Supporting Information for

A Localized Enantioselective Catalytic Site on Short DNA Sequences and their Amphiphiles

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Experimental Procedures

Materials
ssDNA was purchased from Integrated DNA Technologies (Coralville, IA) and the strand concentrations were determined by measuring the absorbance at 260 nm using the extinction coefficient values published in the literature. All other chemicals and materials were purchased from Sigma-Aldrich (St Louis, MO) and used without further purification unless stated otherwise. The organic compounds (1a and 3a) employed for catalysis and calibration were synthesized based on literature reports (see p. 30 for \(^1\)H NMR and \(^{13}\)C NMR).\(^{[1,2]}\) The hydrophobic dendron tail was synthesized following the protocol reported (see p. 30 for \(^1\)H NMR and \(^{13}\)C NMR).\(^{[3]}\) The ssDNA-amphiphiles were synthesized based on a protocol published previously and characterized by matrix assisted laser desorption ionization-time of flight (MALDI-TOF) mass spectrometry (MS) (Table S2).\(^{[4]}\) Cu(II) complexes (dmbipy-Cu) were prepared as reported previously and characterized by MALDI-TOF MS\((\text{Table S3})^{[5]}\).

Methods
\(^1\)H NMR and \(^{13}\)C NMR spectra were recorded on a Bruker Advance 400 MHz FT-NMR spectrometers with CDCl\(_3\) at room temperature. Proton chemical shifts are referenced to TMS (0.00 ppm, in CDCl\(_3\)) and \(^{13}\)C chemical shifts are referenced to TMS (77.16 ppm, in CDCl\(_3\)). ssDNA-amphiphiles were purified by an Agilent 1260 Infinity II high performance liquid chromatography (HPLC) and characterized by MALDI-TOF MS. Analysis of DNA catalytic reactions was performed on an Agilent 1260 Infinity II HPLC with the eluents of hexane and isopropanol (i-PrOH), using a Daicel Chiralcel OD-H column (250 × 4.6 mm) at 25 °C. In all experiments, MilliQ water (Millipore Inc.) with a typical resistivity of 18.2 mΩ/cm was used. All catalysis reactions were carried out in 20 mM MOPS buffer (pH 6.5) unless otherwise stated. Circular dichroism (CD) studies were carried out at 4 °C on an Aviv 420 spectropolarimeter in a 0.1 cm quartz cuvette, with DNA concentrations at 50 µM and the spectra were collected from 320-200 nm with a 1 nm step size, averaging for 3 s at each step. Every CD curve was subtracted by a background CD spectrum of corresponding buffer solution. Melting temperature measurements were carried out on an Aviv 14DS UV-Vis spectrophotometer with Neslab RET-111 temperature controller in a 1 cm quartz cuvette with a 1 °C temperature step.

Preparation of CD samples
An aqueous solution of a ssDNA sequence (final concentration: 50 µM) and its complementary ssDNA sequence (final concentration: 50 µM) were added to MOPS buffer (final volume: 300 µL, final concentration: 20 mM, pH 6.5). The tube was heated in the dry-bath heater to 95 °C, kept for 3 min, and the heater was turned off to let the tube slowly cool down to room temperature. If needed, a solution of dmbipy-Cu (final concentration: 50 µM) would be added.

Typical procedure for Diels-Alder reaction in water
An aqueous solution of a ssDNA sequence (final concentration: 50 µM) and its complementary ssDNA sequence (final concentration: 50 µM) were added to MOPS buffer (final volume: 1 mL, final concentration: 20 mM, pH 6.5). The tube was heated in a dry-bath heater to 95 °C, kept for 3 min, and the heater was then turned off to let the tube slowly cool down to room temperature. A solution of dmbipy-Cu (final concentration: 50 µM) was added and the solution was stirred for 0.5 h at 4 °C. Then, aza-chalcone 1a in CH\(_3\)CN (10 µL of 0.1 M solution) was added. The reaction was initiated by the addition of freshly distilled cyclopentadiene 2 (5.6 µL, 67 eq.) and the mixture was stirred for 3 h at 4 °C, followed by the extraction with diethyl ether (3 × 3 mL) and the filtration through a short pad of silica gel. The solvent was removed under reduced pressure. The conversion, diastereoselectivity (endo:exo) and enantiomeric excess (ee) were determined by chiral HPLC (see p. 36 for a representative HPLC chromatogram and p. 37 for calculation of (1a) conversion).

