

Electronic Supporting Information

Exploring hydrogen bonding and weak aromatic interactions induced assembly of adenine and thymine functionalised naphthalenediimides

Nagarjun Narayanaswamy, M. B. Avinash and T. Govindaraju.*

Bioorganic chemistry Laboratory, New Chemistry Unit, Jawaharlal Nehru Centre for Advanced Scientific Research, Jakkur P.O., Bangalore-560064, India.

* Corresponding author, Email: tgraju@jncasr.ac.in

Tel: +91 (80) 22082969, Fax: +91 (80) 22082627

Table of Contents

Page **2**: General information

Page **2-5**: Synthesis and characterization of compounds

Page **6**: UV-vis absorption spectra of **NDI-AA** and **NDI-TT** in DMSO/CH₃CN solvent system

Page **7**: Temperature dependent absorption spectra of **NDI-AA** and **NDI-TT** with and without PNA templates in DMSO/water system

Page **8**: TEM and AFM micrographs of **NDI-AA**

Page **8**: 2D-NOESY spectra of **NDI-AA** and **NDI-AA/PNA-TT** (1:1)

Page **9**: Mode of hydrogen bonding interactions among adenine (**NDA-AA**) and adenine-thymine (**NDA-AA/PNA-TT**) nucleobases.

Page **10-11**: ¹H and ¹³C-NMR spectra of **NDI-AA** and **NDI-TT**

Page **12**: Mass spectra of **NDI-AA** and **NDI-TT**

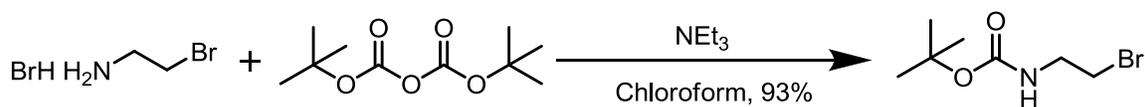
Page **13**: References

General information

Materials: 1, 4, 5, 8-Naphthalenetetracarboxylicdianhydride (NDA), thymine-1-acetic acid, 2-bromo-ethylamine hydrogen bromide were obtained from Sigma–Aldrich. Di-*tert*-butyl dicarbonate, bromo-methyl acetate, adenine, ethylenediamine were obtained from Spectrochem Pvt. Ltd. Mumbai (India). All reagents were used as received unless otherwise mentioned.

Synthetic procedures

Synthesis of *tert*-butyl 2-bromoethylcarbamate

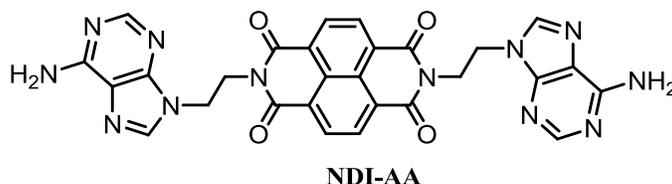


To a stirred solution of 2-bromo-ethylamine hydrogen bromide (2 g, 9.85 mmol) in chloroform, triethyl amine (1.75 mL, 9.85 mmol) was added at 0 °C. After 10 min. Di-*tert*-butyl dicarbonate in chloroform solution was added drop wise to the above solution and the reaction was allowed to stir for 6 h. After completion of the reaction, solvent was evaporated under vacuo. The compound was extracted with DCM (100 mL); organic layer was washed with water (200 mL). Organic layer was dried on anhydrous sodium sulfate. The crude product was purified using column chromatography on silica gel using EtOAc/petroleum ether (15/85) as an eluent to afford colorless liquid in good yield (93 %). ¹H-NMR (400 MHz, CDCl₃)δ 1.38 (s, 9H), 3.39 (t, *J* = 5.32 Hz, 2H), 3.46 (t, *J* = 5.48 Hz, 2H), 4.87 (br, 1H). ¹³C-NMR (100 MHz, CDCl₃)δ 155.2, 79.0, 41.3, 31.8, 27.3. MALDI-TOF-MS: *m/z* = 223.40 [M]⁺ for C₇H₁₄BrNO₂.

