Supplementary Material

Rh^{III}-catalyzed synthesis and investigation of the DNA-binding properties of 11- and 13-substituted berberine derivatives

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1 Equipment

1.1 Reagents and solvents

All reagents were commercially available and were purchased from the following companies: - Alfa Aesar GmbH & Co KG (Haverhill, USA): iodine monochloride, triphenylphosphine, imidazole - Acros Organics (Geel, Belgium): phenylacetylene, 1,1-diphenyleth-2-in-1-ol- Biomers.net GmbH (Ulm, Germany): **22AG** d[A(GGGTTA)$_3$GGG] (HPLC-grade) - Chempur GmbH(Karlsruhe, Germany): Cu(BF$_4$)$_2$ x 6 H$_2$O - Fluka Chemie AG (Brüssel, Belgium): NaOAc, NaH$_2$PO$_4$, Na$_2$HPO$_4$, H$_3$PO$_4$ - Fluorochem Ltd (Hadfield, UK): triisopropylsilyl chloride, Pd(PPh$_3$)Cl$_2$, triisopropylsilylacetylene, DBU - Merck KGaA (Darmstadt, Germany): *i*Pr$_2$NH, boronic acid - Messer GmbH (Bad Soden, Germany): oxygen (purity 4.8) - Oxchem Corp. (Chicago, USA): Pd(PPh$_3$)Cl$_2$ - Sigma-Aldrich Chemie GmbH (Deisenhofen, Germany): calf thymus DNA (ct DNA, type I, high polymerized sodium salt), homoveratrylamine, K$_3$PO$_4$ - VWR Chemicals International GmbH (Langenfeld, Germany): Na$_2$EDTA. The catalyst [RhCp*(CH$_3$CN)$_3$](SbF$_6$)$_2$ was prepared according to literature protocol.[1] All stored solvents were kept under inert gas atmosphere (Ar) and with exclusion from light. Diisopropylamine (DIPA) was stirred with 10 g/L CaH$_2$ under inert gas atmosphere and under reflux for 1 d. The suspension was destilled, and the distillate was stored over molecular sieve (4 Å).Tetrahydrofuran (THF) was stirred under reflux over Na wire and distilled. The distillate was stored over molecular sieve (4 Å). All other reagents and solvents were commercially available and used without further purification. The literature known compounds *N*-(3,4-dimethoxyphenethyl)-2,2,2-trifluoracetamide (**2b**),[2] 2,2,2-trifluoro-*N*-(2-iodo-4,5-dimethoxyphenethyl)acetamide (**2c**),[3] *N*-{2-[4,5-dimethoxy-2-(phenylethynyl)phenyl]ethyl}-2,2,2-trifluoroacetamide (**4a**),[2] 2-[4,5-dimethoxy-2-(phenylethinyl)phenyl]ethan-1-amine (**5a**),[4] 2-{4,5-dimethoxy-2-[(triisopropylsilyl)ethinyl]phenyl}ethan-1-amine (**5b**)[4] and 4-[(triisopropylsilyl)oxy]benzaldehyde (**6c**)[5] were synthesized according to literature protocol.

1.2 Preparation of DNA, ligand, and buffer solutions

Preparation of buffer solutions

For the preparation of buffer solutions E-Pure® water (18 MΩ cm) and the chemicals listed above (1.1) were used. The buffer solutions were kept under exclusion of light at 4 °C for a maximum of three weeks and filtered through a membrane filter (pore size 0.45 µm; Carl Roth GmbH, Karlsruhe, Germany) before usage. The compositions of the buffer solutions are given in the following list:

BPE buffer: *c*(Na$_2$HPO$_4$) = 6.0 mM, *c*(NaH$_2$PO$_4$) = 2.0 mM, *c*(Na$_2$EDTA) = 1.0 mM; with aq. NaOH solution (*c* = 2.0 M) a pH value of 7.0 was adjusted.

Britton-Robinson buffer: *c*(H$_3$BO$_3$) = 4.0 mM, *c*(H$_3$PO$_4$) = 4.0 mM, *c*(NaOAc) = 4.0 mM; with aq. HCl solution (*c* = 1.0 M) a pH value of 2.0 was adjusted.