HPLC condition:
Product 3a: Daicel chiralcel OD-H, hexane/i-PrOH 98:2, 0.5 mL/min, 212 nm. Retention times: 11.5 (Si-exo), 12.8 min (Re-exo); 14.5 (Re-endo), 18.5 min (Si-endo).\(^{[6]}\)

Typical procedure for Diels-Alder reaction in methanol/water (v/v=50/50) mixture
An aqueous solution of a ssDNA sequence (final concentration: 15 µM) and its complementary ssDNA sequence (final concentration: 15 µM) were added to MOPS buffer (500 µL, final concentration: 20 mM, pH 6.5). The tube was heated in a dry-bath heater to 95 °C, kept for 3 min, and the heater was then turned off to let the tube slowly cool down to room temperature. A solution of dmbipy-Cu (final concentration: 15 µM) was added and the solution was stirred for 0.5 h at 4 °C. Then, 500 µL methanol and aza-chalcone 1a in CH\(_3\)CN (10 µL of 0.1 M solution) were
added. The reaction was initiated by the addition of freshly distilled cyclopentadiene 2 (5.6 μL, 67 eq.) and the mixture was stirred for 48 h at 4 °C, followed by the extraction with diethyl ether (3 × 3 mL) and the filtration through a short pad of silica gel. The solvent was removed under reduced pressure. The conversion, diastereoselectivity (endo:exo) and enantiomeric excess (ee) were determined by chiral HPLC.

HPLC condition:
Product 3a: Daicel chiralcel OD-H, hexane/i-PrOH 98:2, 0.5 mL/min, 212 nm. Retention times: 11.5 (Si-exo), 12.8 min (Re-exo); 14.5 (Re-endo), 18.5 min (Si-endo).[6]

Melting temperature measurements
The melting temperatures of the sequences “8-4,5”, “8”, HT21 and st-DNA were measured on an Aviv 14DS UV-Vis spectrophotometer with Neslab RET-111 temperature controller in a 1 cm quartz cuvette. The absorbance was measured at 268 nm every 1 °C for the “8-4, 5” sequence (16 μM), “8” sequence (16 μM), HT21 (3 μM) and st-DNA (8 μM) in phosphate buffer (100 mM, pH 6.5). The samples were held at 80 °C for 10 min and then cooled from 80 °C to 1 °C and heated from 1 °C to 80 °C at 0.2 °C/min. No hysteresis was seen using this temperature gradient. For st-DNA the sample was held at 1 °C for 10 min and then heated to 95 °C at 0.2 °C/min. The melting temperatures were determined from three independent measurements using the first derivative method as described elsewhere.[7]

Binding constant measurements
The binding constants of dmbipy-Cu to different DNA sequences were determined by UV/Vis titration as discussed elsewhere.[5] All measurements were performed in MOPS buffer (20 mM, pH 6.5) at 4 °C. The concentration of dmbipy-Cu was 50 μM and the concentration of the DNA sequences (in base pairs) during the titration process ranged from 32 μM to 160 μM. The absorbance was measured at 307 nm.

Modeling details
Two dsDNA molecules with 21 base pairs of “21” and “21-10, 11” sequences, were modeled in atomistic detail.[8] the sequence of bases along the two strands of the modeled DNAs are shown in Table S1. The modeling of the Cu(II) complex of 4,4’-dimethyl-2,2’-bipyridine (dmbipy-Cu), the crystallographic information file (CIF)[9] of the solid state structure was used.

Molecular dynamics (MD) simulations were conducted in the isobaric, isothermal ensemble (P = 1 bar, T = 310 K); the numerical integration of the differential equations of motion[10-12] was performed by means of the AMBER94[13] force field (details to be published). The partial charges on the dmbipy-Cu atoms were derived using the R.E.D. server[14] by employing the Restricted Electrostatic Potential methodology[15-16] to reproduce the molecular electrostatic potential computed via density functional theory calculations, using the B3LYP exchange correlation functional with basis set 6-31G (d) level of theory.