Synthesis of Boc-protected adeninyl-9-ethylamine (**1**)

Adenine (1 g, 7.40 mmol) and sodium hydride (0.59 g, 14.8 mmol) were dissolved in DMF solution at 0 °C and stirred for 30 min. To this mixture, *tert*-butyl-2-bromoethylcarbamate (1.65 g, 7.40 mmol) was added. The reaction mixture was heated to 65 °C and allowed to stir for 24 h. The completion of the reaction was monitored with TLC. After completion of reaction, the solution was dissolved in 0.5 N HCl solution at 0 °C, solvent was evaporated under vacuo. The crude product was purified using column chromatography on silica gel using MeOH/CHCl₃ (2/98) as an eluent to afford white solid of **1** in good yield (30%). ¹H-NMR (400 MHz, CDCl₃) δ 1.40 (s, 9H), 3.57 (q, *J* = 6Hz, 2H), 4.35 (d, *J* = 5.2 Hz, 2H), 5.03 (s, 1H), 5.61 (br, 2H), 7.76 (s, 1H), 8.34 (s, 1H). ¹³C-NMR (100 MHz, CDCl₃) δ 155.0, 152.9, 150.1, 140.9, 119.7, 79.9, 43.7, 40.4, 28.2. MALDI-TOF-MS: *m/z* = 278.54 [M]⁺ for C₁₂H₁₈N₆O₂.

Preparation of NDI-AA



Boc-protected adeninyl-9-ethylamine **1** (2.2 equiv.) was dissolved in a mixture of DCM/TFA (1:1) (10 mL). The reaction was allowed to stir for 4 h. After completion of the reaction, solvent was evaporated. The trace amount of TFA was removed by formation of azeotropic mixture with toluene. The crude product was dissolved in DMF (10 mL) containing triethyl amine (1mL), to this solution 1,4,5,8-naphthalenetetracarboxylic acid dianhydride (NDA) (1 equiv.) was added

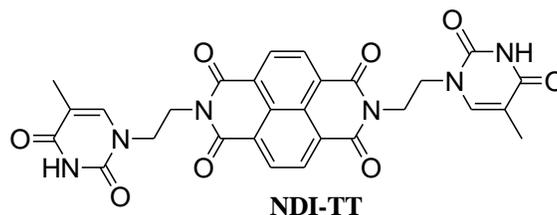
slowly. The resulting solution was heated up to 110 °C and allowed to stir for 6 h. The completion of the reaction was monitored by TLC. The reaction mixture was cooled to room temperature, solution was filtered, and the precipitate was washed with chloroform and methanol. The obtained product (**NDA-AA**) was dried on under vacuum at 40 °C for 12 h. Yellow color powder, Yield 75%. ¹H-NMR (400 MHz, *DMSO-d*₆+*TFA*) δ 4.54 (d, *J* = 5.2 Hz, 4H), 4.64 (d, *J* = 5.2 Hz, 4H), 8.19 (s, 2H), 8.54 (s, 2H), 8.56 (s, 4H), 8.97 (br, 2H), 9.55 (br, 2H). ¹³C-NMR (100 MHz, *DMSO-d*₆+*TFA*) δ 162.9, 150.0, 149.13, 144.7, 130.6, 126.3, 118.2, 42.5, 40.2. LCMS (ESI): *m/z* = 589.25 [*M+H*]⁺ for C₂₈H₂₁N₁₂O₄. Elemental analysis. Found: C, 57.18; H, 3.48, N, 28.50. Calcd: C, 57.14; H, 3.43; N, 28.56 for C₂₈H₂₀N₁₂O₄.

