), 55.1 ($\boldsymbol{C}^{13}H$), 57.1 ($\boldsymbol{C}^8H$), 57.4 ($\boldsymbol{C}^3H$), 60.5 ($\boldsymbol{C}^{17}$), 62.3 ($\boldsymbol{C}^{R5}H_2$), 70.5 ($\boldsymbol{C}^{R2}H$), 74.4 ($\boldsymbol{C}^{Pr2}H$), 75.5 ($\boldsymbol{C}^{R3}H$), 76.4 ($\boldsymbol{C}^{2Pt}$ S_N), 76.6 ($\boldsymbol{C}^{19}H$), 83.6 ($\boldsymbol{C}^{R4}H$), 86.4 ($\boldsymbol{C}^1$), 88.2 ($\boldsymbol{C}^{R1}H$), 95.4 ($\boldsymbol{C}^{10}H$), 104.9 ($\boldsymbol{C}^{15}$), 109.0 ($\boldsymbol{C}^5$), 112.8 ($\boldsymbol{C}^{B7}H$), 112.9 ($\boldsymbol{C}^{bPt}H_2$ R_N), 117.4 ($\boldsymbol{C}^{B4}H$), 124.4 ($\boldsymbol{C}N$) [52,53], 129.1/130.0 ($\boldsymbol{C}_5H_{5/PhPt}$ S_N), 131.3 ($\boldsymbol{C}^{B8}$), 134.1 ($\boldsymbol{C}^{B5}$), 135.9 ($\boldsymbol{C}^{B6}$), 138.1 ($\boldsymbol{C}^{B9}$), 143.0 (i-$\boldsymbol{C}^{Pt}$ S_N), 143.3 ($\boldsymbol{C}^{B2}H$), 147.0 ($\boldsymbol{C}^{aPt}$ S_N), 166.7 ($\boldsymbol{C}^{14}$), 167.4 ($\boldsymbol{C}^6$), 174.5 ($\boldsymbol{C}^{57}$), 174.8 ($\boldsymbol{C}^{38}$), 175.4 ($\boldsymbol{C}^{61}$), 175.7 ($\boldsymbol{C}^{27}$), 176.2 ($\boldsymbol{C}^{43}$), 176.7 ($\boldsymbol{C}^{32}$), 177.4 ($\boldsymbol{C}^9$), 177.5 ($\boldsymbol{C}^{50}$), 177.6 ($\boldsymbol{C}^{11}$), 178.3 ($\boldsymbol{C}^{1Pt}$ S_N), 180.9 ($\boldsymbol{C}^{16}$), 182.5 ($\boldsymbol{C}^4$) ppm. $^{31}P\{^1H\}$ NMR (162 MHz; CD_3OD; 298 K): δ -0.2 ($\boldsymbol{P}O_4^-$) ppm.



Peak List

m/z	z	Abund
121.0509	1	107621.52
921.3409		80172.95
921.843	1	150314.69
922.01	1	78187.82
922.3437	1	184514.05
922.8445	1	146677.75
923.344	1	119528.5
923.8444	1	64409.46
1843.6763	2	40775.96
1844.6786	2	33415.59


B12-Pt2
%Transmittance
Wavenumbers (cm-1)
3362.80
3198.33
2972.67
2927.77
2882.67
2802.51
2206.26
1669.66
1617.04
1600.43
1577.85
1544.20
1499.70
1477.56
1454.57
1409.81
1357.68
1353.98
1318.30
1238.80
1202.40
1164.00
1144.25
1113.39
1049.64
1003.29
928.22
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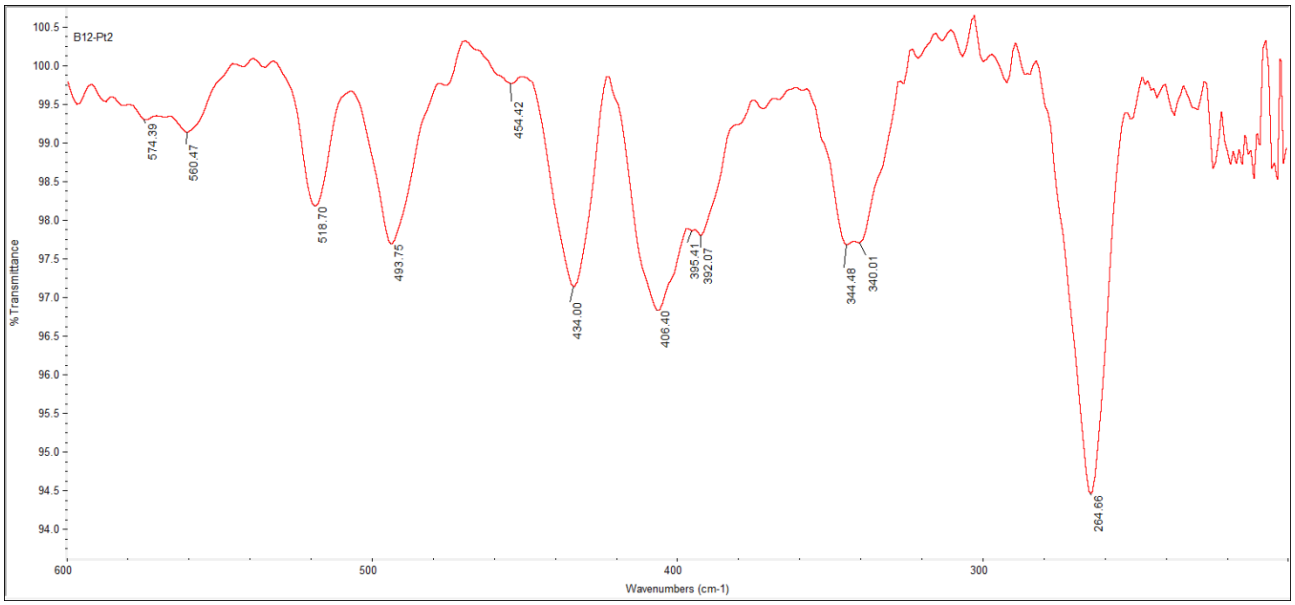
707.96
598.59
520.04
491.76
456.67
436.53


B12-Pt2
%Transmittance
Wavenumbers (cm-1)
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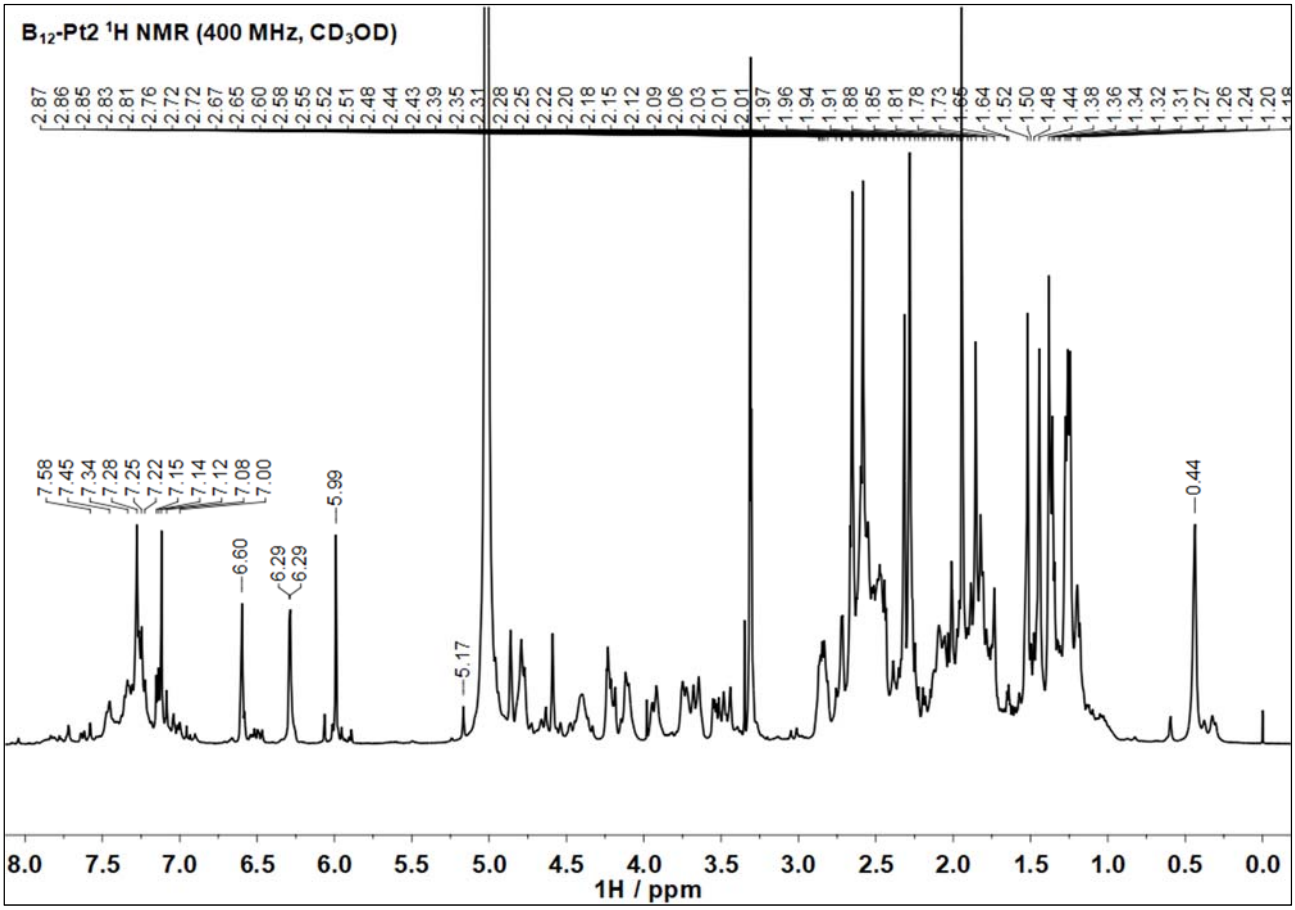
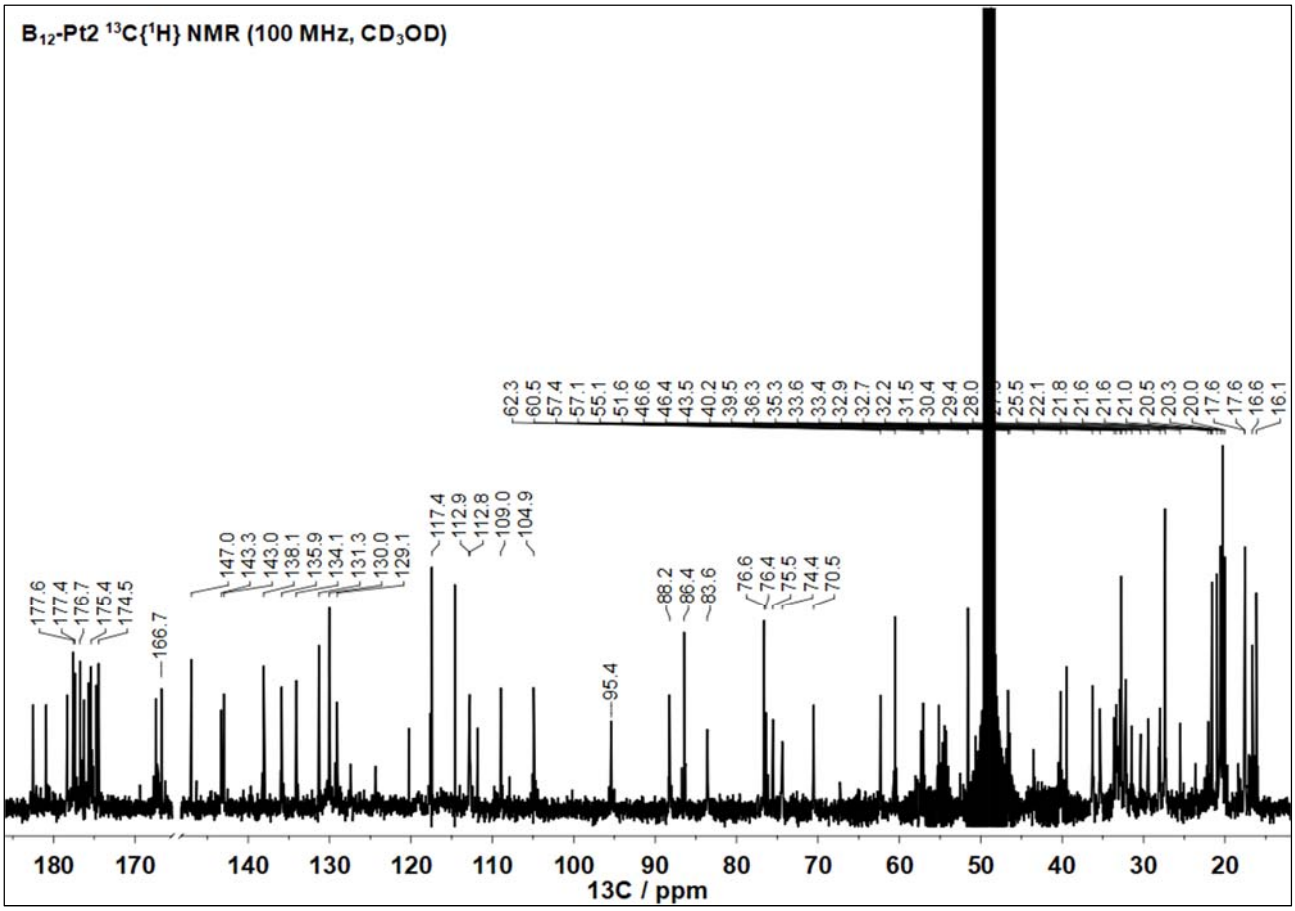
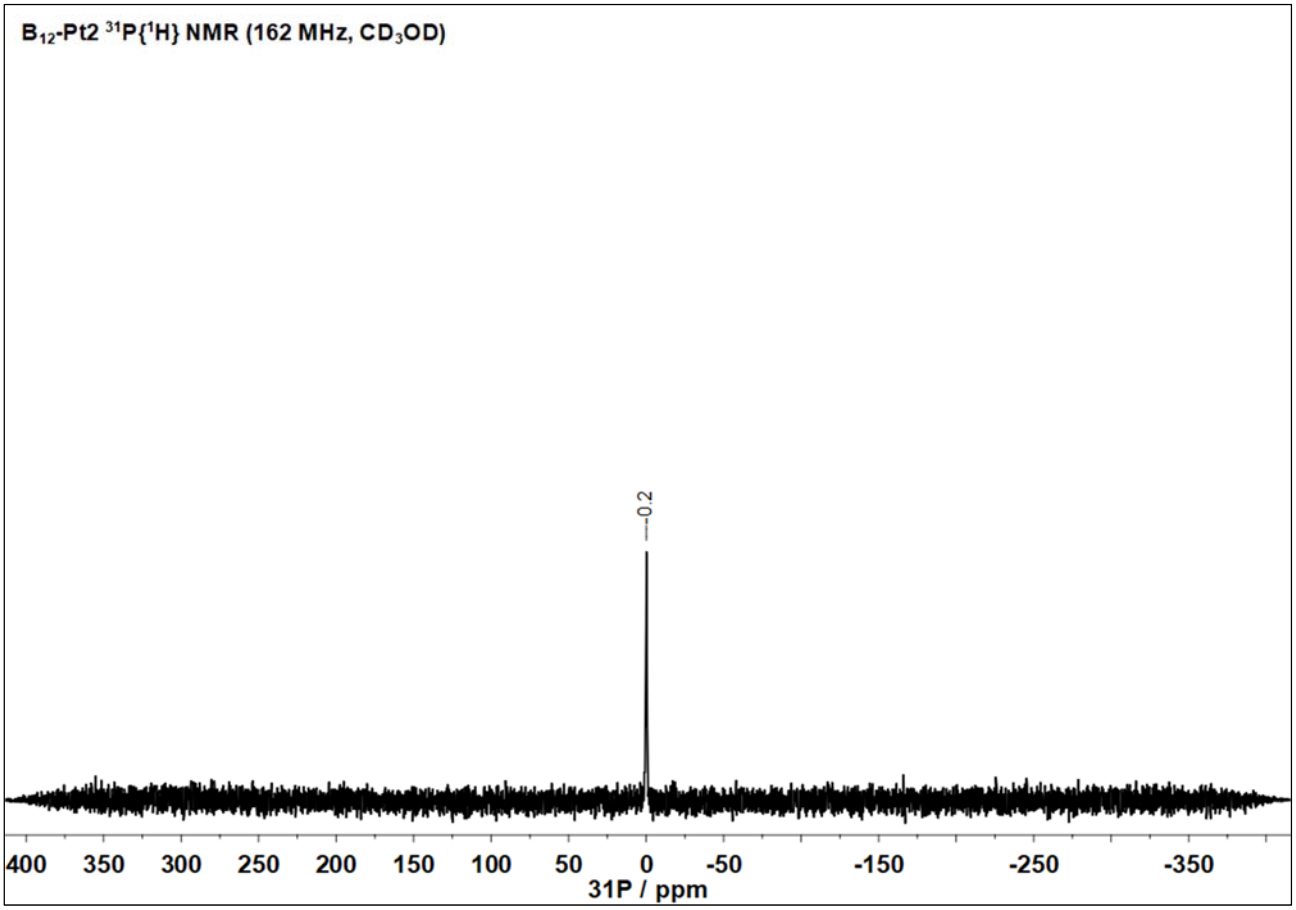
560.47
518.70
493.75
454.42
434.00
406.40
395.41
392.07
344.48
340.01
264.66