The rhombic dodecahedron simulation box dimensions were set so that to ensure at least 3 nm thickness for the solvent surrounding the dmbipy-Cu and DNA molecules in all directions. This value is well above the minimum value of 0.8 to 1.2 nm recommended in protocols for nucleic acids simulations.[17] Randomly selected water molecules were replaced by the appropriate number of Na cations to neutralize the system. The TIP3P water model was employed,[18] which enables high computational efficiency and has been successfully combined with the AMBER force fields in modeling nucleic acids hydration.[19]

For the equilibration of the system, prior to the MD production runs, we followed the equilibration protocol described in our previous work on the binding dynamics of siRNAs with lipopeptide molecules.[20] The Berendsen algorithm was utilized so that to allow for fluctuating the pressure tensor (diagonal components) isotropically under the constrain of a constant trace (the pressure).[21] with a time constant of 2.0 ps; the isothermal compressibility of water set equal to 4.5 × 10^{-5} bar^{-1}. For the thermostating a velocity-rescaling scheme[22] was employed on two separate groups, namely the DNA with the dmbipy-Cu, and the ionic solvent, with a time constant of 0.1 ps. For the dispersion interactions a shifted cutoff of 1 nm was applied. Bond lengths were constrained as usually in most biomolecular systems.[23-24] The smooth particle-mesh Ewald method[25] was employed for the computation of the electrostatic interactions. MD trajectories were produced up to 230 ns, using a timestep of 2 fs, and recorded every 10 ps. The geometrical characteristics of the dsDNAs were extracted using CURVES+. Visualization of the MD trajectory and image rendering was performed ultimately by means of VMD.[27]

In the post-processing stage, the distance between the minor groove base atoms and the copper atom was computed as the minimum distance of all the following minor groove base atoms with the copper atom. For “21” sequence, the minor groove base atoms include N3 of adenine, C2 of adenine and O2 of thymine. For “21-10, 11” sequence, the minor groove base atoms include N3 of adenine, C2 of adenine, O2 of thymine, N3 of guanine, N2 of guanine and O2 of cytosine. The notation of the base atoms follows the standard IUPAC-IUB numbering.
convention for the nucleic acid bases. The computed distances were binned to bins of 0.1 nm width, and, subsequently, normalized histograms over the total number of configurations (time frames) were constructed.

The same procedure was followed for computing the distance between the minor groove base atoms and each of the pyridine methyl carbons. Also, the same procedure was followed for computing the distance between the minor or major groove base atoms and the dmbipy-Cu for all the atoms of the dmbipy-Cu. For “21” sequence, the major groove base atoms include N7 of adenine, N6 of adenine and O4 of thymine; for “21-10, 11” sequence, the major groove base atoms include N7 of adenine, N6 of adenine, O4 of thymine, N7 of guanine, O6 of guanine and N4 of cytosine.

In Figures S19 and S23, computations for the base pairs at the edges of the DNA were excluded due to spurious values caused by terminal base-pair fraying events, a known artifact in nucleic acids simulations using the AMBER force fields.
Supplementary Schemes

**Scheme S1.** The absolute configurations of endo and exo isomers of the Diels-Alder product 3a.\(^6\)

**Scheme S2.** Examples for designating DNA sequences used in this study.
Scheme S3. Typical procedure for the synthesis and hybridization of the dsDNA-amphiphiles.
### Supplementary Tables

*Table S1.* dsDNA sequences employed in this work.

| Name      | Sequence                                      |
|-----------|-----------------------------------------------|
| HT21      | ![Sequence Image](image1.png)                |
| 21        | ![Sequence Image](image2.png)                |
| 21-6      | ![Sequence Image](image3.png)                |
| 21-6, 7   | ![Sequence Image](image4.png)                |
| 21-6, 7, 8| ![Sequence Image](image5.png)                |
| 21-6, 7, 8, 9| ![Sequence Image](image6.png)            |
| 21-6, 16  | ![Sequence Image](image7.png)                |
| 21-10, 11 | ![Sequence Image](image8.png)                |
| 21-15, 16 | ![Sequence Image](image9.png)                |
| 21-18, 19 | ![Sequence Image](image10.png)               |
| 21-20, 21 | ![Sequence Image](image11.png)               |
Table S2. Molecular weight of ssDNA-amphiphiles.

| Complex | Calculated | Measured^[a] |
|---------|------------|--------------|
|         | 7517.5     | 7525.9       |
|         | 7096.2     | 7060.0       |
|         | 3285.8     | 3290.6       |
|         | 3291.8     | 3298.2       |

[a] Mass observed in MALDI-TOF MS analysis.