Preparation of Boc-protected thymine-1-ethyl amine (**2**)

To a stirred solution of thymine (0.1 g, 0.79 mmol) and potassium carbonate (0.13 g, 0.95 mmol) in DMF *tert*-butyl -2-bromoethylcarbamate (0.18 g, 0.79 mmol) was added to the above reaction mixture. The reaction mixture was heated up to 65 °C and allowed to stir for 24 hours. After completion of the reaction the solvent was evaporated, the crude product was purified using column chromatography on silica gel using MeOH/CHCl₃ (3:97) as an eluent afford white color solid compound **2** in good yield (45 %).

¹H-NMR (400 MHz, CDCl₃) δ 1.43 (9H, s), 1.90 (d, *J* = 1.2 Hz, 3H), 3.39 (q, *J* = 6.0 Hz, 2H), 3.84 (t, *J* = 6 Hz, 2H), 4.85 (s, 1H), 6.99 (s, 1H), 8.42 (s, 1H). ¹³C-NMR (100 MHz, CDCl₃) δ 164.1, 156.1, 150.9, 141.0, 110.4, 79.9, 48.0, 40.2, 28.3, 12.2 MALDI-TOF-MS: *m/z* = 269.23[*M*]⁺ for C₁₂H₁₉N₃O₄

Preparation of NDI-TT



Boc-protected thyminy-1-ethyl amine **2** (2.2 equiv.) was dissolved in a mixture of DCM/TFA (1:1) (10 mL). The reaction was allowed to stir for 4 h. After completion of the reaction, solvent was evaporated. The trace amount of TFA was removed by the formation of azeotropic mixture with toluene. The crude product was dissolved in DMF (10 mL) containing triethyl amine (1mL), to this solution 1,4,5,8-naphthalenetetracarboxylic acid dianhydride (NDA) (1 equiv.) was added slowly. The resulting solution was heated up to 110 °C and allowed to stir for 6 h. The completion of the reaction was monitored by TLC. The reaction mixture was cooled to room temperature, solution was filtered, and the precipitate was washed with chloroform and methanol. The obtained product **NDA-TT** was dried under vacuum at 40 °C for 10 h. White colour powder, Yield 58%. ¹H-NMR (400 MHz, *DMSO-d*₆) δ 1.64 (d, *J* = 0.8 Hz, 6H), 4.02 (t, *J* = 4.8 Hz, 4H), 4.36 (t, *J* = 5.2 Hz, 4H), 7.58 (d, *J* = 0.8 Hz, 2H), 8.67 (s, 4H), 11.06 (s, 2H). ¹³C-NMR (100 MHz, *DMSO-d*₆) δ 164.2, 162.7, 151.4, 141.5, 130.5, 126.2, 108.6, 45.8, 40.2, 11.8 MALDI-TOF-MS: *m/z* = 571.066 [M+H]⁺ for C₂₈H₂₃N₆O₈. Elemental analysis. Found: C, 58.90; H, 3.88, N, 14.69. Calcd: C, 58.95; H, 3.89; N, 14.73 for C₂₈H₂₂N₆O₈.

PNA dimers **PNA-TT** and **PNA-AA** were prepared following the procedure reported in literature.¹