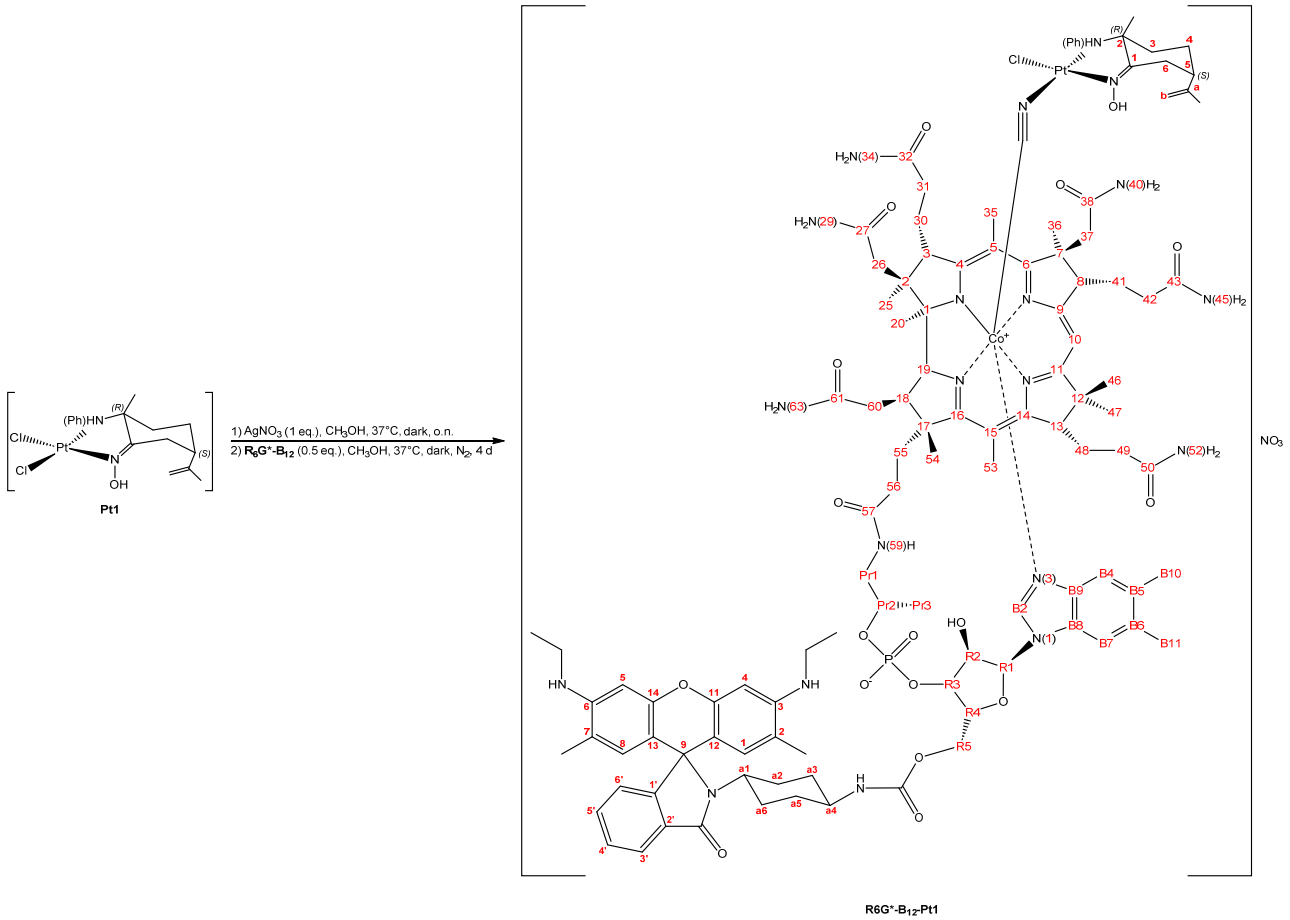

B12-Pt2 1H NMR (400 MHz, CD3OD)
1H / ppm


B12-Pt2 13C{1H} NMR (100 MHz, CD3OD)
13C / ppm



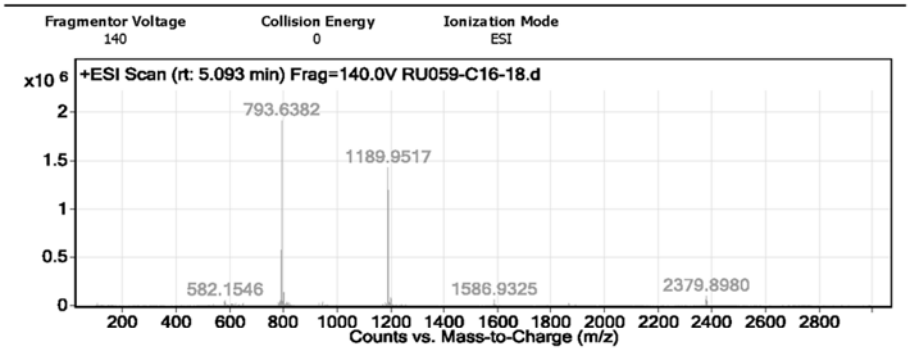
R6G*-B_{12}-Pt1

A water solution (0.5 mL) of $AgNO_3$ (0.017 g, 0.10 mmol) was added dropwise to a methanol solution (5 mL) of **Pt1** (0.052 g, 0.10 mmol) and the mixture was stirred at 37°C overnight in the dark. The off-white insoluble residue (AgCl) was filtered off and the resulting clear pale yellow filtrate was added dropwise to a methanol solution (4 mL) of the rhodamine 6G–*trans*-1,4-diaminocyclohexane–vitamin B_{12} conjugate (**R6G*-B_{12}**, 0.095 g, 0.05 mmol). The mixture was stirred at 37°C for 4 d in the dark under an inert atmosphere of nitrogen and the solvent was subsequently evaporated to dryness. The crude product was then purified by semi-preparative reversed-phase HPLC (column: Kromasil 100-5-C18, 10×250 mm, 5 µm particle size) over 40 min at a flow rate of 3.0 mL min^{-1} (solvent A: H_2O + 0.1% TFA; solvent B: CH_3CN + 0.1% TFA; 0-5 min: isocratic flow of 60:40 parts A:B; 5-30 min linear gradient to 30:70 parts A:B; 30-35 min: isocratic flow of 30:70 parts A:B; 35-40 min: isocratic flow of 60:40 parts A:B). The desired compound was eluted at a retention time of 16 min. The solvent was then evaporated to dryness and the residue was dried under vacuum over P_2O_5, yielding a dark pink solid identified as a mixture of R_N and S_N epimers (R_N:S_N ratio of *ca.* 2.5:1 in CD_3OD as estimated by ^1H NMR spectroscopy) of the title compound (MM($C_{112}H_{146}ClCoN_{21}O_{21}PPt$) = 2442.97 g mol^{-1}, 0.055 g, 45% yield).



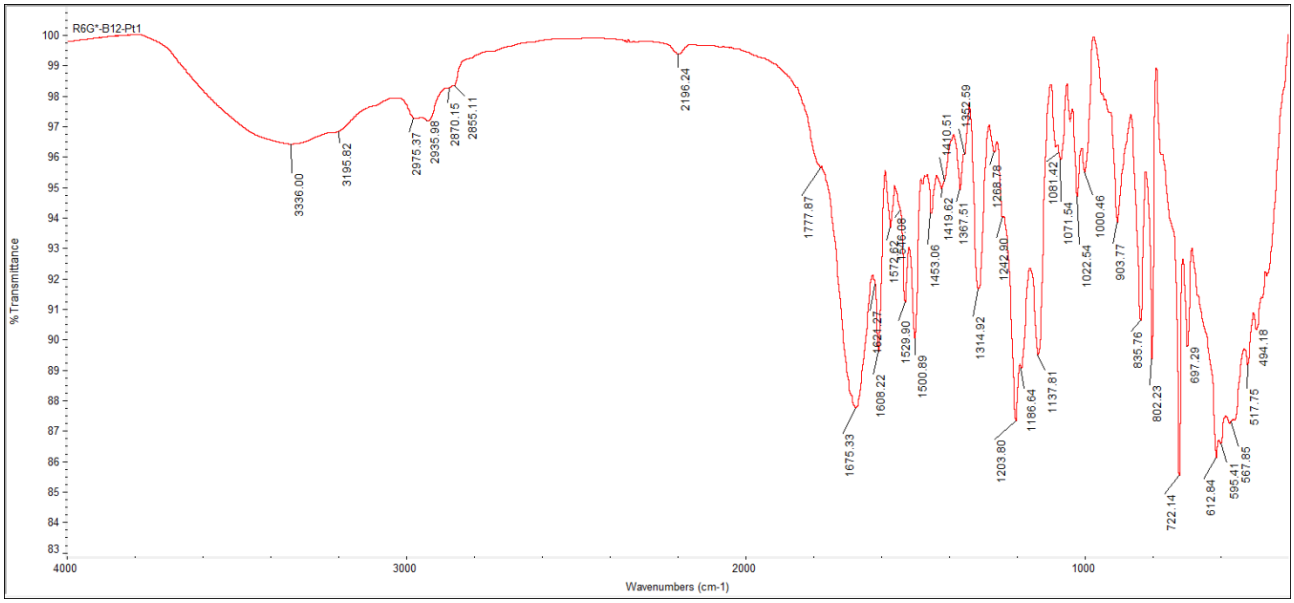
HRMS (CH_3OH, *m/z*) calcd. for $[M\text{-}NO_3]^+$: 2379.9558; found: 2379.8980; $[M\text{-}NO_3+H]^{2+}$: 1190.4815; found: 1189.9517; $[M\text{-}NO_3+2H]^{3+}$: 793.9901; found: 793.6382. FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3336/3196 ($\nu_{a,s}(NH_2)_{amide}$ + $\nu(NH)_{amide}$ + $\nu(NH)_{amine}$ + $\nu(OH)_{oxime}$ overlapped), 2197 ($\nu(C{\equiv}N)$), 1675 ($\nu(C{=}O)$/amide I + ($\nu(C{=}O)_{lactam}$ + $\nu(C{=}C)_{alkene}$ overlapped), 1621 ($\delta(NH_2)$/amide II), 1608 ($\nu(C{=}N)_{oxime}$), 1572/1546 (corrin ring breathing mode), 1530/1501 ($\delta(C{-}N{-}H)_{amine}$ + $\delta_{ip}(C{-}N{-}H)$/amide II + $\nu(C{=}C)$ + $\nu(C{=}N)$ overlapped), 1411 ($\nu_a(NO_3^-)$, 1353 ($\nu(N{-}Ph)_{amine}$), 1269 ($\nu(C{-}O{-}C)_{xanthene}$), 1243 ($\nu(Ph{-}N)$), 1138 ($\nu_{oop}(PO_2^-)$), 1072 ($\nu_{ip}(PO_2^-)$), 1001 ($\nu(P{-}O{-}C)$), 904 ($\nu(N{-}OH)_{oxime}$), 836 ($w(NO_3^-)$, 570 ($\delta(Co{-}C{\equiv}N)$), 491 ($\nu(Co{-}N)$), 453 ($\nu(Pt{-}NOH)$), 436 ($\nu(Pt{-}NC)$, 401 ($\nu(Co{-}CN)$), 342 ($\nu(Pt{-}NPh)$ + $\nu(Co{-}N)$ overlapped), 336 ($\nu(Pt{-}Cl)$) cm^{-1}. ^1H NMR (600 MHz; CD_3OD; 298 K): δ 0.41 (3 H, s, $C^{20}\boldsymbol{H}_3$), 1.16-1.19 (2 H, m, $C^{a3,a5}\boldsymbol{H}_{ax}$), 1.11-1.17 (3 H, m, $C^{41}\boldsymbol{H}'$ + $C^{a2,a6}\boldsymbol{H}_{ax}$ overlapped), 1.14-1.21 (2 H, m, $C^{3Pt}\boldsymbol{H}_{2/diastereotopic}$ R_N), 1.28 (3 H, s, $C^{46}\boldsymbol{H}_3$), 1.23 (3 H, br s, $C^{Pr3}\boldsymbol{H}_3$), 1.25-1.33 (1 H, m, $C^{42}\boldsymbol{H}'$), 1.33 (3 H, s, $C^{54}\boldsymbol{H}_3$), 1.35 (3 H, s, $C^{25}\boldsymbol{H}_3$), 1.36 (6 H, td, $C^{R6G}\boldsymbol{H}_{3(amine)}$), 1.38 (3 H, s, $C^{47}\boldsymbol{H}_3$), 1.50 (3 H, br t, $C^{aPt}C\boldsymbol{H}_3$ R_N), 1.52-1.57 (2 H, m, $C^{a3,a5}\boldsymbol{H}_{eq}$), 1.58-1.63 (2 H, m, $C^{42}\boldsymbol{H}''$ + $C^{55}\boldsymbol{H}'$ overlapped), 1.75 (1.2 H, br t, $C^{aPt}C\boldsymbol{H}_3$ S_N), 1.76-1.82 (3 H, m, $C^{41}\boldsymbol{H}''$ + $C^{4Pt}\boldsymbol{H}_{2/diastereotopic}$ R_N overlapped), 1.80-1.89 (4 H, m, $C^{48}\boldsymbol{H}_{2/diastereotopic}$ + $C^{30}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 1.87 (3 H, s, $C^{36}\boldsymbol{H}_3$), 1.91 (1.2 H, s, $C^{2Pt}C\boldsymbol{H}_3$ S_N), 1.93 (3 H, s, $C^{2Pt}C\boldsymbol{H}_3$ R_N), 1.93 (6 H, 2s, $C^{2,7R6G}C\boldsymbol{H}_3$), 1.99-2.09 (1 H, m, $C^{56}\boldsymbol{H}'$), 2.21-2.27 (3 H, m, $C^{a2,a6}\boldsymbol{H}_{eq}$ + $C^{6Pt}\boldsymbol{H}_{ax}$ R_N overlapped),