Table S3. Molecular weight of dmbipy-Cu.

| Complex  | Calculated          | Measured^[a] |
|----------|---------------------|--------------|
| dmbipy-Cu| 308.02 (dmbipy-CuNO$_3^-$) | 309.0        |

[a] Mass observed in MALDI-TOF MS analysis.
Table S4. The sequence of bases along the strands of the dsDNA molecules modeled in this study; the numbering of bases and base pairs are also shown.

| “21” sequence | Strand 5’ to 3’ | Strand 3’ to 5’ |
|----------------|-----------------|-----------------|
| Number of nucleotide pair or base pair | Base No | Base type | Base type | Base No |
| 1              | 1               | T             | A         | 42       |
| 2              | 2               | T             | A         | 41       |
| 3              | 3               | T             | A         | 40       |
| 4              | 4               | T             | A         | 39       |
| 5              | 5               | T             | A         | 38       |
| 6              | 6               | T             | A         | 37       |
| 7              | 7               | T             | A         | 36       |
| 8              | 8               | T             | A         | 35       |
| 9              | 9               | T             | A         | 34       |
| 10             | 10              | T             | A         | 33       |
| 11             | 11              | T             | A         | 32       |
| 12             | 12              | T             | A         | 31       |
| 13             | 13              | T             | A         | 30       |
| 14             | 14              | T             | A         | 29       |
| 15             | 15              | T             | A         | 28       |
| 16             | 16              | T             | A         | 27       |
| 17             | 17              | T             | A         | 26       |
| 18             | 18              | T             | A         | 25       |
| 19             | 19              | T             | A         | 24       |
| 20             | 20              | T             | A         | 23       |
| 21             | 21              | T             | A         | 22       |

| “21-10, 11” sequence | Strand 5’ to 3’ | Strand 3’ to 5’ |
|-----------------------|-----------------|-----------------|
| Number of nucleotide pair or base pair | Base No | Base type | Base type | Base No |
| 1                     | 1               | T             | A         | 42       |
| 2                     | 2               | T             | A         | 41       |
| 3                     | 3               | T             | A         | 40       |
| 4                     | 4               | T             | A         | 39       |
| 5                     | 5               | T             | A         | 38       |
| 6                     | 6               | T             | A         | 37       |
| 7                     | 7               | T             | A         | 36       |
| 8                     | 8               | T             | A         | 35       |
| 9                     | 9               | T             | A         | 34       |
| 10                    | 10              | G             | C         | 33       |
| 11                    | 11              | G             | C         | 32       |
| 12                    | 12              | T             | A         | 31       |
| 13                    | 13              | T             | A         | 30       |
| 14                    | 14              | T             | A         | 29       |
| 15                    | 15              | T             | A         | 28       |
| 16 | 16 | T | A | 27 |
| 17 | 17 | T | A | 26 |
| 18 | 18 | T | A | 25 |
| 19 | 19 | T | A | 24 |
| 20 | 20 | T | A | 23 |
| 21 | 21 | T | A | 22 |
Figure S1. Conversion and ee% for Diels-Alder reaction catalyzed by 21-base pair dsDNA sequences containing two G•C pairs at different positions with dmbipy-Cu. Same reaction conditions as in Figure 1.
Figure S2. CD spectra of HT21 sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.). Spectra are shown from three independent experiments.