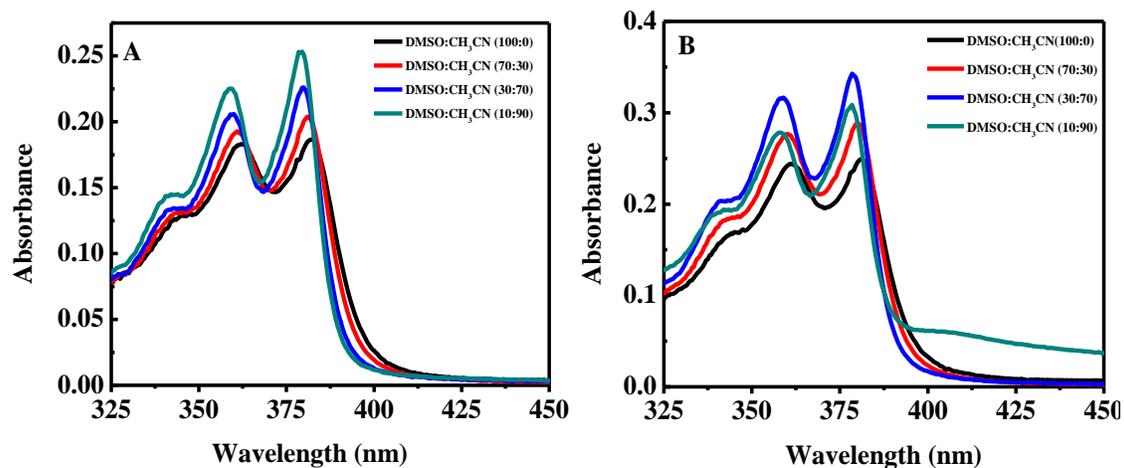


Fig. S1 UV-vis absorption spectra of **NDI-conjugates**. (A) **NDI-AA** in DMSO/ CH₃CN system. (B) **NDI-TT** in DMSO/ CH₃CN system.

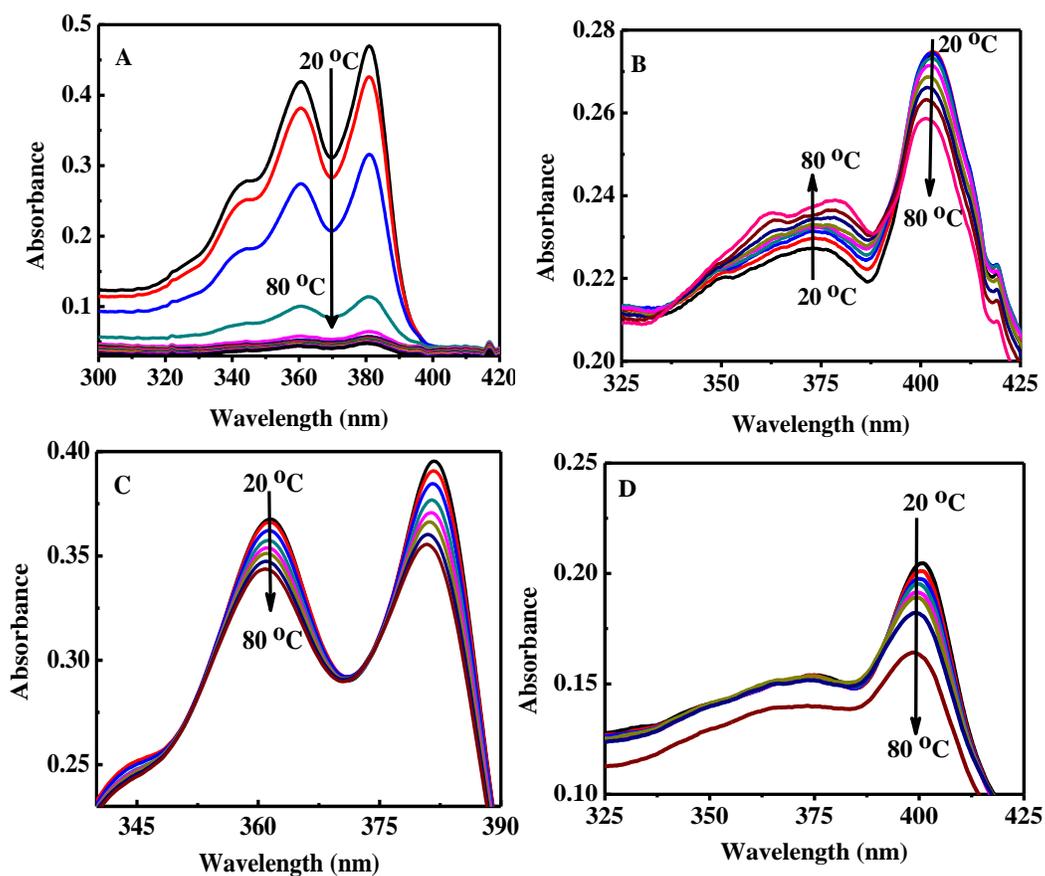


Fig. S2 Temperature dependent UV-vis absorption spectra. (A) and (B) spectra of NDI-AA and NDI-TT respectively in DMSO/Water solvent. (C) and (D) spectra of NDI-AA/PNA-TT and NDI-TT/PNA-AA respectively in DMSO/Water solvent.