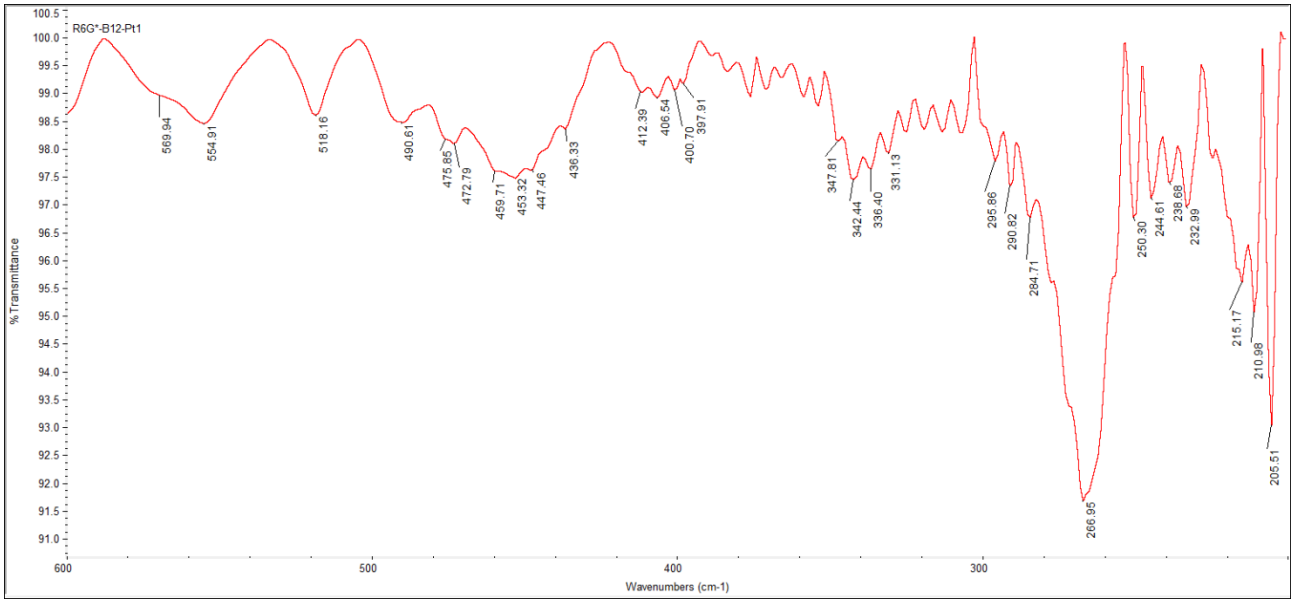
2.27 (3 H, s, $C^{B11}\boldsymbol{H}_3$), 2.30 (3 H, s, $C^{B10}\boldsymbol{H}_3$), 2.36-2.41 (3 H, m, $C^{37}\boldsymbol{H}_{2/diastereotopic}$ + $C^{26}\boldsymbol{H}'$ overlapped), 2.38-2.61 (8 H, m, $C^{5Pt}\boldsymbol{H}$ R_N + $C^{31}\boldsymbol{H}_{2/diastereotopic}$ + $C^{49}\boldsymbol{H}_{2/diastereotopic}$ + $C^{26}\boldsymbol{H}''$ + $C^{55}\boldsymbol{H}''$ + $C^{56}\boldsymbol{H}''$ overlapped), 2.51 (3 H, s, $C^{53}\boldsymbol{H}_3$), 2.61 (3 H, s, $C^{35}\boldsymbol{H}_3$), 2.68-2.74 (2 H, m, $C^{60}\boldsymbol{H}_{2/diastereotopic}$), 2.76-2.94 (2 H, m, $C^{18}\boldsymbol{H}$ + $C^{Pr1}\boldsymbol{H}'$ overlapped), 3.04-3.11 (1 H, m, $C^{a1}\boldsymbol{H}$), 3.16-3.24 (1 H, m, $C^{a4}\boldsymbol{H}$), 3.29-3.35 (1 H, m, $C^{13}\boldsymbol{H}$), 3.37-3.44 (1 H, m, $C^{6Pt}\boldsymbol{H}_{eq}$ R_N), 3.53 (4 H, qd, $C^{R6G}\boldsymbol{H}_{2(amine)}$), 3.57-3.68 (2 H, m, $C^{8}\boldsymbol{H}$ + $C^{Pr1}\boldsymbol{H}''$ overlapped), 3.94-4.01 (1 H, m, $C^{19}\boldsymbol{H}$), 4.07-4.10 (1 H, m, $C^{R5}\boldsymbol{H}'$), 4.10-4.25 (2 H, m, $C^{R4}\boldsymbol{H}$ + $C^{R2}\boldsymbol{H}$ overlapped), 4.34-4.38 (1 H, m, $C^{Pr2}\boldsymbol{H}$), 4.51-4.56 (1 H, m, $C^{R5}\boldsymbol{H}''$), 4.61-4.69 (1 H, m, $C^{R3}\boldsymbol{H}$), 4.63-4.69 (2 H, m, $C^{bPt}\boldsymbol{H}_{2/diastereotopic}$ R_N), 4.71-4.75 (1 H, m, $C^{3}\boldsymbol{H}$), 5.95 (1 H, s, $C^{10}\boldsymbol{H}$), 6.14-6.26 (1 H, m, $C^{R1}\boldsymbol{H}$), 6.49 (1 H, s, $C^{B4}\boldsymbol{H}$), 6.78-6.84 (1 H, m, $C^{6'R6G}\boldsymbol{H}$), 6.91 (2 H, s, $C^{4,5R6G}\boldsymbol{H}$), 7.02 (2 H, s, $C^{1,8R6G}\boldsymbol{H}$), 7.12 (1 H, s, $C^{B2}\boldsymbol{H}$), 7.32 (1 H, s, $C^{B7}\boldsymbol{H}$), 6.67-7.60 (7 H, m, $C_5\boldsymbol{H}_{5/PhPt}$ R_N + S_N overlapped), 7.72-7.80 (2 H, m, $C^{4'R6G}\boldsymbol{H}$ + $C^{5'R6G}\boldsymbol{H}$ overlapped), 7.83 (1 H, br s, $C^{3'R6G}\boldsymbol{H}$) ppm. $^{13}C\{^1H\}$ NMR (151 MHz; CD_3OD; 298 K): δ 14.1 ($\boldsymbol{C}^{R6G}H_{3(amine)}$), 16.3 ($\boldsymbol{C}^{53}H_3$), 16.8 ($\boldsymbol{C}^{35}H_3$), 17.3 ($\boldsymbol{C}^{25}H_3$), 17.5 ($\boldsymbol{C}^{54}H_3$), 17.6 ($C^{2,7R6G}\boldsymbol{C}H_3$), 20.1 ($\boldsymbol{C}^{36}H_3$), 20.2 ($\boldsymbol{C}^{Pr3}H_3$), 20.3 ($\boldsymbol{C}^{B11}H_3$), 20.4 ($\boldsymbol{C}^{47}H_3$), 20.8 ($\boldsymbol{C}^{20}H_3$), 20.9 ($\boldsymbol{C}^{B10}H_3$), 21.0 ($C^{aPt}\boldsymbol{C}H_3$ R_N), 21.5 ($C^{2Pt}\boldsymbol{C}H_3$ S_N), 21.6 ($C^{aPt}\boldsymbol{C}H_3$ S_N), 25.4 ($\boldsymbol{C}^{4Pt}H_2$ R_N), 26.1 ($\boldsymbol{C}^{41}H_2$), 26.2 ($C^{2Pt}\boldsymbol{C}H_3$ R_N), 27.2 ($\boldsymbol{C}^{30}H_2$), 29.0 ($\boldsymbol{C}^{48}H_2$), 29.8 ($\boldsymbol{C}^{a2,a6}H_2$), 30.0 ($\boldsymbol{C}^{6Pt}H_2$ R_N), 31.7 ($\boldsymbol{C}^{3Pt}H_2$ R_N), 31.9 ($\boldsymbol{C}^{42}H_2$ + $\boldsymbol{C}^{a3,a5}H_2$ + $\boldsymbol{C}^{55}H_2$ overlapped), 32.5 ($\boldsymbol{C}^{46}H_3$ + $\boldsymbol{C}^{56}H_2$ + $\boldsymbol{C}^{60}H_2$ overlapped), 35.2 ($\boldsymbol{C}^{49}H_2$), 36.1 ($\boldsymbol{C}^{31}H_2$), 39.4 ($\boldsymbol{C}^{R6G}H_{2(amine)}$ + $\boldsymbol{C}^{5Pt}H$ R_N overlapped), 39.5 ($\boldsymbol{C}^{18}H$), 43.4 ($\boldsymbol{C}^{37}H_2$), 43.5 ($\boldsymbol{C}^{26}H_2$), 46.6 ($\boldsymbol{C}^{Pr1}H_2$), ~49 ($\boldsymbol{C}^{2}$ + $\boldsymbol{C}^{12}$ overlapped with the residual peak of CD_3OD), 49.6 ($\boldsymbol{C}^{a4}H$), 50.5 ($\boldsymbol{C}^{7}$), 54.3 ($\boldsymbol{C}^{a1}H$), 54.4 ($\boldsymbol{C}^{13}H$), 57.2 ($\boldsymbol{C}^{8}H$), 57.5 ($\boldsymbol{C}^{3}H$), 60.5 ($\boldsymbol{C}^{17}$), 64.1 ($\boldsymbol{C}^{R5}H_2$), 67.5 ($\boldsymbol{C}^{9}$), 70.4 ($\boldsymbol{C}^{R2}H$), 73.3 ($\boldsymbol{C}^{R3}H$), 73.8 ($\boldsymbol{C}^{Pr2}H$), 76.1 ($\boldsymbol{C}^{2Pt}$ R_N), 76.8 ($\boldsymbol{C}^{19}H$), 81.6 ($\boldsymbol{C}^{R4}H$), 86.5 ($\boldsymbol{C}^{1}$), 88.5 ($\boldsymbol{C}^{R1}H$), 94.8 ($\boldsymbol{C}^{4,5R6G}H$), 95.6 ($\boldsymbol{C}^{10}H$), 105.3 ($\boldsymbol{C}^{15}$), 106.9 ($\boldsymbol{C}^{12,13R6G}$), 108.6 ($\boldsymbol{C}^{5}$), 111.3 ($\boldsymbol{C}^{bPt}H_2$ R_N), 113.0 ($\boldsymbol{C}^{B7}H$), 117.0 ($\boldsymbol{C}^{B4}H$), 118.9 ($\boldsymbol{C}^{2,7R6G}$), 123.1 ($\boldsymbol{C}^{3'R6G}H$), 125.4 ($\boldsymbol{C}$N), 126.5 ($\boldsymbol{C}^{1,8R6G}H$), 126.7 ($\boldsymbol{C}^{6'R6G}H$), 129.0-130.6 ($\boldsymbol{C}_5H_{5/PhPt}$ R_N), 131.3 ($\boldsymbol{C}^{4'R6G}H$), 131.6 ($\boldsymbol{C}^{B8}$), 133.0 ($\boldsymbol{C}^{2'R6G}$), 133.1 ($\boldsymbol{C}^{5'R6G}H$), 134.5 ($\boldsymbol{C}^{B5}$), 135.6 ($\boldsymbol{C}^{B6}$), 138.4 ($\boldsymbol{C}^{B9}$), 142.9 (i-$\boldsymbol{C}^{Pt}$ R_N), 143.1 ($\boldsymbol{C}^{B2}H$), 146.8 ($\boldsymbol{C}^{aPt}$ R_N), 148.4 ($\boldsymbol{C}^{3,6R6G}$), 157.6 ($\boldsymbol{C}^{11,14R6G}$), 158.5 ($\boldsymbol{C}^{1'}$), 166.8 ($\boldsymbol{C}^{14}$), 167.2 ($\boldsymbol{C}^{6}$), 169.3 ($\boldsymbol{C}^{R6G}$=O$_{(lactam)}$), 174.8 ($\boldsymbol{C}^{57}$), 175.1 ($\boldsymbol{C}^{38}$), 175.5 ($\boldsymbol{C}^{61}$), 175.6 ($\boldsymbol{C}^{27}$), 175.7 ($\boldsymbol{C}^{43}$), 176.4 ($\boldsymbol{C}^{32}$), 177.1 ($\boldsymbol{C}^{9}$), 177.3 ($\boldsymbol{C}^{R6G}$=O$_{(amide)}$), 177.8 ($\boldsymbol{C}^{50}$), 177.9 ($\boldsymbol{C}^{11}$), 178.7 ($\boldsymbol{C}^{1Pt}$ R_N), 180.6 ($\boldsymbol{C}^{16}$), 181.6 ($\boldsymbol{C}^{4}$) ppm. $^{31}P\{^1H\}$ NMR (162 MHz; CD_3OD; 298 K): δ -0.3 ($\boldsymbol{P}O_4^-$) ppm.