Figure S3. CD spectra of st-DNA (1.05 mM base pairs, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).
**Figure S4.** CD spectra of “21” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

**Figure S5.** CD spectra of “21-6” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).
Figure S6. CD spectra of “21-6, 7” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

Figure S7. CD spectra of “21-6, 7, 8” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).
**Figure S8.** CD spectra of “21-6, 7, 8, 9” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

**Figure S9.** CD spectra of “21-10, 11” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).
Figure S10. CD spectra of “12-6, 7” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

Figure S11. CD spectra of “8-4, 5” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).
Figure S12. CD spectra of “6-3, 4” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

Figure S13. CD spectra of “21-6, 16” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).
**Figure S14.** CD spectra of “21-20, 21” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

**Figure S15.** CD spectra of “21-18, 19” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).
Figure S16. CD spectra of “21-15, 16” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

Figure S17. Atoms notation in the modeled dmbipy-Cu complex.
Figure S18. Time evolution of the distance between the DNA and the dmbipy-Cu, computed as the minimum distance of all the DNA atoms with all the dmbipy-Cu atoms, over the last 170 ns of the equilibrated MD trajectory for “21” (left) and “21-10, 11” (right); distances below 0.4 nm denote bound molecules.

Figure S19. Rise per base pair along the helix axis computed over the entire MD trajectory, for “21” (red) and “21-10, 11” (blue); the crystallographic value of the rise per base pair\textsuperscript{[30]} is also depicted (dotted line) for comparison.

Figure S20. Probability density functions of finding the dmbipy-Cu at various distances from the minor (continuous line), or major (dashed line), groove base atoms (see text), for “21” (left) and “21-10, 11” (right), computed over the last 170 ns of the equilibrated MD trajectory.
Figure S21. Minimum distance between the dmbipy-Cu and each of the 21 DNA nucleotide pairs, computed over the equilibrated MD trajectory for “21” (red) and “21-10, 11” (blue). The numbering of nucleotide pairs for the two sequences can be seen in Table S4.

Figure S22. Probability density functions of finding the pyridine methyl carbon C9 of the dmbipy-Cu at various distances from the minor groove base atoms (see text) in “21” (red) and “21-10, 11” (blue), computed over the last 170 ns of the equilibrated MD trajectory. For the notation of carbon atom see Figure S17.
**Figure S23.** Minor groove width along the DNA helix for the “21” (red) and “21-10, 11” (blue), computed over the entire MD trajectory (continuous lines); the minor groove width for the initial DNA structures is also shown (dotted lines). Shadowed rectangles denote the nucleotide pairs whereat the bipy-Cu binds in “21” (light red) and “21-10, 11” (light blue).

**Figure S24.** Indicative configurations rendered from the MD trajectory of the dmbipy-Cu binding to the DNA in “21” (left) and “21-10, 11” (right), depicting also: the water molecules surrounding the complex (top); only the first-shell coordination waters around the copper atom of the dmbipy-Cu at the square-planar equatorial and axial positions (bottom). Color code for the dmbipy-Cu and waters: Cu (magenta), N (blue), C (cyan), H (white), O (red); color code for the DNA: guanine (green), cytosine (cyan), adenine and thymine (yellow).
Figure S25. (a) Radial distribution function of water oxygens with respect to the copper atom, and (b) probability density functions of finding the copper atom at various distances from the oxygen atoms of the DNA phosphate groups (b), in “21” (red) and “21-10, 11” (blue), computed over the equilibrated MD trajectory.

Figure S26. The reactant [1a], added by following the bidentate coordination pattern of the enone to the copper atom of the dmbipy-Cu as shown in Figure 2e, is depicted in (a) for “21”, and (b, c) for “21-10, 11” (b, c are rotated views of the same configuration). The configurations depicted in (a), (b) and (c) correspond to the ones shown in Figure 2 (f), (g) and (h), with only difference being that the reactant [1a] is rotated by 180° to present its Re-face, i.e., in (a) and (c) the Re-face is presented, while Figure 2 (f) and (h) presents the Si-face. Water molecules in the rendered pictures are omitted for clarity. Color code for the dmbipy-Cu and reactant [1a]: Cu (magenta), N (blue), C (cyan), H (white), O (red); color code for the DNA: guanine (green), cytosine (cyan), adenine and thymine (yellow).
Figure S27. Melting curves of sequences (a) “8-4, 5” at 16 μM, (b) “8” at 16 μM, (c) HT21 at 3 μM, and (d) st-DNA at 8 μM in phosphate buffer. Measurements were taken at 268 nm at 0.2 °C/min. Representative curves are shown from three independent experiments (n=3).