Potassium phosphate buffer: *c*(K$_2$HPO$_4$) = 25 mM; with KH$_2$PO$_4$ solution (*c* = 25 mM) a pH value of 7.0 was adjusted.

Preparation of ct DNA solution

A ct DNA concentration of approximately 1–2 mg mL^{-1} was adjusted in a BPE buffer solution. The solution was kept at 4 °C for 2 d under exclusion of light and was filtered through a

membrane filter (pore size 0.45 µm; Carl Roth GmbH, Karlsruhe, Germany). The concentration was determined photometrically after dilution with BPE buffer (1:40) according to the Lambert-Beer law (ε_{264} = 12824 $cm^{-1}M^{-1}$, *c* in base pairs).[1]

*Preparation of **22AG** solution*

The lyophilised oligonucleotide strand **22AG** was dissolved in potassium phosphate buffer and heated in a water bath at 90 °C for 5 min. The solution was slowly cooled to r.t. and kept at –25 °C. The concentration of the solution was c_{DNA} = 200 µM.

Preparation of ligand solutions

For spectroscopic measurements stock solutions of the ligands with a concentration *c* = 1.00 mM in MeOH were prepared and kept with exclusion from light at –4 °C.

2 Photometric and fluorimetric analysis of derivatives 1b and 1e

2.1 Absorption and emission properties

Starting from the stock solutions of the derivatives **1b** and **1e** a solution (c = 20 µM or 40 µM) in MeOH, DMSO, H_2O, $CHCl_3$, $HClO_4$, and in BPE buffer was prepared. For every solution, an absorption and emission spectrum was recorded. The absorption spectra were recorded in a range of 200–500 nm with a measurement speed of 120 nm min^{-1}. The emission spectra were recorded in a range of 365 to 685 nm with a measurement speed of 120 nm min^{-1}. The excitation wavelength was set to λ_{ex} = 350 nm and the detector voltage was adjusted to 600 V. All spectra were processed with Origin software[6] ("moving average") or in the program "Scan" with the implemented smoothing function with a factor of 10.

2.2 Photometric and fluorimetric DNA titrations

Starting from the stock solutions of the derivative **1e** a solution in BPE buffer or potassium phosphate buffer (c = 20 µM or 40 µM) was prepared for the titration with ct DNA or **22AG**. The sample volumes were V_{sample} = 1500 µL for ct DNA titrations and V_{sample} = 500 µL for titrations with **22AG**. The ligand solutions were titrated with DNA solution, that contained the same ligand concentration as the analyte sample, to prevent a dilution effect. The solutions contained up to 5% (v/v) DMSO to ensure sufficient solubility. After every titration step, the solution was equilibrated for 3 min, and an absorption spectrum was recorded. In both titrations an emission spectrum of the ligand solution and of the DNA-ligand solution after the titration was recorded. The volume of the DNA solution was increased when the change in the absorption decreased between two titration steps. When no change in absorption was observed or the DNA precipitated from the solution the titration was stopped. For the fluorimetric titrations the solution was excited at the wavelength that corresponded to the isosbestic points, as obtained from the photometric titrations.

2.3 Photometric and fluorimetric acid-base titrations of 1b

A solution of **1b** (c = 20 µM) in Britton-Robinson buffer (pH = 2.0) was titrated with aq. NaOH solution (c = 2 M) in a pH range of 2–11. After every titration step, an absorption and an emission spectrum was recorded, and the corresponding pH value was determined. The fluorescence intensity was corrected based on the change in absorption at the excitation wavelength.

To determine the pK_a value of **1b**, the experimental titration data were fitted to the theoretical curve considering the Henderson-Hasselbalch equation for weak acids (eq.1).

$$A = \frac{A_{HA}10^{-pH} + A_A10^{-pK_a}}{10^{-pH} + 10^{-pK_a}} \quad \text{(eq. 1)}$$

A is the absorption at a given wavelength, A_A is the absorption of the deprotonated form of compound **1b**^{cB} and A_{HA} is the absorption of **1b**.