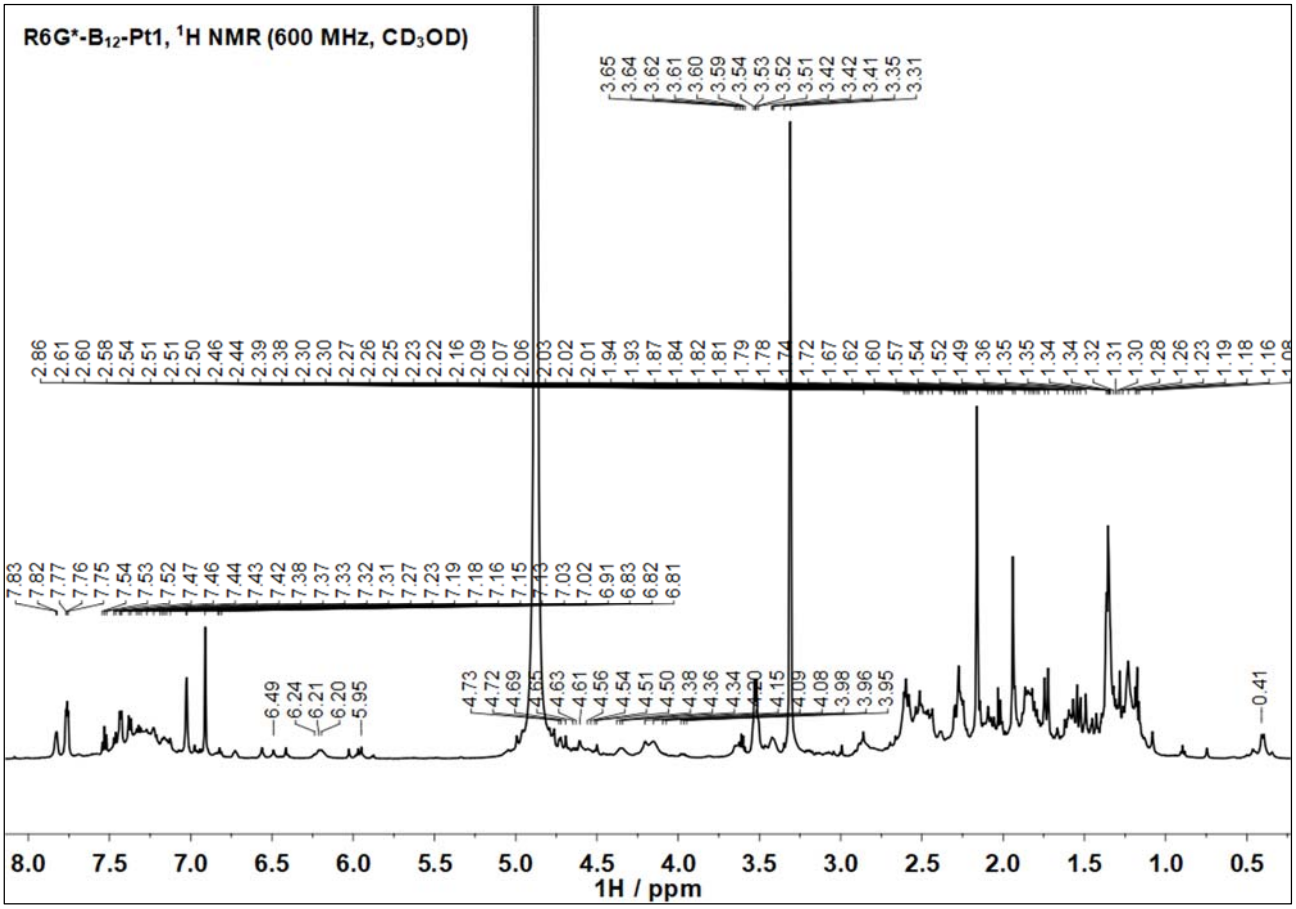


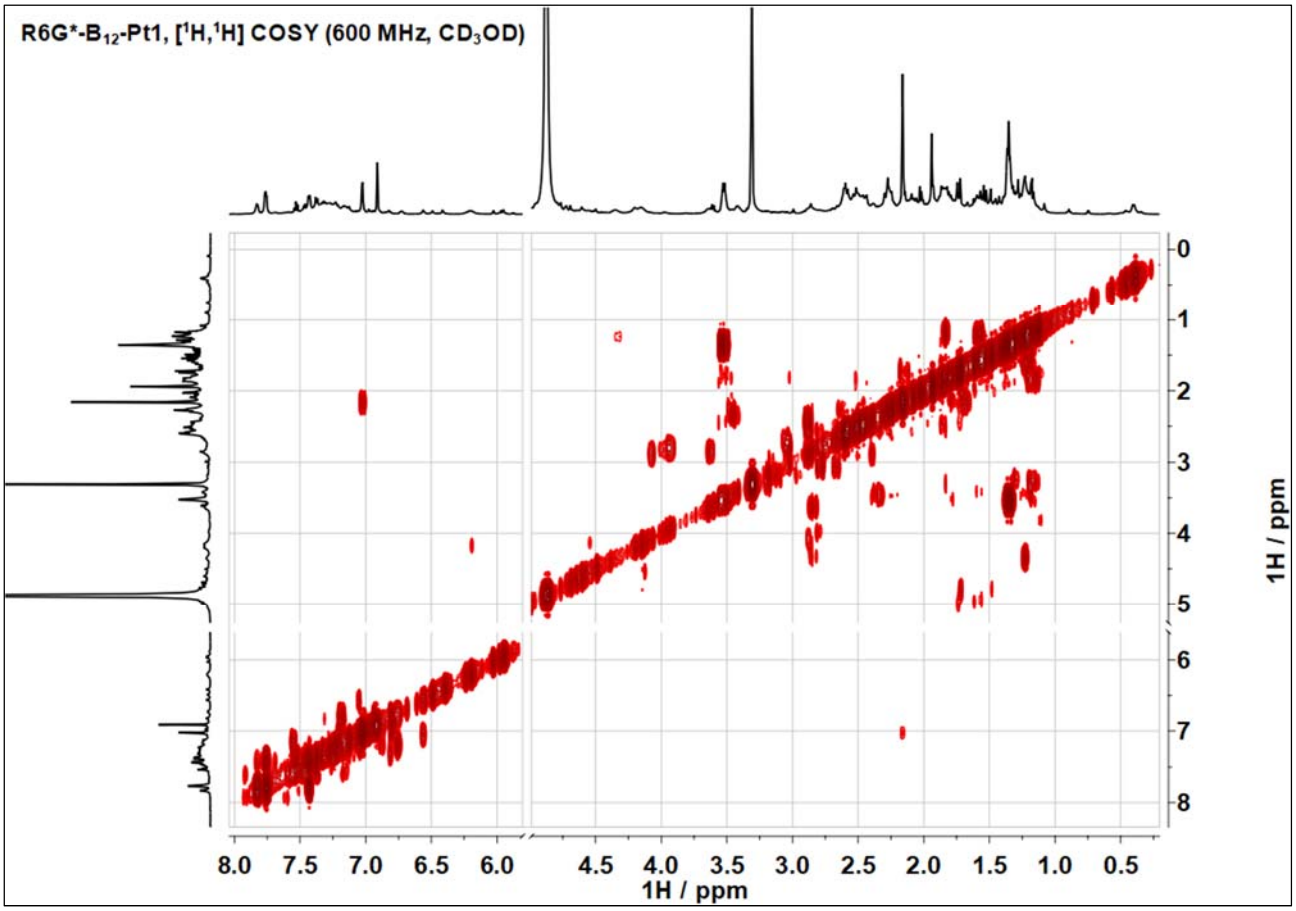
Peak List

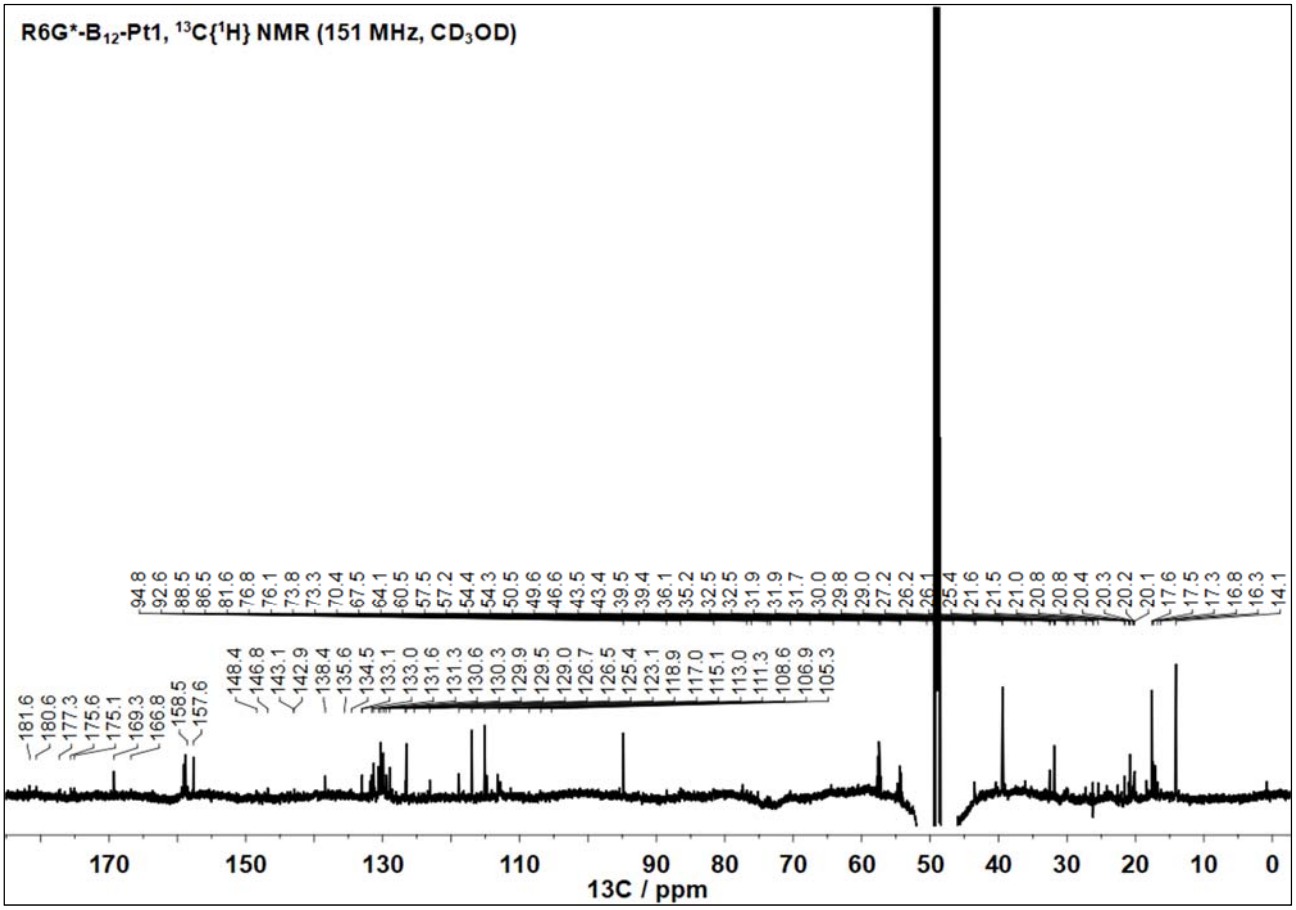
m/z	z	Abund
793.3029		1385792.75
793.6382	1	1914046.25
793.9718	1	1869493.63
794.3049	1	1638358
794.6378	1	1154626
1189.45	2	991210.75
1189.9517	2	1427130
1190.4524	2	1401596.13
1190.9521	2	1195008.13
1191.4527	2	799483.31