Figure S28. Binding constant of dmbipy-Cu to different sequences. Data are shown as mean ± SD (n=3). Statistical difference was determined using one-way ANOVA with Tukey’s honest significant difference post-hoc test; * P < 0.05, for all other pairs P > 0.05.
Figure S29. (a) HPLC trace for the Diels-Alder reaction mixture catalyzed by right-handed "8-4, 5" sequence with dmbipy-Cu (ee: 95%). (b) HPLC trace for the Diels-Alder reaction mixture catalyzed by left-handed "8-4, 5" sequence with dmbipy-Cu (ee: -95%). Same reaction and conditions as in Figure 1.
Figure S30. CD spectra of left-handed “8-4, 5” (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

Figure S31. Conversion and ee% for Diels-Alder reaction catalyzed by different 8-base pair dsDNA sequences with dmbipy-Cu. Same reaction and conditions as in Figure 1.
Figure S32. CD spectra of (TTTGCTTT) • (AAACGAAA) sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

Figure S33. CD spectra of 8-base pair alternating G•C sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).
Figure S34. CD spectra of “8” sequence (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).

Figure S35. CD spectra of 8-base pair dsDNA based on alternative TA base pairs (50 µM, in 20 mM MOPS buffer, pH: 6.5) in the absence or presence of dmbipy-Cu (1 eq.).
Figure S36. CD spectra of HT21 and its amphiphile “HT21-amph.” in methanol-water (v/v = 50/50) mixture (15 µM, in 20 mM MOPS buffer, pH: 6.5) in the presence of dmbipy-Cu (1 eq.). Spectra are shown from three independent experiments.

Figure S37. CD spectra of “8-4, 5” and its amphiphile “8-4, 5-amph.” in methanol-water (v/v = 50/50) mixture (15 µM, in 20 mM MOPS buffer, pH: 6.5) in the presence of dmbipy-Cu (1 eq.).
References