3 NMR spectra

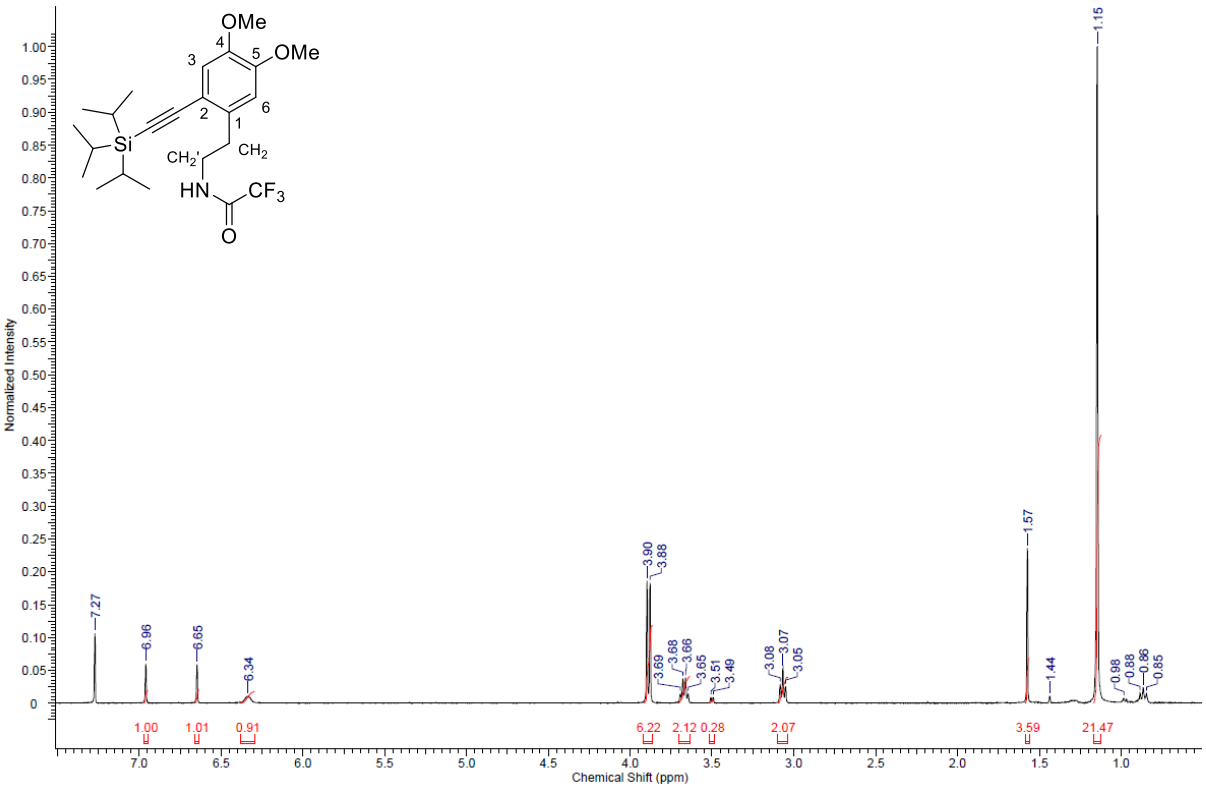

Figure S1. ^{1}H NMR spectrum (400 MHz) of **5b** in $CDCl_3$.