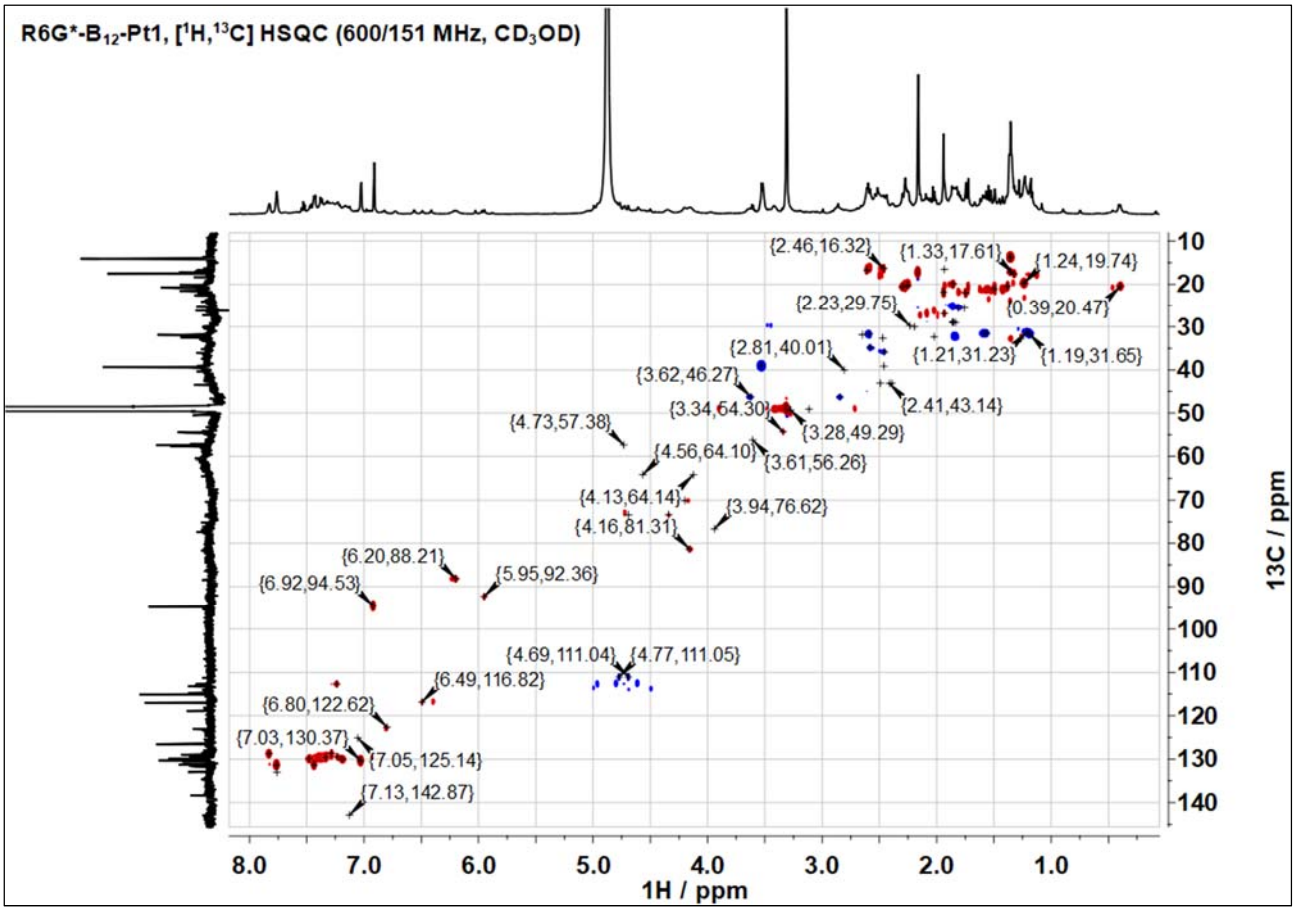


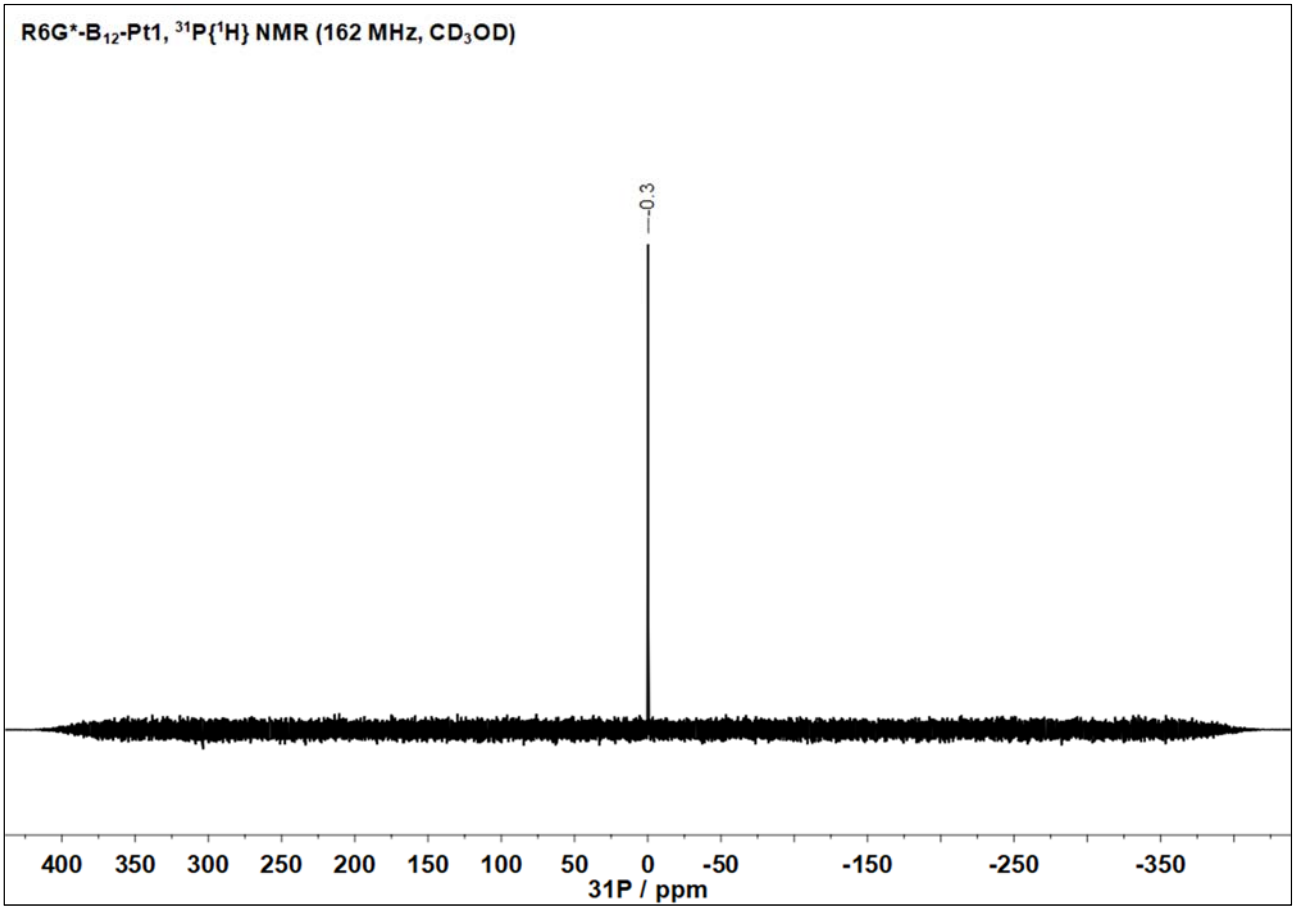



R6G*-B12-Pt1, 1H NMR (600 MHz, CD3OD)
1H / ppm


R6G*-B12-Pt1, [1H,1H] COSY (600 MHz, CD3OD)
1H / ppm
1H / ppm

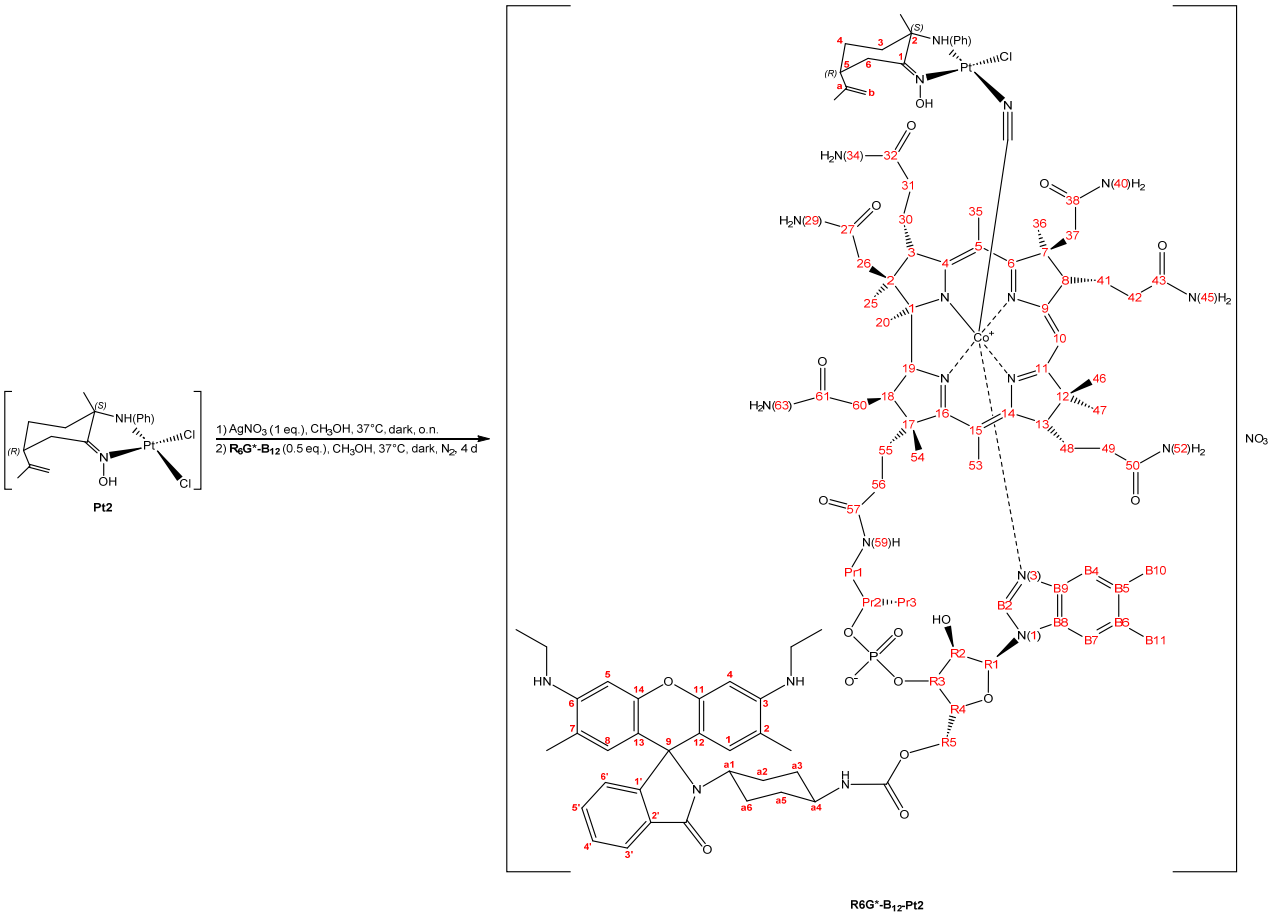

R6G*-B12-Pt1, 13C{1H} NMR (151 MHz, CD3OD)
94.8 92.6 88.5 86.5 81.6 76.8 76.1 73.8 73.3 70.4 67.5 64.1 60.5 57.5 57.2 54.4 54.3 50.5 49.6 46.6 43.5 43.4 39.5 39.4 36.1 35.2 32.5 32.5 31.9 31.9 31.7 30.0 29.8 29.0 27.2 26.2 26.1 25.4 21.6 21.5 21.0 20.8 20.4 20.3 20.2 20.1 17.6 17.5 17.3 16.8 16.3 14.1
148.4 146.8 143.1 142.9 138.4 135.6 134.5 133.1 133.0 131.6 131.3 130.6 130.3 129.9 129.5 129.0 126.7 126.5 125.4 123.1 118.9 117.0 115.1 113.0 111.3 108.6 106.9 105.3
181.6 180.6 177.3 175.6 175.1 169.3 166.8 158.5 157.6
170 150 130 110 90 80 70 60 50 40 30 20 10 0
13C / ppm


R6G*-B12-Pt1, [1H,13C] HSQC (600/151 MHz, CD3OD)
{2.46,16.32}
{1.33,17.61}
{1.24,19.74}
{2.23,29.75}
{0.39,20.47}
{2.81,40.01}
{1.21,31.23}
{1.19,31.65}
{3.62,46.27}
{2.41,43.14}
{3.34,64.30}
{3.28,49.29}
{4.73,57.38}
{4.56,64.10}
{3.61,56.26}
{4.13,64.14}
{3.94,76.62}
{4.16,81.31}
{6.20,88.21}
{6.92,94.53}
{5.95,92.36}
{4.69,111.04}
{4.77,111.05}
{6.49,116.82}
{6.80,122.62}
{7.03,130.37}
{7.05,125.14}
{7.13,142.87}
10 20 30 40 50 60 70 80 90 100 110 120 130 140
13C / ppm
8.0 7.0 6.0 5.0 4.0 3.0 2.0 1.0
1H / ppm



R6G*-B_{12}-Pt2

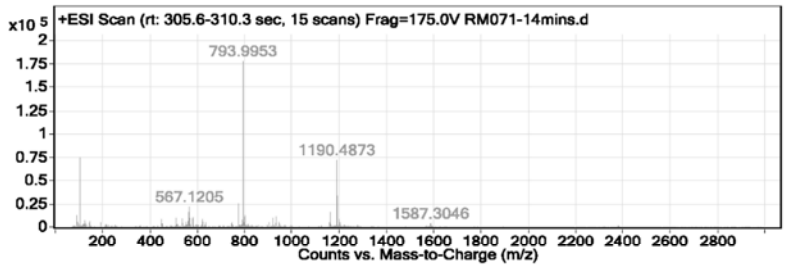
A water solution (1 mL) of $AgNO_3$ (0.008 g, 0.05 mmol) was added dropwise to a methanol solution (4 mL) of **Pt2** (0.025 g, 0.05 mmol) and the mixture was stirred at 37°C overnight in the dark. The off-white insoluble residue (AgCl) was filtered off and the resulting clear pale yellow filtrate was added dropwise to a methanol solution (4 mL) of the rhodamine 6G–*trans*-1,4-diaminocyclohexane–vitamin B_{12} conjugate (**R6G*-B_{12}**, 0.031 g, 0.02 mmol). The mixture was stirred at 37°C for 4 d in the dark under an inert atmosphere of nitrogen and the solvent was subsequently evaporated to dryness. The crude product was then purified by semi-preparative reversed-phase HPLC (column: Kromasil 100-5-C18, 10×250 mm, 5 µm particle size) over 40 min at a flow rate of 3.5 mL min^{-1} (solvent A: H_2O + 0.1% TFA; solvent B: CH_3CN + 0.1% TFA; 0-5 min: isocratic flow of 60:40 parts A:B; 5-30 min linear gradient to 30:70 parts A:B; 30-35 min: isocratic flow of 30:70 parts A:B; 35-40 min: isocratic flow of 60:40 parts A:B). The desired compound was eluted at a retention time of 14 min. The solvent was then evaporated to dryness and the residue was dried under vacuum over P_2O_5, yielding a dark pink solid identified as a mixture of S_N and R_N epimers (S_N:R_N ratio of *ca.* 2:1 in CD_3OD as estimated by ^1H NMR spectroscopy) of the title compound (MM($C_{112}H_{146}ClCoN_{21}O_{21}PPt$) = 2442.97 g mol^{-1}, 0.018 g, 37% yield).



HRMS (CH_3OH, *m/z*) calcd. for $[M\text{-}NO_3{+}H]^{2+}$: 1190.4815; found: 1190.4873; $[M\text{-}NO_3{+}2H]^{3+}$: 793.9901; found: 793.9953. FT-IR (CsI disk; 298 K): $\tilde{\nu}_{max}$ 3393/3196 ($\nu_{a,s}(NH_2)_{amide}$ + $\nu(NH)_{amide}$ + $\nu(NH)_{amine}$ + $\nu(OH)_{oxime}$ overlapped), 2199 ($\nu(C{\equiv}N)$), 1674 ($\nu(C{=}O)$/amide I + $(\nu(C{=}O)_{lactam}$ + $\nu(C{=}C)_{alkene}$ overlapped), 1622 ($\delta(NH_2)$/amide II), 1608 ($\nu(C{=}N)_{oxime}$), 1577/1545 (corrin ring breathing mode), 1530/1500 ($\delta(C{-}N{-}H)_{amine}$ + $\delta_{ip}(C{-}N{-}H)$/amide II + $\nu(C{=}C)$ + $\nu(C{=}N)$ overlapped), 1410 ($\nu_a(NO_3^-)$, 1353 ($\nu(N{-}Ph)_{amine}$), 1265 ($\nu(C{-}O{-}C)_{xanthene}$), 1243 ($\nu(Ph{-}N)$), 1137 ($\nu_{oop}(PO_2^-)$), 1075 ($\nu_{ip}(PO_2^-)$), 1000 ($\nu(P{-}O{-}C)$), 902 ($\nu(N{-}OH)_{oxime}$), 836 ($w(NO_3^-)$, 561 ($\delta(Co{-}C{\equiv}N)$), 488 ($\nu(Co{-}N)$), 458 ($\nu(Pt{-}NOH)$), 434 ($\nu(Pt{-}NC)$, 401 ($\nu(Co{-}CN)$), 340 ($\nu(Pt{-}NPh)$ + $\nu(Co{-}N)$ overlapped), 333 ($\nu(Pt{-}Cl)$) cm^{-1}. ^1H NMR (400 MHz; CD_3OD; 298 K): δ 0.44 (3 H, s, $C^{20}\boldsymbol{H}_3$), 1.16-1.19 (2 H, m, $C^{a3,a5}\boldsymbol{H}_{ax}$), 1.11-1.17 (3 H, m, $C^{41}\boldsymbol{H}'$ + $C^{a2,a6}\boldsymbol{H}_{ax}$ overlapped), 1.14-1.21 (2 H, m, $C^{3Pt}\boldsymbol{H}_{2/diastereotopic}$ S_N), 1.23 (3 H, s, $C^{46}\boldsymbol{H}_3$), 1.24 (3 H, br s, $C^{Pr3}\boldsymbol{H}_3$), 1.25-1.33 (1 H, m, $C^{42}\boldsymbol{H}'$), 1.31 (3 H, s, $C^{54}\boldsymbol{H}_3$), 1.33 (3 H, s, $C^{25}\boldsymbol{H}_3$), 1.36 (6 H, td, $C^{R6G}\boldsymbol{H}_{3(amine)}$), 1.40 (3 H, s, $C^{47}\boldsymbol{H}_3$), 1.48 (3 H, br t, $C^{aPt}C\boldsymbol{H}_3$ S_N), 1.52-1.57 (2 H, m, $C^{a3,a5}\boldsymbol{H}_{eq}$), 1.58-1.63 (2 H, m, $C^{42}\boldsymbol{H}''$ + $C^{55}\boldsymbol{H}'$ overlapped), 1.74 (1.1 H, br t, $C^{aPt}C\boldsymbol{H}_3$ R_N), 1.76-1.82 (3 H, m, $C^{41}\boldsymbol{H}''$ + $C^{4Pt}\boldsymbol{H}_{2/diastereotopic}$ S_N overlapped), 1.80-1.90 (4 H, m, $C^{48}\boldsymbol{H}_{2/diastereotopic}$ + $C^{30}\boldsymbol{H}_{2/diastereotopic}$ overlapped), 1.85 (3 H, s, $C^{36}\boldsymbol{H}_3$), 1.93 (10.1 H, br s, $C^{2Pt}C\boldsymbol{H}_3$ R_N + $C^{2Pt}C\boldsymbol{H}_3$ S_N + $C^{2,7R6G}C\boldsymbol{H}_3$ overlapped), 1.99-2.09 (1 H, m, $C^{56}\boldsymbol{H}'$), 2.21-2.27 (3 H, m, $C^{a2,a6}\boldsymbol{H}_{eq}$ + $C^{6Pt}\boldsymbol{H}_{ax}$ S_N overlapped), 2.28 (3 H, s, $C^{B11}\boldsymbol{H}_3$), 2.31 (3 H, s, $C^{B10}\boldsymbol{H}_3$), 2.36-2.41 (3 H, m,