[1] S. Otto, F. Bertoncin, J. B. Engberts, *J. Am. Chem. Soc.* **1996**, *118*, 7702-7707.
[2] D. A. Evans, K. R. Fandrick, H.-J. Song, *J. Am. Chem. Soc.* **2005**, *127*, 8942-8943.
[3] V. Percec, W.-D. Cho, G. Ungar, D. J. Yeardley, *J. Am. Chem. Soc.* **2001**, *123*, 1302-1315.
[4] H. Kuang, T. E. Gartner lli, M. D. de Mello, J. Guo, X. Zuo, M. Tsapatsis, A. Jayaraman, E. Kokkoli, *Nanoscale* **2019**, *11*, 19850-19861.
[5] G. Roelfes, A. J. Boersma, B. L. Feringa, *Chem. Commun.* **2006**, 635-637.
[6] C. Wang, Q. Qi, W. Li, J. Dang, M. Hao, S. Lv, X. Dong, Y. Gu, P. Wu, W. Zhang, *Nanoscale* **2019**, *11*, 19850-19861.
[7] M. Z. Akhter, A. Sharma, M. R. Rajeswari, *Mol. Biosyst.* **2011**, *7*, 1336-1346.
[8] S. Li, W. K. Olson, X.-J. Lu, *Nucleic Acids Res.* **2019**, *47*, W26-W34.
[9] A. Draksharapu, A. J. Boersma, M. Leising, A. Meetsma, W. R. Browne, G. Roelfes, *Dalton Trans.* **2015**, *44*, 3647-3655.
[10] M. J. Abraham, T. Murtola, R. Schulz, S. Páll, J. C. Smith, B. Hess, E. Lindahl, *SoftwareX* **2015**, *1*, 19-25.
[11] D. Van Der Spoel, E. Lindahl, B. Hess, G. Groenhof, A. E. Mark, H. J. Berendsen, *J. Comput. Chem.* **2005**, *26*, 1701-1718.
[12] H. J. Berendsen, D. van der Spoel, R. van Drunen, *Comput. Phys. Commun.* **1995**, *91*, 43-56.
[13] W. D. Cornell, P. Cieplak, C. I. Bayly, I. R. Gould, K. M. Merz, D. M. Ferguson, D. C. Spellmeyer, T. Fox, J. W. Caldwell, P. A. Kollman, *J. Am. Chem. Soc.* **1995**, *117*, 5179-5197.
[14] E. Vanquelef, S. Simon, G. Marquant, E. Garcia, G. Klimerak, J. C. Delepine, P. Cieplak, F.-Y. Dupradaeu, *Nucleic Acids Res.* **2011**, *39*, W511-W517.
[15] C. I. Bayly, P. Cieplak, W. Cornell, P. A. Kollman, *J. Phys. Chem.* **1993**, *97*, 10269-10280.
[16] R. H. Henchman, J. W. Essex, *J. Comput. Chem.* **1999**, *20*, 483-498.
[17] R. Galindo-Murillo, C. Bergonzo, T. E. Cheatham III, *Curr. Protoc. Nucleic Acid Chem.* **2014**, *56*, 7.10. 11-17.10. 21.
[18] W. L. Jorgensen, J. Chandrasekhar, J. D. Madura, R. W. Impey, M. L. Klein, *J. Chem. Phys.* **1983**, *79*, 926-935.
[19] P. Kührovará, M. Otyepka, J. í. Šponer, P. Banáš, *J. Chem. Theory Comput.* **2014**, *10*, 401-411.
[20] E. Pantatosaki, G. K. Papadopoulos, *J. Chem. Theory Comput.* **2020**, *16*, 3842-3855.
[21] H. J. Berendsen, J. v. Postma, W. F. van Gunsteren, A. DiNola, J. R. Haak, *J. Chem. Phys.* **1984**, *81*, 3684-3690.
[22] G. Bussi, D. Donadio, M. Parrinello, *J. Chem. Phys.* **2007**, *126*, 014101.
[23] B. Hess, H. Bekker, H. J. Berendsen, J. G. Fraaije, *J. Comput. Chem.* **1997**, *18*, 1463-1472.
[24] B. Hess, *J. Chem. Theory Comput.* **2008**, *4*, 116-122.
[25] U. Essmann, L. Perera, M. L. Berkowitz, T. Darden, H. Lee, L. G. Pedersen, *J. Chem. Phys.* **1995**, *103*, 8577-8593.
[26] R. Lavery, M. Moakher, J. H. Maddocks, D. Petkevicute, K. Zakrzewska, *Nucleic Acids Res.* **2009**, *37*, 5917-5929.
[27] W. Humphrey, A. Dalke, K. Schulten, *J. Mol. Graph.* **1996**, *14*, 33-38.
[28] J. A. Secrist III, *Curr. Protoc. Nucleic Acid Chem.* **2000**, A. 1D. 1-A. 1D. 3.
[29] M. Zgarbová, M. Otyepka, J. Sponer, F. Lankas, P. Jurečka, *J. Chem. Theory Comput.* **2014**, *10*, 3177-3189.
[30] J. M. Berg, J. L. Tymoczko, L. Stryer, *Biochemistry. 5th Edition, New York: W H Freeman* **2002**.
1H-NMR Spectra and 13C-NMR Spectra

(1) (E)-3-phenyl-1-(pyridin-2-yl)prop-2-en-1-one (1a)

1a: 1H NMR (400 MHz, CDCl3) δ = 8.76 (ddd, J=4.8, 1.7, 0.9, 1H), 8.32 (d, J=16.1, 1H), 8.24 – 8.17 (m, 1H), 7.96 (d, J=16.1, 1H), 7.92 – 7.86 (m, 1H), 7.77 – 7.72 (m, 2H), 7.50 (ddd, J=7.6, 4.8, 1.3, 1H), 7.43 (dd, J=5.1, 1.8, 3H).
$^{13}$C NMR (101 MHz, CDCl$_3$) $\delta$ = 189.62, 154.26, 148.95, 144.98, 136.91, 135.15, 130.57, 128.97, 127.01, 123.02, 121.03.
(2) 3-phenylbicyclo[2.2.1]hept-5-en-2-yl(pyridin-2-yl)methanone (3a)