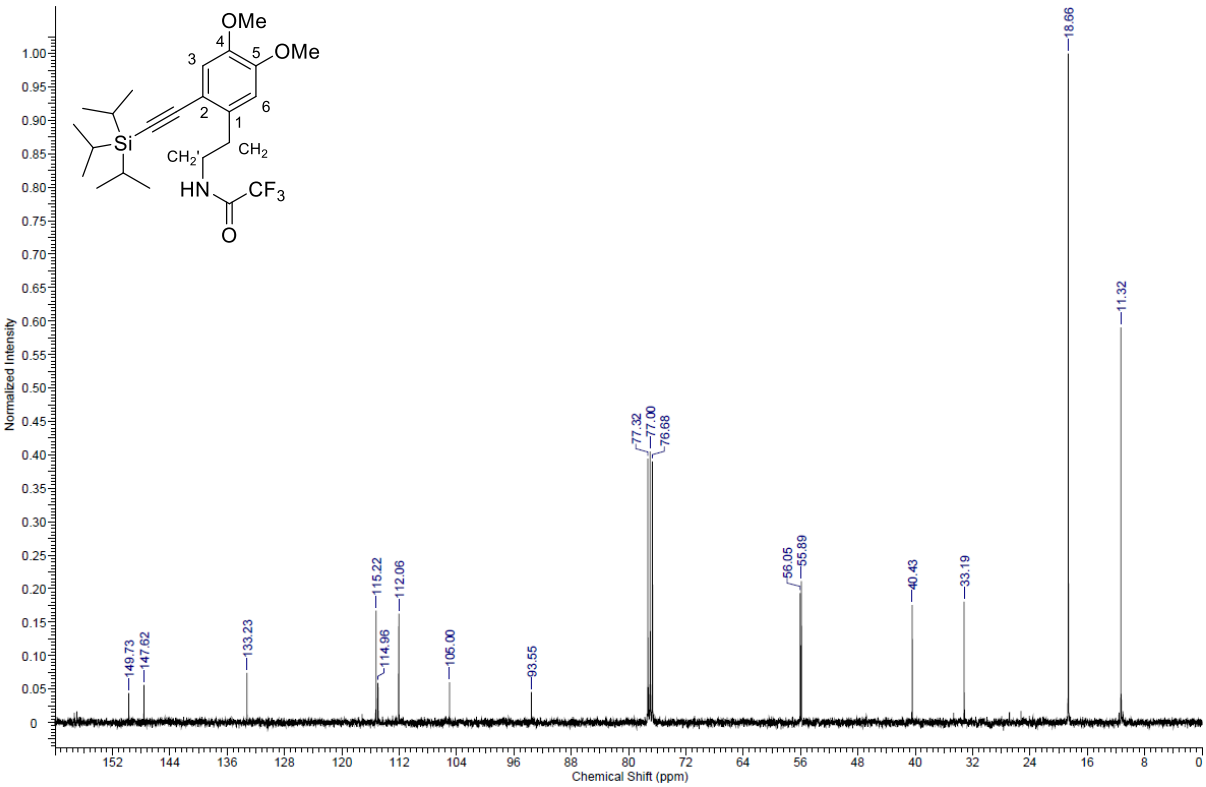

Figure S2. ^{13}C NMR spectrum (100 MHz) of **5b** in $CDCl_3$.