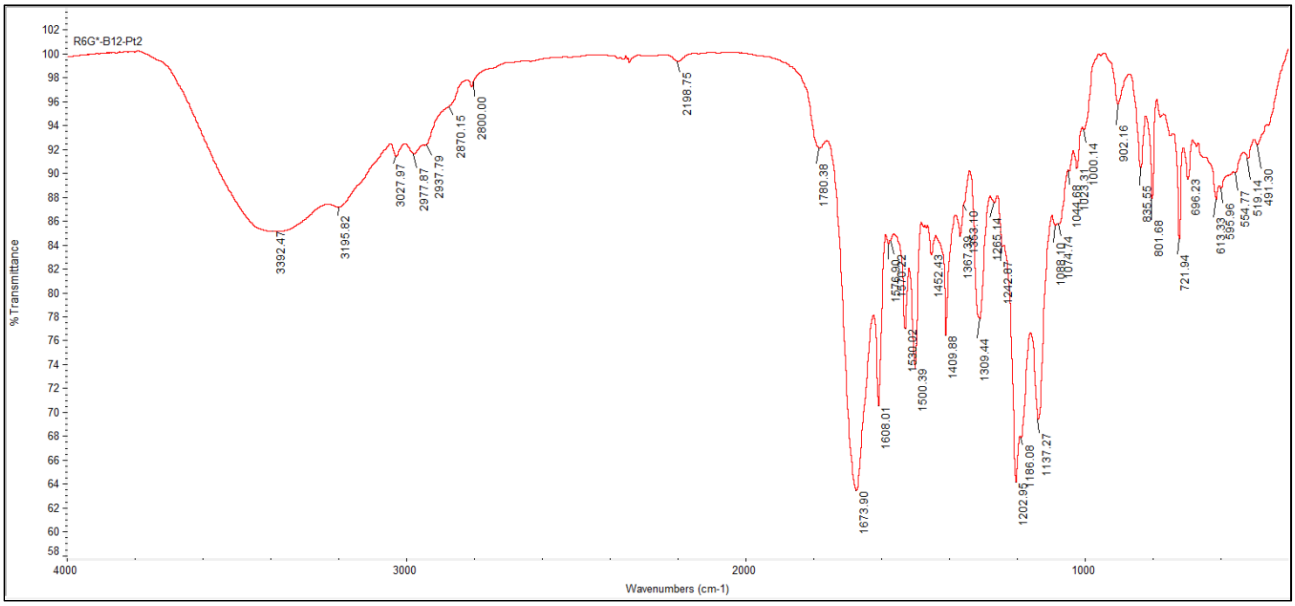
$C^{37}\boldsymbol{H}_{2/\text{diastereotopic}}$ + $C^{26}\boldsymbol{H}'$ overlapped), 2.36-2.60 (8 H, m, $C^{5Pt}\boldsymbol{H}$ R_N + $C^{31}\boldsymbol{H}_{2/\text{diastereotopic}}$ + $C^{49}\boldsymbol{H}_{2/\text{diastereotopic}}$ + $C^{26}\boldsymbol{H}''$ + $C^{55}\boldsymbol{H}''$ + $C^{56}\boldsymbol{H}''$ overlapped), 2.58 (3 H, s, $C^{53}\boldsymbol{H}_3$), 2.66 (3 H, s, $C^{35}\boldsymbol{H}_3$), 2.68-2.74 (2 H, m, $C^{60}\boldsymbol{H}_{2/\text{diastereotopic}}$), 2.76-2.94 (2 H, m, $C^{18}\boldsymbol{H}$ + $C^{Pr1}\boldsymbol{H}'$ overlapped), 3.12-3.15 (1 H, m, $C^{a1}\boldsymbol{H}$), 3.16-3.26 (1 H, m, $C^{a4}\boldsymbol{H}$), 3.29-3.35 (1 H, m, $C^{13}\boldsymbol{H}$), 3.37-3.44 (1 H, m, $C^{6Pt}\boldsymbol{H}_{eq}$ S_N), 3.53 (4 H, qd, $C^{R6G}\boldsymbol{H}_{2(\text{amine})}$), 3.57-3.68 (2 H, m, $C^{8}\boldsymbol{H}$ + $C^{Pr1}\boldsymbol{H}''$ overlapped), 3.94-4.01 (1 H, m, $C^{19}\boldsymbol{H}$), 4.07-4.10 (1 H, m, $C^{R5}\boldsymbol{H}'$), 4.10-4.25 (2 H, m, $C^{R4}\boldsymbol{H}$ + $C^{R2}\boldsymbol{H}$ overlapped), 4.33-4.38 (1 H, m, $C^{Pr2}\boldsymbol{H}$), 4.51-4.55 (1 H, m, $C^{R5}\boldsymbol{H}''$), 4.60-4.63 (1 H, m, $C^{R3}\boldsymbol{H}$), 4.59-4.71 (2 H, m, $C^{bPt}\boldsymbol{H}_{2/\text{diastereotopic}}$ S_N), 4.75-4.80 (1 H, m, $C^{3}\boldsymbol{H}$), 5.96 (1 H, s, $C^{10}\boldsymbol{H}$), 6.21 (1 H, br s, $C^{R1}\boldsymbol{H}$), 6.59 (1 H, s, $C^{B4}\boldsymbol{H}$), 6.88-6.2 (1 H, m, $C^{6'R6G}\boldsymbol{H}$), 6.92 (2 H, s, $C^{4,5R6G}\boldsymbol{H}$), 7.03 (2 H, d, $C^{1,8R6G}\boldsymbol{H}$), 7.12 (1 H, s, $C^{B2}\boldsymbol{H}$), 7.24 (1 H, s, $C^{B7}\boldsymbol{H}$), 7.10-7.50 (7 H, m, $C_5\boldsymbol{H}_{5/\text{PhPt}}$ S_N + R_N overlapped), 7.75-7.78 (2 H, m, $C^{4'R6G}\boldsymbol{H}$ + $C^{5'R6G}\boldsymbol{H}$ overlapped), 7.82-7.84 (1 H, m, $C^{3'R6G}\boldsymbol{H}$) ppm. $^{13}C\{^1H\}$ NMR (100 MHz; CD_3OD; 298 K): δ 14.1 ($\boldsymbol{C}^{R6G}H_{3(\text{amine})}$), 16.4 ($\boldsymbol{C}^{53}H_3$), 16.6 ($\boldsymbol{C}^{35}H_3$), 17.4 ($\boldsymbol{C}^{25}H_3$), 17.5 ($\boldsymbol{C}^{54}H_3$), 17.6 ($C^{2,7R6G}\boldsymbol{C}H_3$), 20.1 ($\boldsymbol{C}^{36}H_3$), 20.3 ($\boldsymbol{C}^{Pr3}H_3$), 20.4 ($\boldsymbol{C}^{B11}H_3$), 20.7 ($\boldsymbol{C}^{47}H_3$), 20.9 ($\boldsymbol{C}^{20}H_3$), 21.0 ($\boldsymbol{C}^{B10}H_3$), 21.6 ($C^{aPt}\boldsymbol{C}H_3$ S_N), 21.6 ($C^{2Pt}\boldsymbol{C}H_3$ R_N), 21.7 ($C^{aPt}\boldsymbol{C}H_3$ R_N), 25.5 ($\boldsymbol{C}^{4Pt}H_2$ S_N), 26.6 ($\boldsymbol{C}^{41}H_2$), 27.2 ($C^{2Pt}\boldsymbol{C}H_3$ S_N), 27.3 ($\boldsymbol{C}^{30}H_2$), 29.3 ($\boldsymbol{C}^{48}H_2$), 30.0 ($\boldsymbol{C}^{a2,a6}H_2$), 30.3 ($\boldsymbol{C}^{6Pt}H_2$ S_N), 31.5 ($\boldsymbol{C}^{3Pt}H_2$ S_N), 31.8 ($\boldsymbol{C}^{42}H_2$ + $\boldsymbol{C}^{a3,a5}H_2$ + $\boldsymbol{C}^{55}H_2$ overlapped), 32.5 ($\boldsymbol{C}^{46}H_3$ + $\boldsymbol{C}^{56}H_2$ + $\boldsymbol{C}^{60}H_2$ overlapped), 35.2 ($\boldsymbol{C}^{49}H_2$), 36.2 ($\boldsymbol{C}^{31}H_2$), 39.4 ($\boldsymbol{C}^{R6G}H_{2(\text{amine})}$ + $\boldsymbol{C}^{5Pt}H$ R_N overlapped), 39.5 ($\boldsymbol{C}^{18}H$), 43.2 ($\boldsymbol{C}^{37}H_2$), 43.9 ($\boldsymbol{C}^{26}H_2$), 46.6 ($\boldsymbol{C}^{Pr1}H_2$), ~49 ($\boldsymbol{C}^{2}$ + $\boldsymbol{C}^{12}$ overlapped with the residual peak of CD_3OD), 50.0 ($\boldsymbol{C}^{a4}H$), 50.4 ($\boldsymbol{C}^{7}$), 54.2 ($\boldsymbol{C}^{a1}H$), 54.5 ($\boldsymbol{C}^{13}H$), 56.8 ($\boldsymbol{C}^{8}H$), 57.3 ($\boldsymbol{C}^{3}H$), 60.7 ($\boldsymbol{C}^{17}$), 64.5 ($\boldsymbol{C}^{R5}H_2$), 67.7 ($\boldsymbol{C}^{9}$), 70.4 ($\boldsymbol{C}^{R2}H$), 73.3 ($\boldsymbol{C}^{R3}H$), 73.8 ($\boldsymbol{C}^{Pr2}H$), 76.1 ($\boldsymbol{C}^{2Pt}$ S_N), 76.9 ($\boldsymbol{C}^{19}H$), 81.6 ($\boldsymbol{C}^{R4}H$), 86.5 ($\boldsymbol{C}^{1}$), 88.5 ($\boldsymbol{C}^{R1}H$), 94.8 ($\boldsymbol{C}^{4,5R6G}H$), 95.5 ($\boldsymbol{C}^{10}H$), 105.5 ($\boldsymbol{C}^{15}$), 106.8 ($\boldsymbol{C}^{12,13R6G}$), 108.7 ($\boldsymbol{C}^{5}$), 111.8 ($\boldsymbol{C}^{bPt}H_2$ S_N), 114.6 ($\boldsymbol{C}^{B7}H$), 117.4 ($\boldsymbol{C}^{B4}H$), 120.3 ($\boldsymbol{C}^{2,7R6G}$), 123.5 ($\boldsymbol{C}^{3'R6G}H$), 124.0 ($\boldsymbol{C}N$), 126.5 ($\boldsymbol{C}^{1,8R6G}H$), 126.6 ($\boldsymbol{C}^{6'R6G}H$), 129.0-130.6 ($\boldsymbol{C}_5H_{5/\text{PhPt}}$ S_N), 131.3 ($\boldsymbol{C}^{4'R6G}H$), 131.8 ($\boldsymbol{C}^{B8}$), 133.0 ($\boldsymbol{C}^{2'R6G}$), 133.1 ($\boldsymbol{C}^{5'R6G}H$), 134.6 ($\boldsymbol{C}^{B5}$), 136.3 ($\boldsymbol{C}^{B6}$), 138.4 ($\boldsymbol{C}^{B9}$), 142.6 (i-$\boldsymbol{C}^{Pt}$ S_N), 142.8 ($\boldsymbol{C}^{B2}H$), 147.0 ($\boldsymbol{C}^{aPt}$ S_N), 148.8 ($\boldsymbol{C}^{3,6R6G}$), 157.6 ($\boldsymbol{C}^{11,14R6G}$), 158.9 ($\boldsymbol{C}^{1'}$), 166.7 ($\boldsymbol{C}^{14}$), 167.1 ($\boldsymbol{C}^{6}$), 169.3 ($\boldsymbol{C}^{R6G}{=}O_{(\text{lactam})}$), 174.3 ($\boldsymbol{C}^{57}$), 175.1 ($\boldsymbol{C}^{38}$), 175.6 ($\boldsymbol{C}^{61}$), 175.6 ($\boldsymbol{C}^{27}$), 175.7 ($\boldsymbol{C}^{43}$), 176.3 ($\boldsymbol{C}^{32}$), 177.2 ($\boldsymbol{C}^{9}$), 177.3 ($\boldsymbol{C}^{R6G}{=}O_{(\text{amide})}$), 177.4 ($\boldsymbol{C}^{50}$), 178.2 ($\boldsymbol{C}^{11}$), 178.9 ($\boldsymbol{C}^{1Pt}$ S_N), 180.8 ($\boldsymbol{C}^{16}$), 181.6 ($\boldsymbol{C}^{4}$) ppm. $^{31}P\{^1H\}$ NMR (162 MHz; CD_3OD; 298 K): δ -0.4 ($\boldsymbol{P}O_4^-$) ppm.

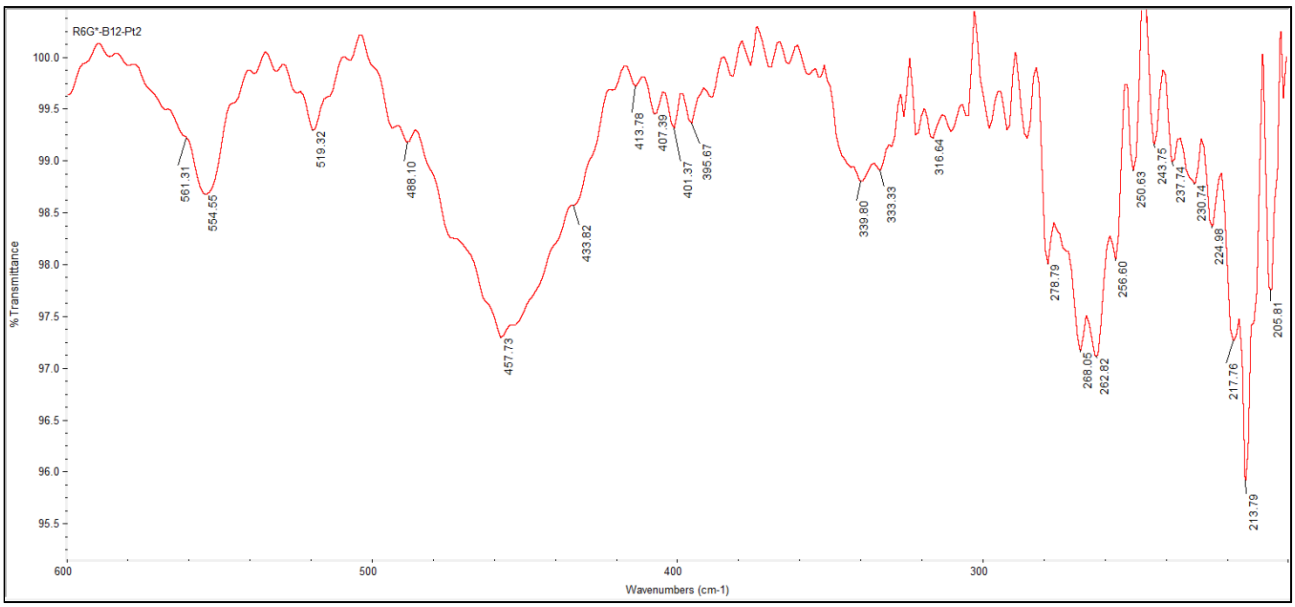
Spectrum Source	Fragmentor Voltage	Collision Energy	Ionization Mode
Peak (7) in "+ BPC Scan"	175	0	ESI