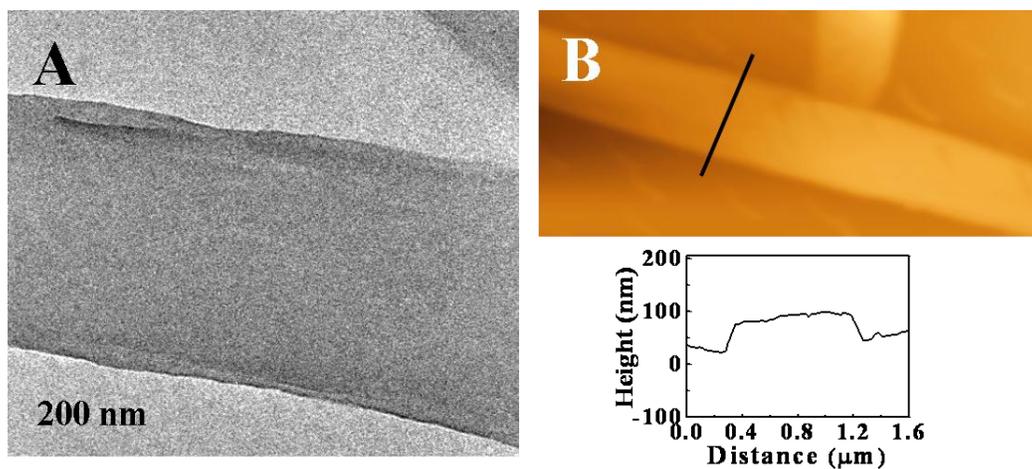


Fig. S3 (A) TEM micrograph of the NDI-AA nanoribbon. (B) AFM image of NDI-AA nanoribbon and the corresponding height profile (along the black trace).

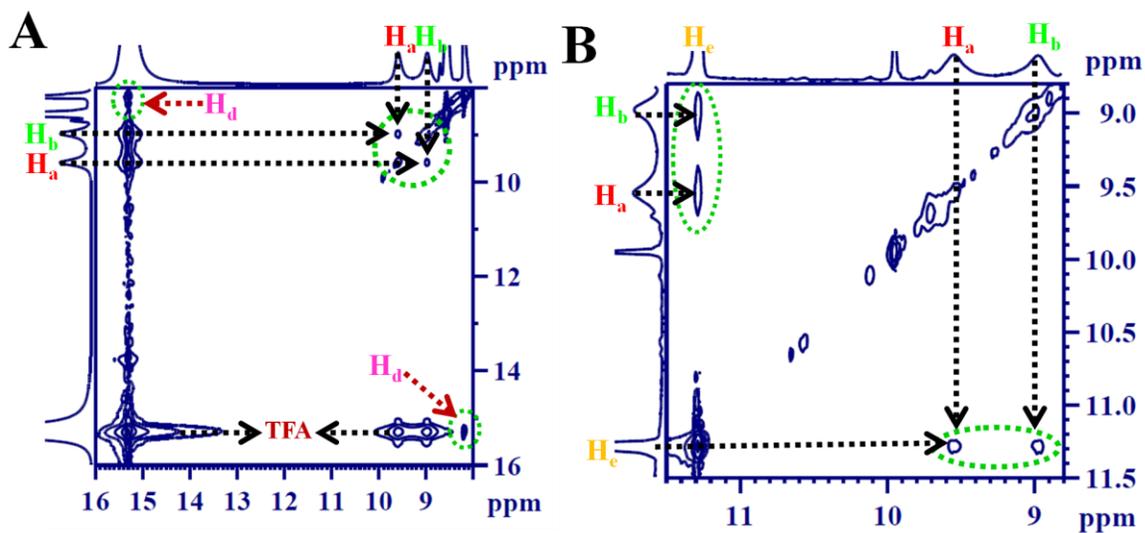


Fig. S4 (A) 2D-NOESY spectra of NDI-AA (with trace amount of TFA) in $DMSO-d_6$. (B) 2D-NOESY spectra of NDI-AA/PNA-TT (1:1) in $DMSO-d_6$.