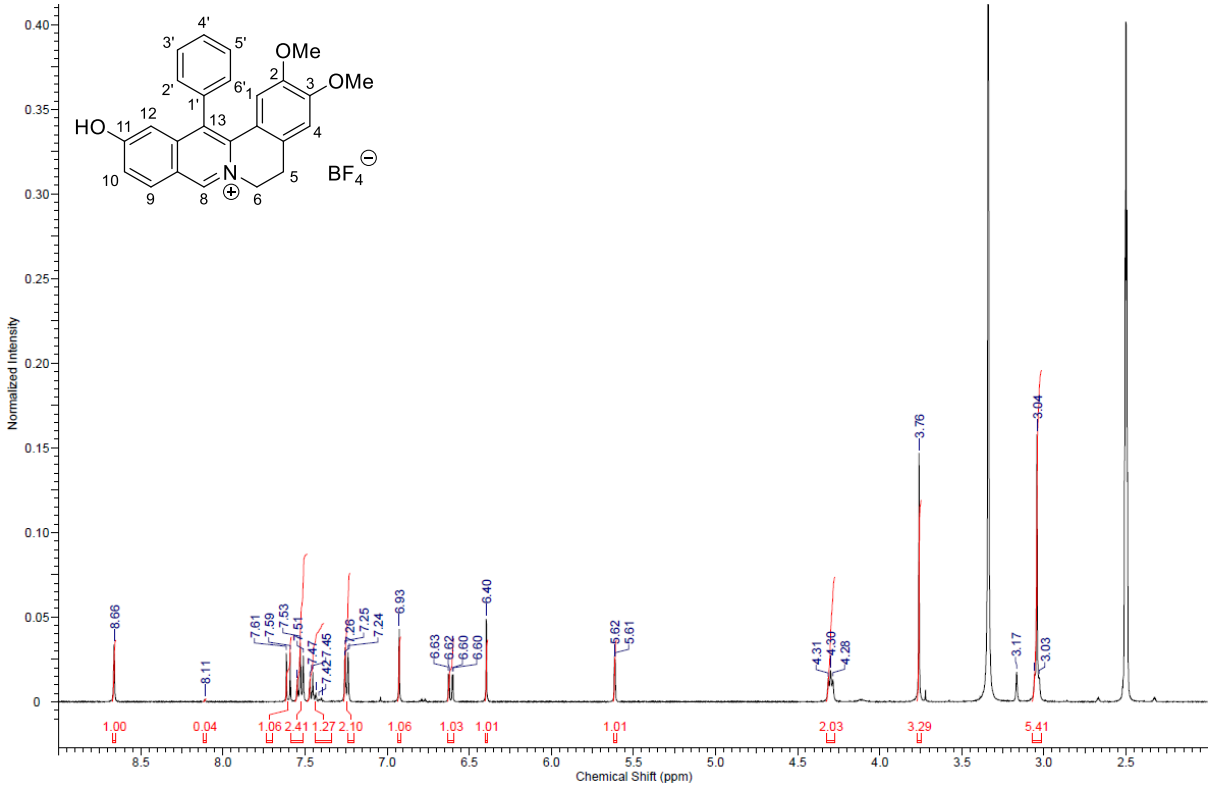

Figure S3. ^{1}H NMR spectrum (400 MHz) of **1b** in DMSO-d_6.