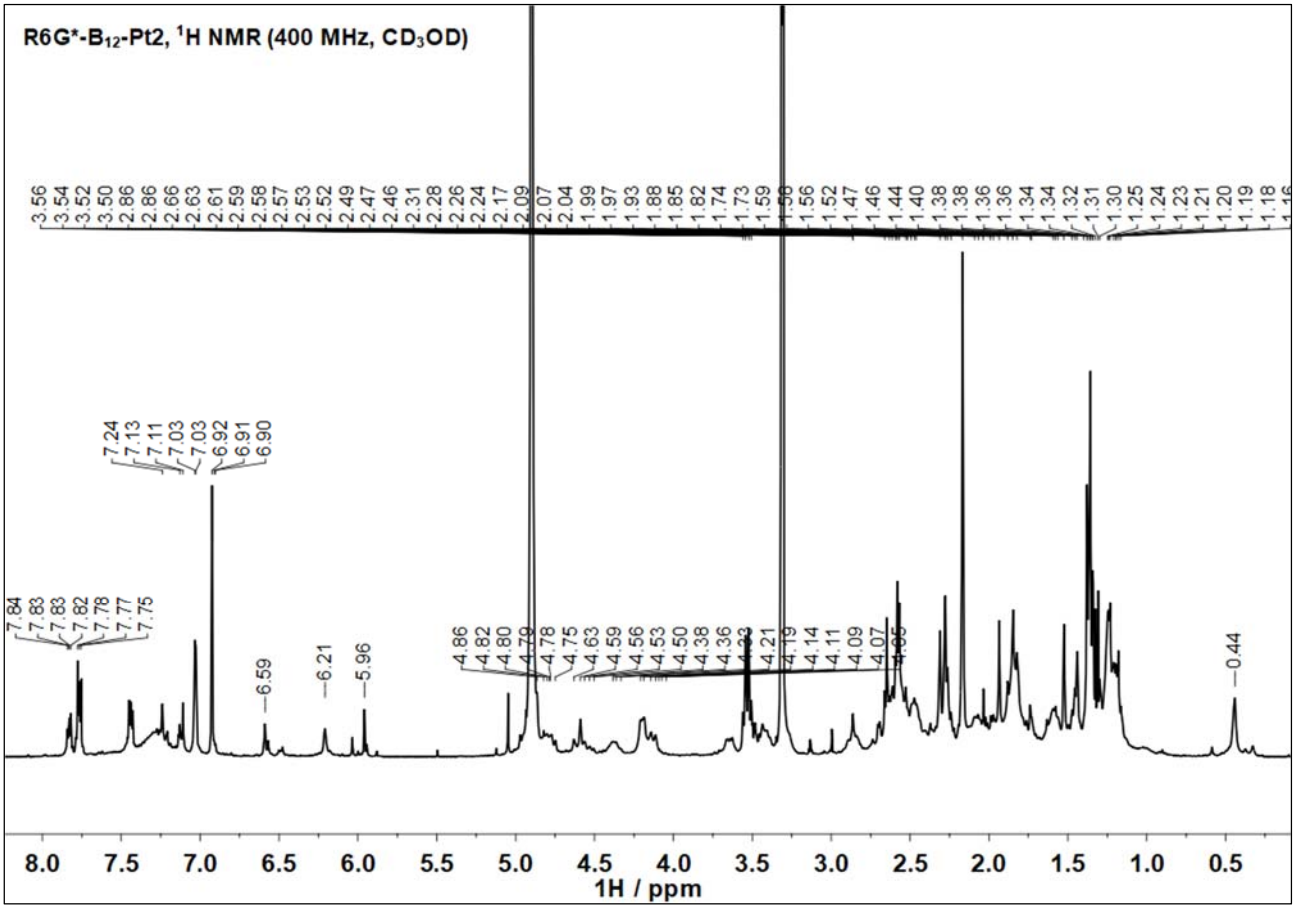


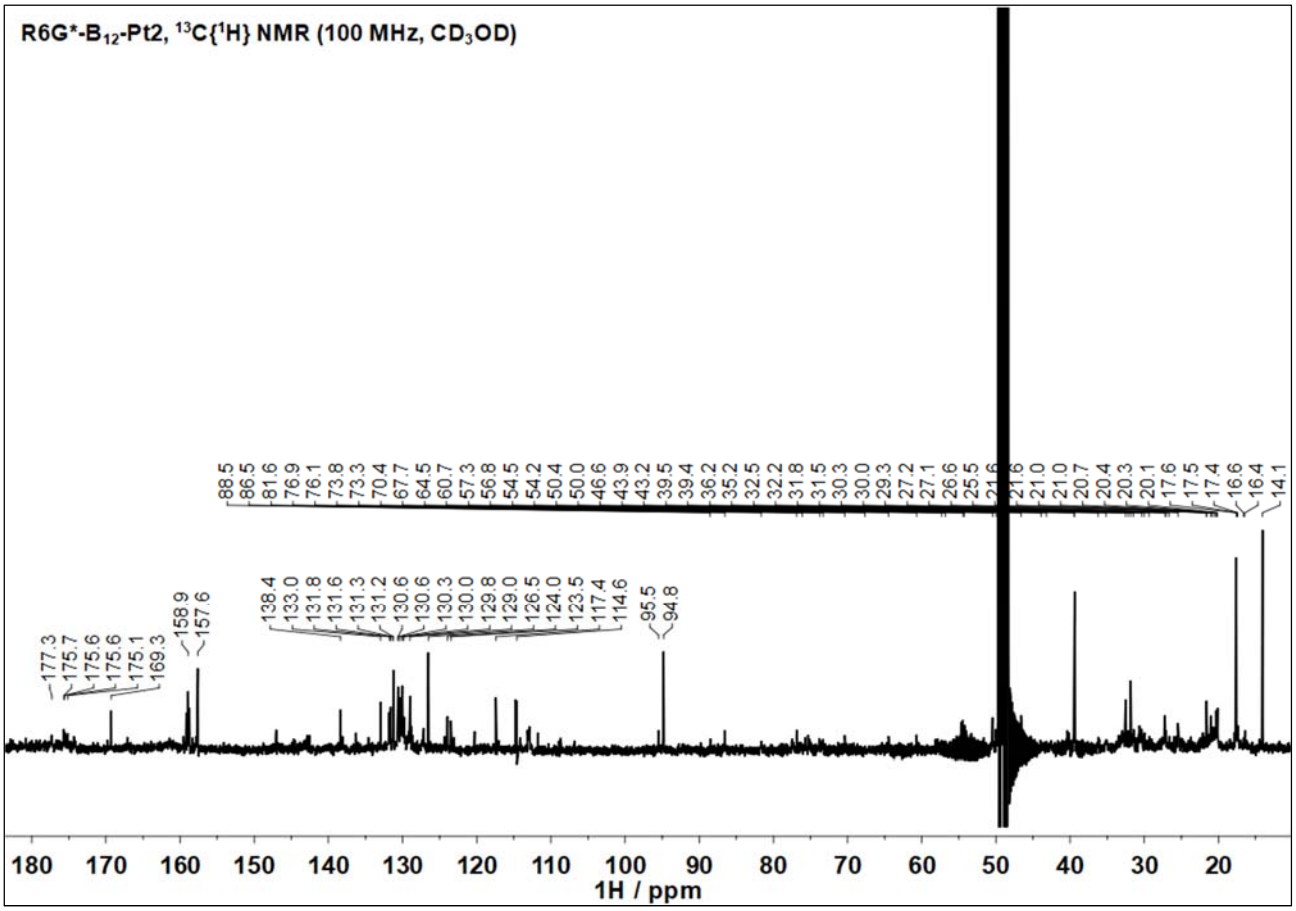
Peak List

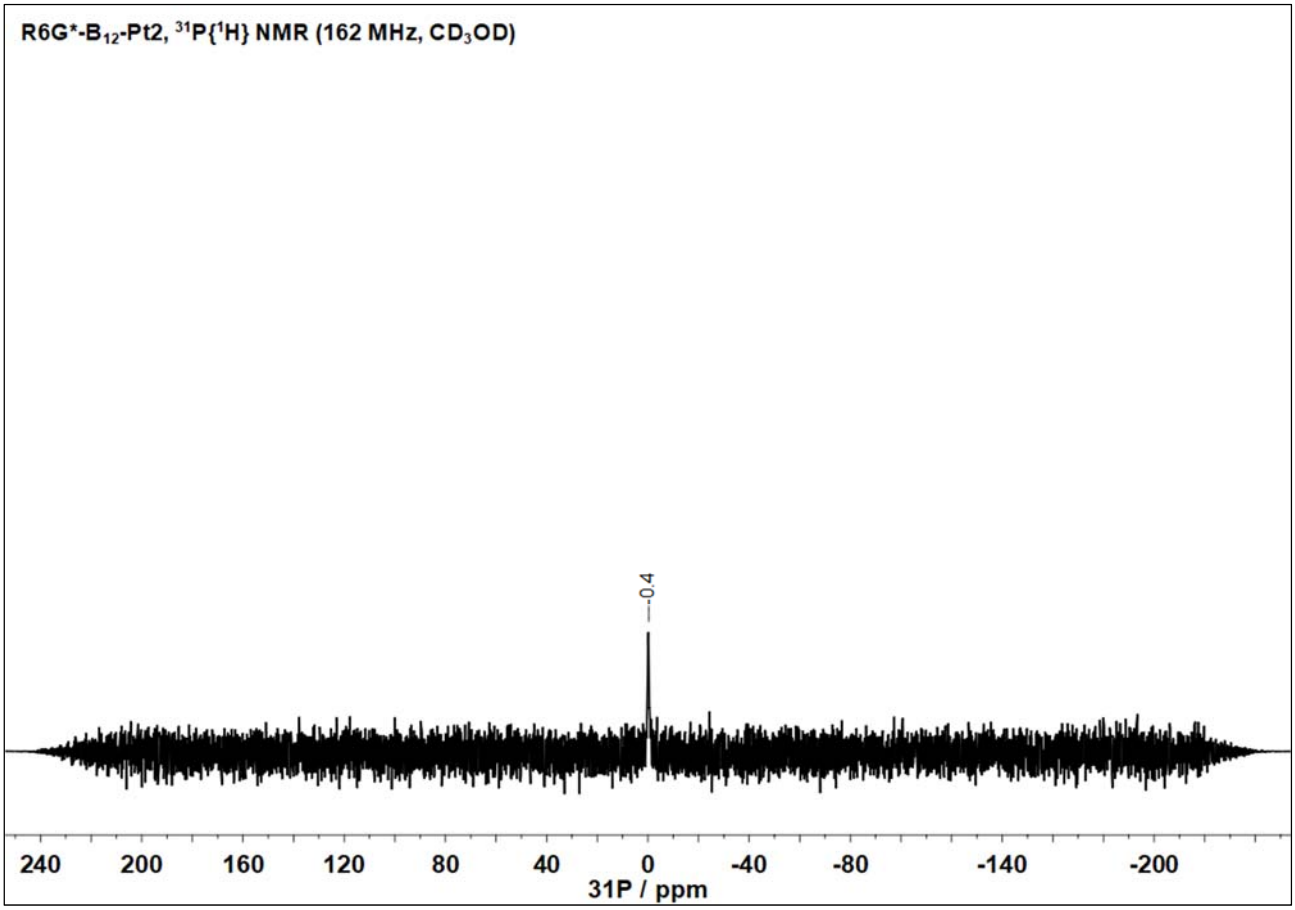
m/z	z	Abund
103.9559		74638.33
793.3252		57323.81
793.661		123466.39
793.9953	1	178119.47
794.3295	1	170561.91
794.6633	1	130580.73
794.9963	1	87339.17
1190.4873	2	72165.13
1190.9884	2	69283.36
1191.4879	2	54647.9



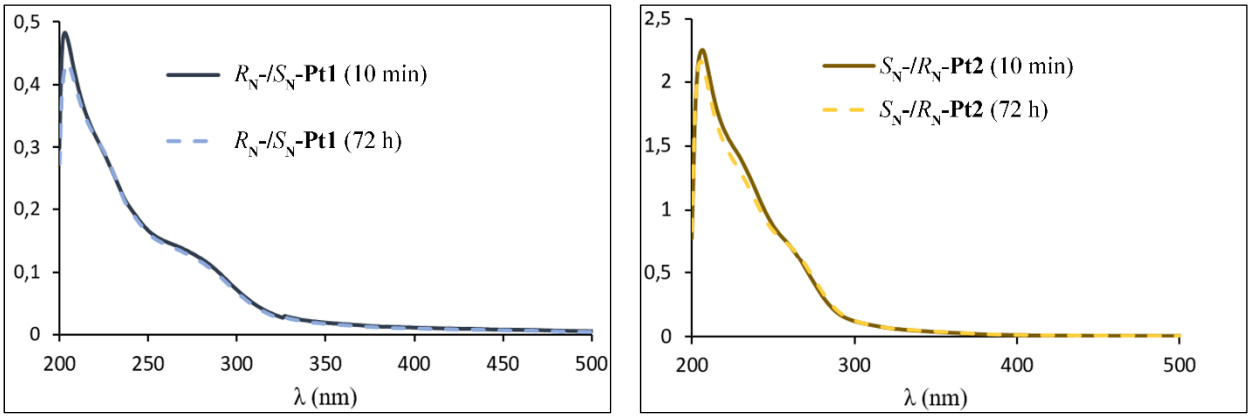



R6G*-B12-Pt2, 1H NMR (400 MHz, CD3OD)
1H / ppm

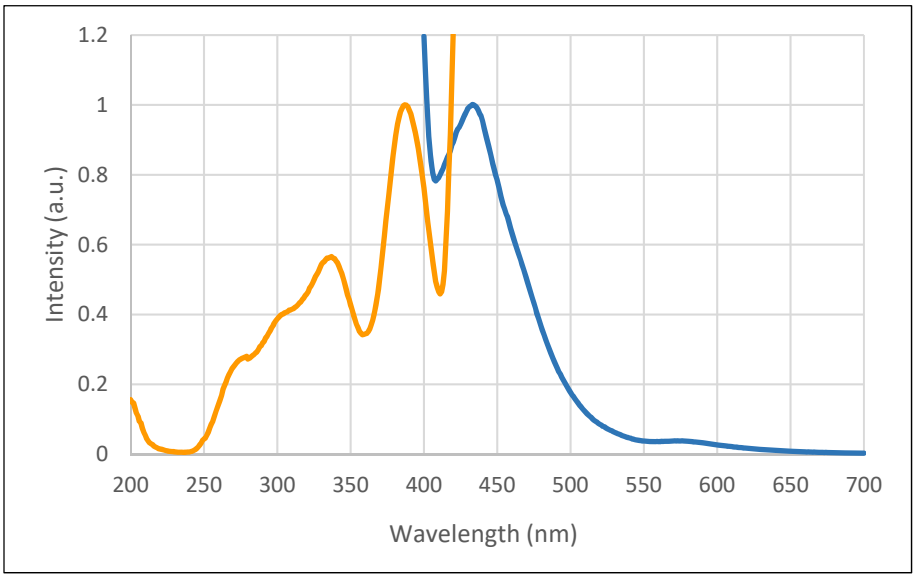

R6G*-B12-Pt2, 13C{1H} NMR (100 MHz, CD3OD)
1H / ppm


R6G*-B12-Pt2, 31P{1H} NMR (162 MHz, CD3OD)
-0.4
240
200
160
120
80
40
0
-40
-80
-140
-200
31P / ppm

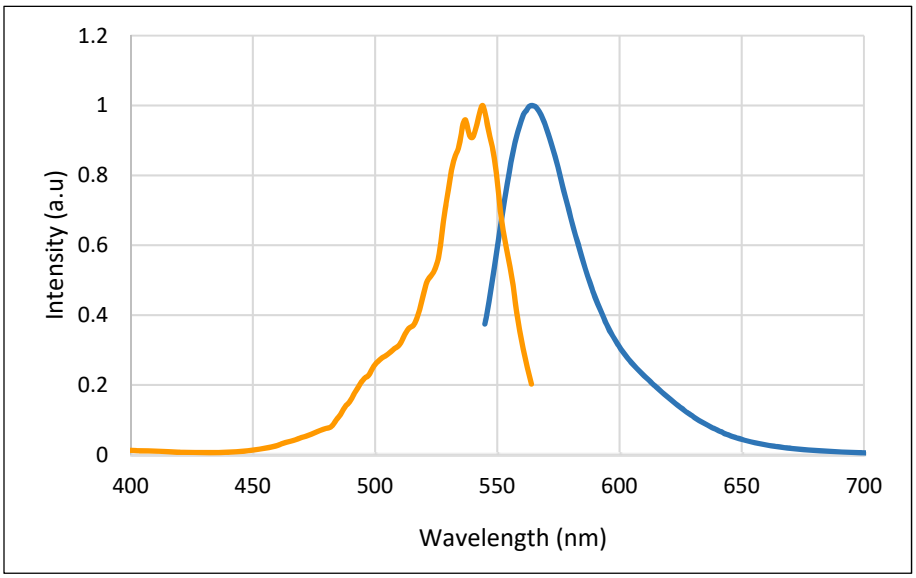
Absorption and emission spectra



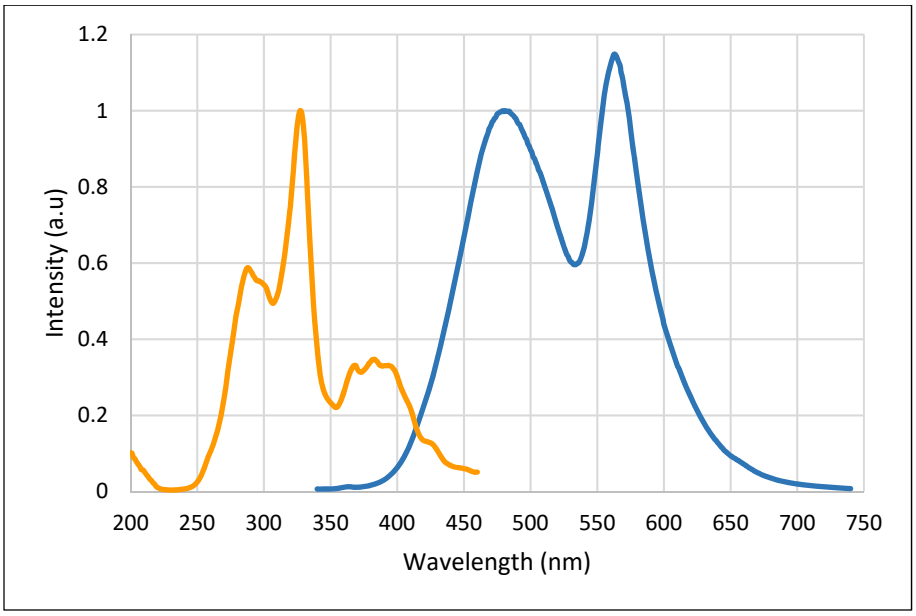
UV-Vis spectra of complexes **Pt1** and **Pt2** in water at room temperature over 72 h.