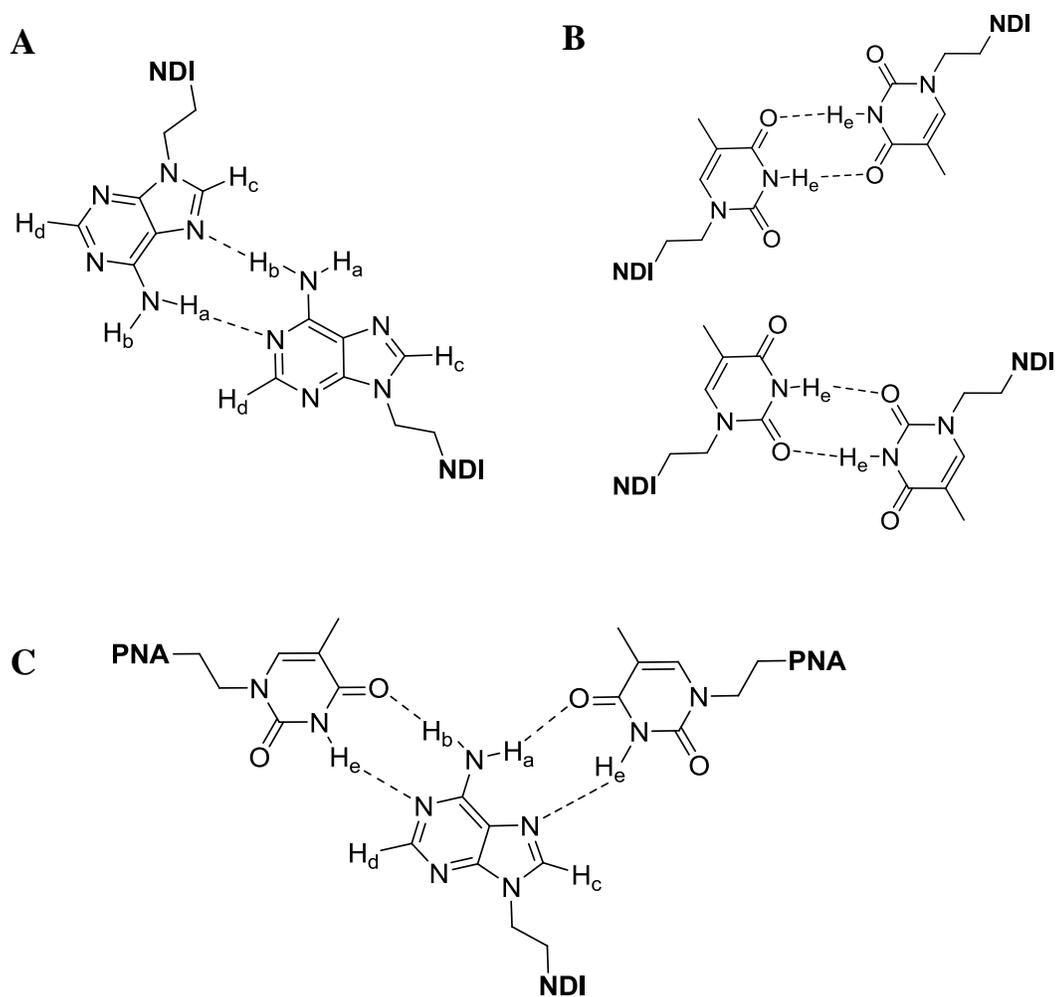