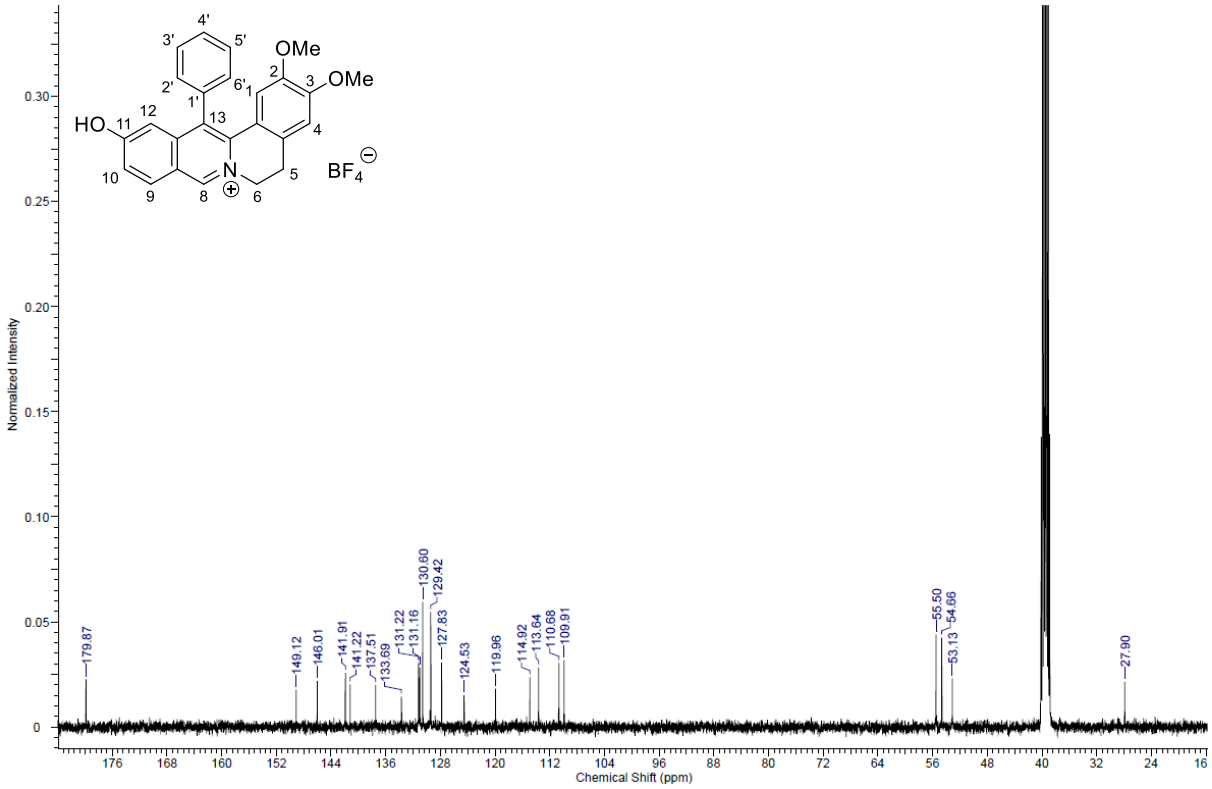

Figure S4. ^{13}C NMR spectrum (100 MHz) of **1b** in DMSO-d_6.

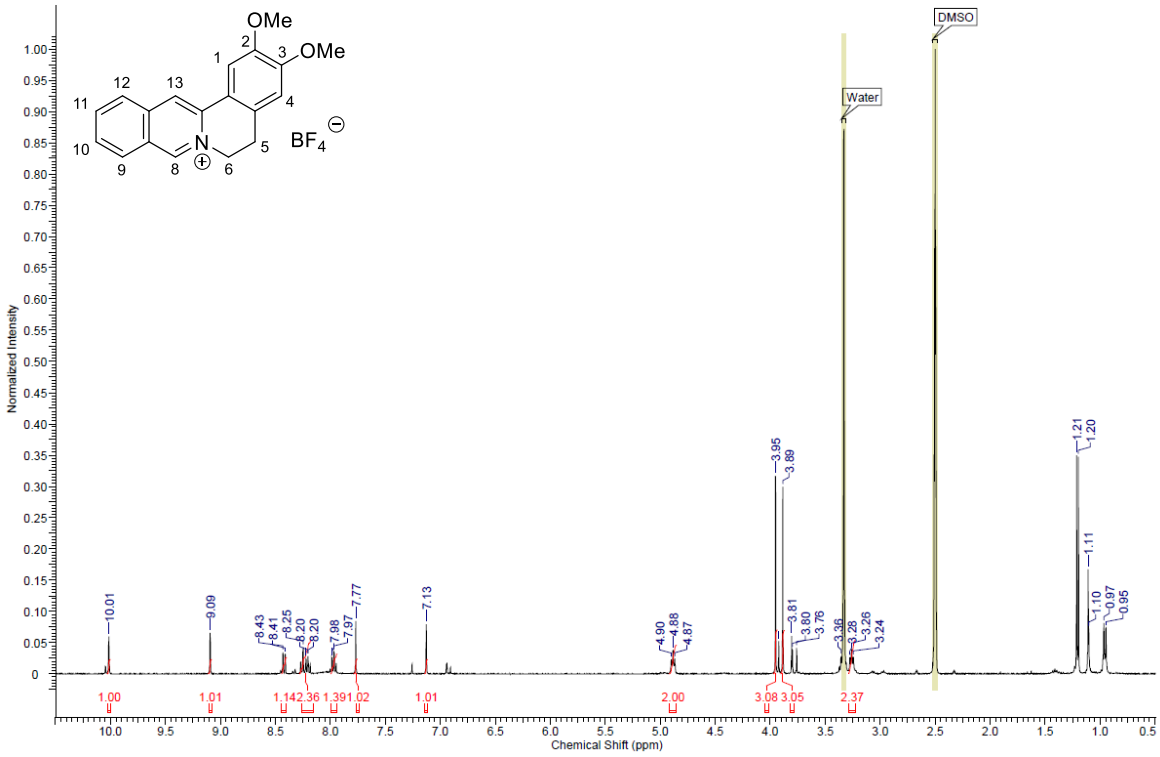

Figure S5. ^{1}H NMR spectrum (400 MHz) of **1e** in DMSO-d_6.

4 References

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