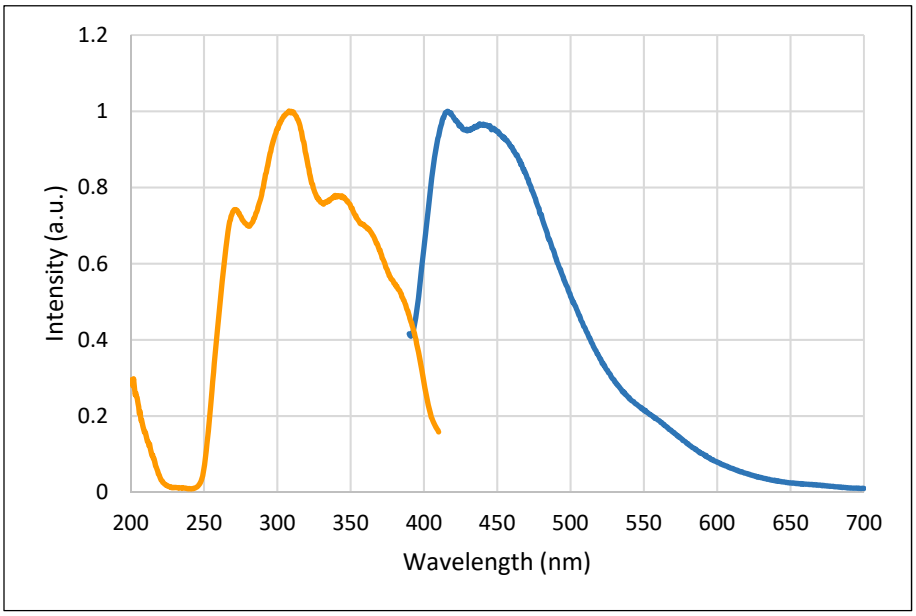
Absorption (orange) and emission (blue) spectra of vitamin B_{12} 25 µM in DMSO. Intensities have been normalized to the maxima.



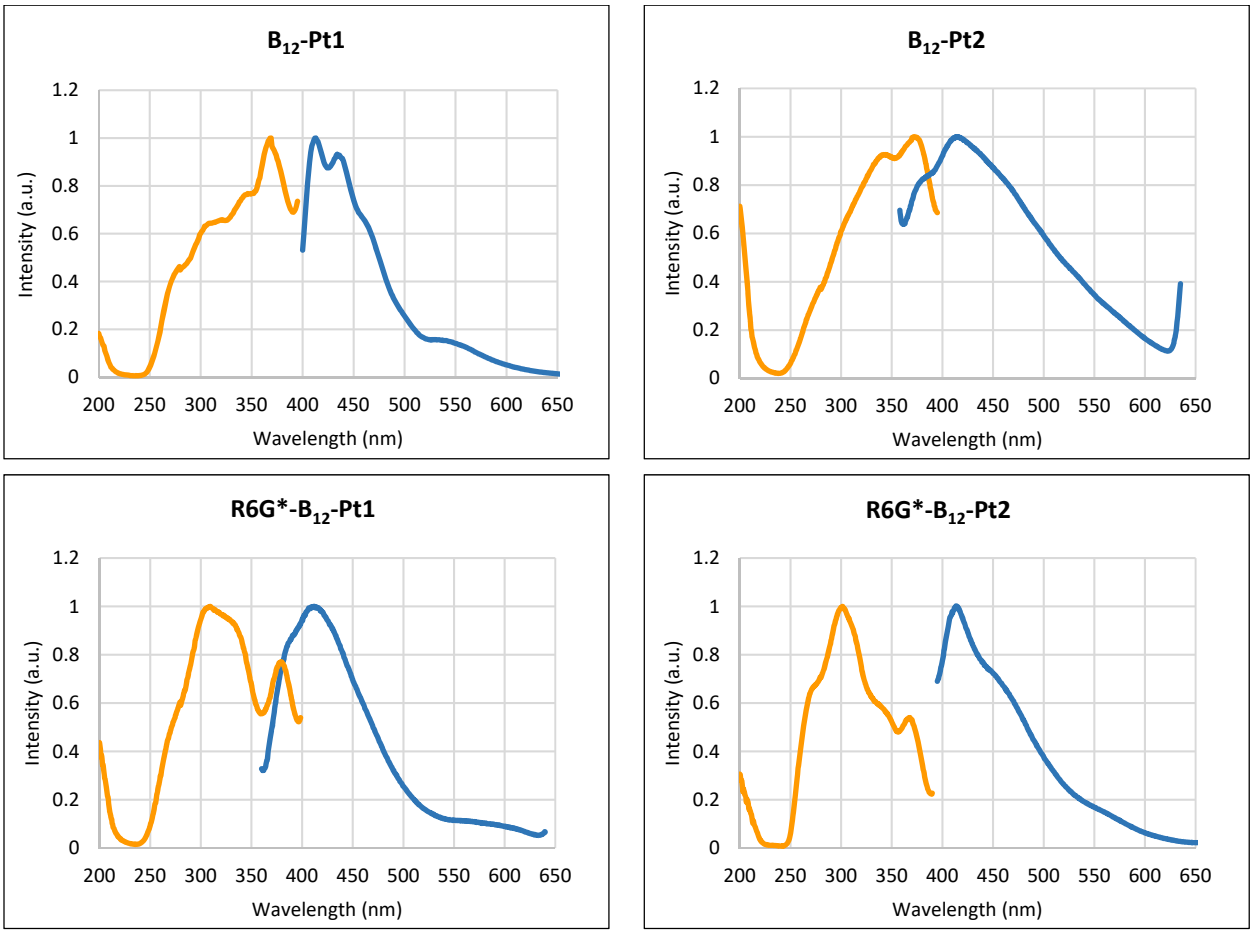
Absorption (orange) and emission (blue) spectra of rhodamine 6G (**R6G**) 25 µM in DMSO. Intensities have been normalized to the maxima.



Absorption (orange) and emission (blue) spectra of **R6G*** 25 µM in DMSO. Intensities have been normalized to the maxima.



Absorption (orange) and emission (blue) spectra of **R6G*-B$_{12}$** 25 µM in DMSO. Intensities have been normalized to the maxima.



Absorption (orange) and emission (blue) spectra of **B_{12}-Pt1**, **B_{12}-Pt2**, **R6G*-B_{12}-Pt1** and **R6G*-B_{12}-Pt2** 25 µM in DMSO. Intensities have been normalized to the maxima.

Single-crystal X-ray diffraction data

Table S1. Selection of experimental data for the X-ray diffraction studies on **R6G*** and ***R*$_N$-Pt1**.

	R6G*	***R*$_N$-Pt1**
CCDC code	2321413	2321414
Formula	$C_{32}H_{39}ClN_4O_4$	$C_{16}H_{22}Cl_2N_2OPt$
M_r	579.12	524.34
T [K]	301(2)	200(2)
λ [Å]	0.71073	0.71073
crystal system	Triclinic	Monoclinic
space group	$P\bar{1}$	$P2_1$
a [Å]; α [º]	11.996(1); 100.96(1)	8.362(1)
b [Å]; β [º]	12.301(1); 114.32(1)	12.743(1); 95.31(1)
c [Å]; γ [º]	13.450(1); 108.00(1)	8.522(1)
V [Å^3]	1600.7(3)	1343.8(2)
Z	2	2
ρ_{calcd} [g cm^{-3}]	1.202	1.926
$\mu_{MoK\alpha}$ [mm^{-1}]	0.160	8.058
F(000)	616	504
crystal size [mm^3]	0.40×0.30×0.05	0.20×0.17×0.12
θ range [deg]	3.48 to 29.24	3.20 to 27.50
index ranges	-16 to 15	-10 to 10
	-16 to 9	-16 to 16
	-17 to 18	-11 to 11
Reflections collected	14309	17625
Unique data	7375 [R_{int} = 0.071]	4134 [R_{int} = 0.073]
obsd data [I>2σ(I)]	2584	3419
Goodness-of-fit on F^2	0.974	0.905
final R[a] indices [I>2σ(I)]	R1 = 0.091	R1 = 0.043
	wR2 = 0.209	wR2 = 0.095
R indices (all data)[a]	R1 = 0.241	R1 = 0.066
	wR2 = 0.299	wR2 = 0.111
largest diff. peak/hole [e Å^{-3}]	0.530/-0.445	4.229/-1.237

[a] $R1 = \Sigma||F_0|-|F_c||/[\Sigma|F_0|]$; $wR2 = \{[\Sigma w(F_0^2-F_c^2)^2]/[\Sigma w(F_0^2)^2]\}^{1/2}$

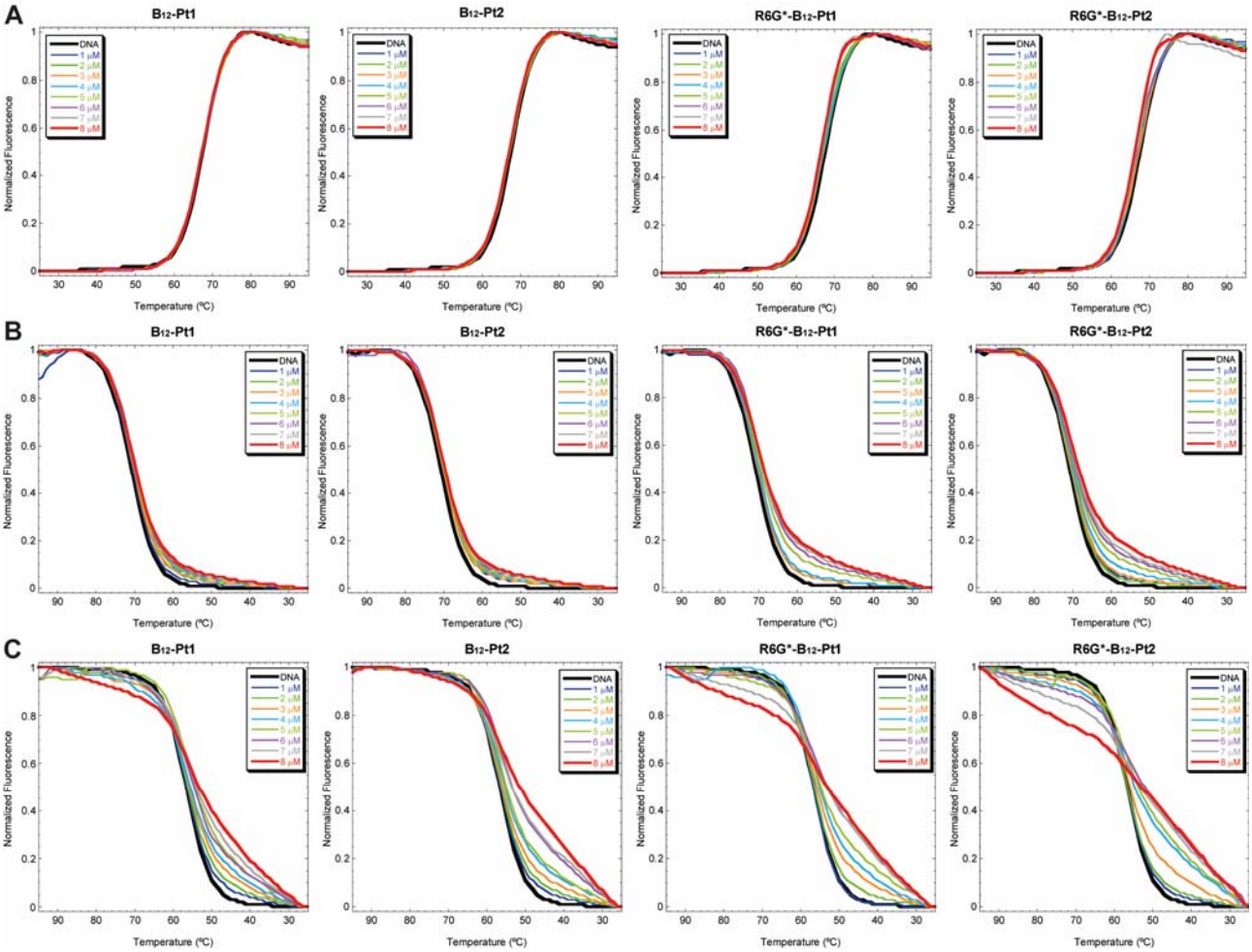
Table S2. Selected bond lengths (Å), bond angles (deg) and torsion angles (deg) for **R6G***.

N(3)–C(19)	1.490(5)	N(3)–C(26)	1.344(5)
N(3)–C(27)	1.445(5)	O(2)–C(26)	1.237(5)
C(19)–N(3)–C(26)	113.2(3)	C(19)–N(3)–C(27)	121.5(3)
C(26)–N(3)–C(27)	125.0(3)	N(3)–C(26)–O(2)	125.5(4)
N(3)–C(26)–C(25)	107.4(4)	C(26)–C(25)–C(20)	108.6(4)
C(19)–C(20)–C(25)	110.9(4)	N(3)–C(19)–C(20)	99.8(3)
N(3)–C(19)–C(6)	112.7(3)	N(3)–C(19)–C(15)	111.0(3)
C(20)–C(19)–C(6)	112.4(3)	C(20)–C(19)–C(15)	109.9(4)
C(26)–N(3)–C(19)–C(6)	-121.7(4)	C(27)–N(3)–C(19)–C(6)	64.7(4)
C(26)–N(3)–C(19)–C(15)	113.6(4)	C(27)–N(3)–C(19)–C(15)	-60.0(5)
C(25)–C(20)–C(19)–C(6)	120.7(4)	C(21)–C(20)–C(19)–C(6)	-60.6(6)
C(25)–C(20)–C(19)–C(15)	-115.7(4)	C(21)–C(20)–C(19)–C(15)	62.9(6)
N(3)–C(19)–C(6)–C(5)	53.5(5)	C(20)–C(19)–C(6)–C(5)	-58.4(5)
N(3)–C(19)–C(6)–C(7)	-130.3(4)	C(20)–C(19)–C(6)–C(7)	117.8(4)
N(3)–C(19)–C(15)–C(10)	130.6(4)	C(20)–C(19)–C(15)–C(10)	-119.9(4)
N(3)–C(19)–C(15)–C(14)	-51.9(5)	C(20)–C(19)–C(15)–C(14)	57.6(5)
O(2)–C(26)–N(3)–C(27)	-6.1(7)		

Table S3. Selected bond lengths (Å) and bond angles (deg) for ***R*$_N$-Pt1**.

Pt(1)–Cl(1)	2.301(4)	Pt(1)–Cl(2)	2.284(4)
Pt(1)–N(1)	1.971(14)	Pt(1)–N(2)	2.046(12)
N(1)–O(1)	1.396(17)	N(1)–C(2)	1.30(2)
C(1)–C(2)	1.50 (3)	N(2)–C(1)	1.55(2)
Cl(1)–Pt(1)–Cl(2)	93.5(2)	Cl(1)–Pt(1)–N(1)	93.3(4)
Cl(1)–Pt(1)–N(2)	174.5(4)	Cl(2)–Pt(1)–N(1)	173.2(4)
Cl(2)–Pt(1)–N(2)	91.4(4)	N(1)–Pt(1)–N(2)	81.8(5)
Pt(1)–N(1)–C(2)	118.8(13)	Pt(1)–N(1)–O(1)	124.7(11)
C(2)–N(1)–O(1)	116.4(15)	Pt(1)–N(2)–C(1)	108.5(9)
Pt(1)–N(2)–C(11)	113.6(9)	C(1)–N(2)–C(11)	113.8(12)
N(1)–C(2)–C(1)	115.8(16)	N(2)–C(1)–C(2)	109.0(13)

FRET-DNA melting curves



FRET-based DNA melting assays using oligonucleotide F10T (0.2 mM) and vitamin B_{12}-platinum(II) conjugates **B_{12}-Pt1**, **B_{12}-Pt2**, **R6G*-B_{12}-Pt1** or **R6G*-B_{12}-Pt2**, at a concentration between 1 and 8 μM. **A**) melting curves; **B**) renaturing curves representing dsDNA annealing; **C**) renaturing curves representing dsDNA annealing (in the absence of chloride ions).