3a: ^1^H NMR (400 MHz, CDCl$_3$) $\delta$ = 8.69 (ddd, $J$=4.7, 1.7, 0.9, 1H), 8.01 (dt, $J$=7.9, 1.1, 1H), 7.83 (td, $J$=7.7, 1.7, 1H), 7.46 (ddd, $J$=7.6, 4.8, 1.3, 1H), 7.35 – 7.25 (m, 4H), 7.20 – 7.14 (m, 1H), 6.50 (dd, $J$=5.6, 3.2, 1H), 5.84 (dd, $J$=5.6, 2.8, 1H), 4.54 (dd, $J$=5.2, 3.4, 1H), 3.55 (s, 1H), 3.46 (d, $J$=5.2, 1H), 3.10 (s, 1H), 2.08 (d, $J$=10.4, 1H), 1.62 (ddd, $J$=8.6, 3.4, 1.7, 1H).
$^{13}$C NMR (101 MHz, CDCl$_3$) $\delta = 148.97, 144.82, 139.50, 136.84, 133.02, 128.45, 127.69, 127.05, 125.88, 122.37, 54.36, 49.50, 48.89, 48.37, 45.70.$
Methyl 3,4-Bis[4-(n-dodecan-1-yloxy)benzyloxy]benzoate: $^1$H NMR (400 MHz, CDCl$_3$) δ 7.62 (d, $J = 10.9$ Hz, 1H), 7.34 (d, $J = 12.2$ Hz, 2H), 7.30 (d, $J = 9.2$ Hz, 2H), 6.92 (d, $J = 8.4$ Hz, 1H), 6.87 (d, $J = 8.7$ Hz, 4H), 5.11 (d, $J = 11.4$ Hz, 3H), 3.94 (q, $J = 6.9$ Hz, 4H), 3.87 (s, 3H), 1.83 – 1.70 (m, 4H), 1.45 – 1.23 (m, 36H), 0.88 (t, $J = 6.8$ Hz, 6H).
Methyl 3,4-Bis[4-(n-dodecan-1-yloxy)benzyloxy]benzoate: $^{13}$C NMR (75 MHz, CDCl$_3$) δ 166.80 (s), 158.95 (s), 153.02 (s), 148.36 (s), 129.99 (s), 129.25 (d, $J = 18.5$ Hz), 128.74 (d, $J = 16.9$ Hz), 128.31 (s), 123.89 (s), 122.89 (s), 115.71 (s), 115.19 – 113.89 (m), 113.39 (s), 77.42 (s), 77.00 (s), 76.58 (s), 71.09 (s), 70.70 (s), 68.01 (s), 51.92 (s), 31.89 (s), 29.79 – 28.97 (m), 26.03 (s), 22.67 (s), 14.10 (s).
HPLC Chromatogram

HPLC condition: Daicel chiralcel-ODH, hexane/i-PrOH 98:2, 0.5 mL/min, 212 nm. Product 3a from the Diels-Alder reaction catalyzed by dmbipy-Cu in the presence of “21-10, 11” sequence (ee: 95%). Retention times: 11.5 (Si-exo), 12.8 mins (Re-exo); 14.5 (Re-endo), 18.5 mins (Si-endo).
Calculation of Conversion of (1a)

Conversion of 1a was calculated based on the equation below:

\[
\text{Conversion of 1a (\%) = } \frac{n_{3a}}{n_{1a}+n_{3a}} = \frac{P_{3a}}{P_{3a} + P_{1a}/k_1}
\]

- \(P_{3a}\): HPLC peak areas of 3a
- \(P_{1a}\): HPLC peak areas of 1a

Determination of correction factor \(k_1\). The HPLC peak areas ratios \((P_{1a}/P_{3a})\) were determined with the standard molar ratios \((n_{1a}/n_{3a})\) of 0.1, 0.2, 0.5, 1, 2, 5, 10. The correction factor \(k_1\) determined to be 0.396 (\(R^2 = 0.997\)).