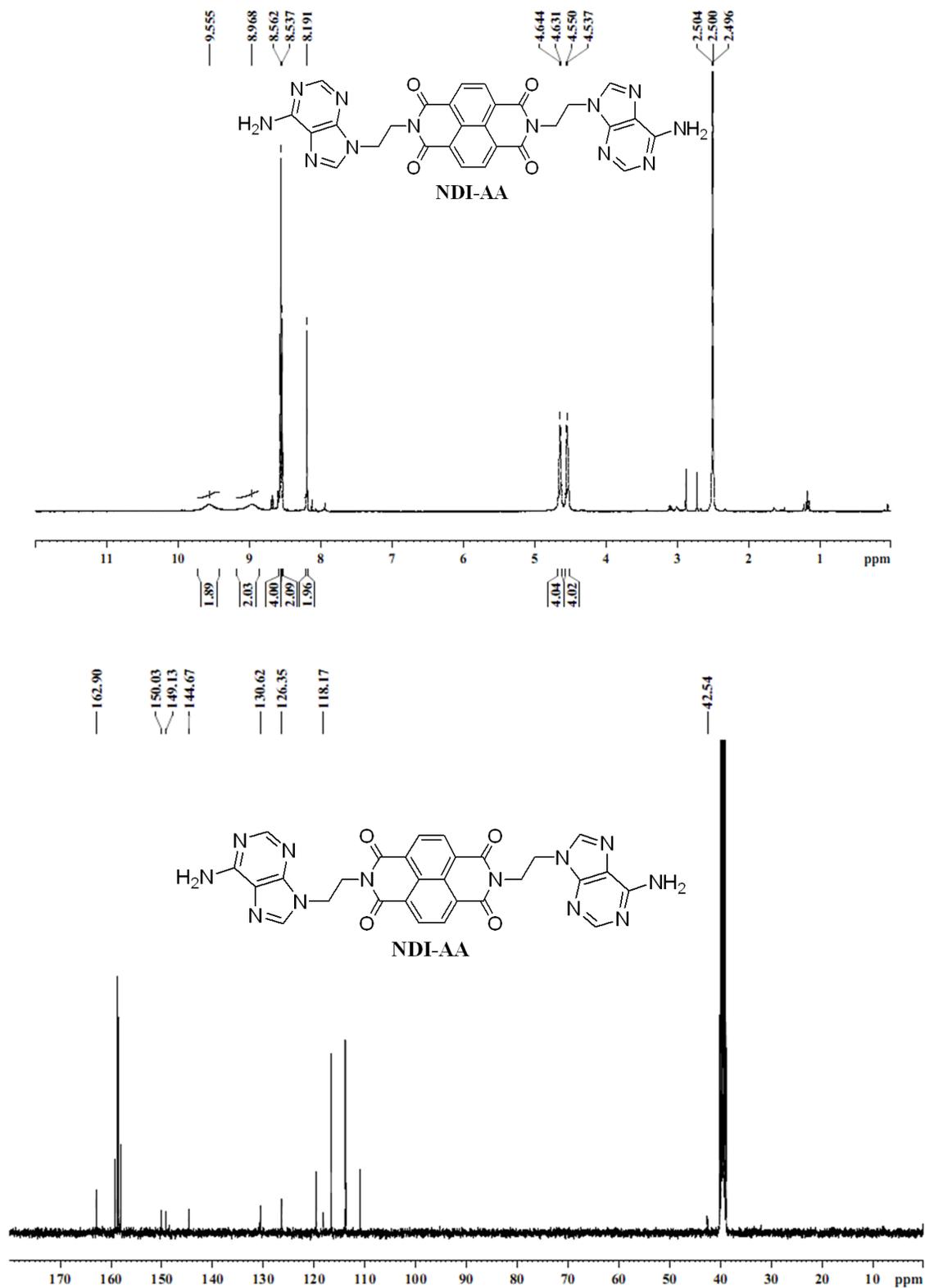
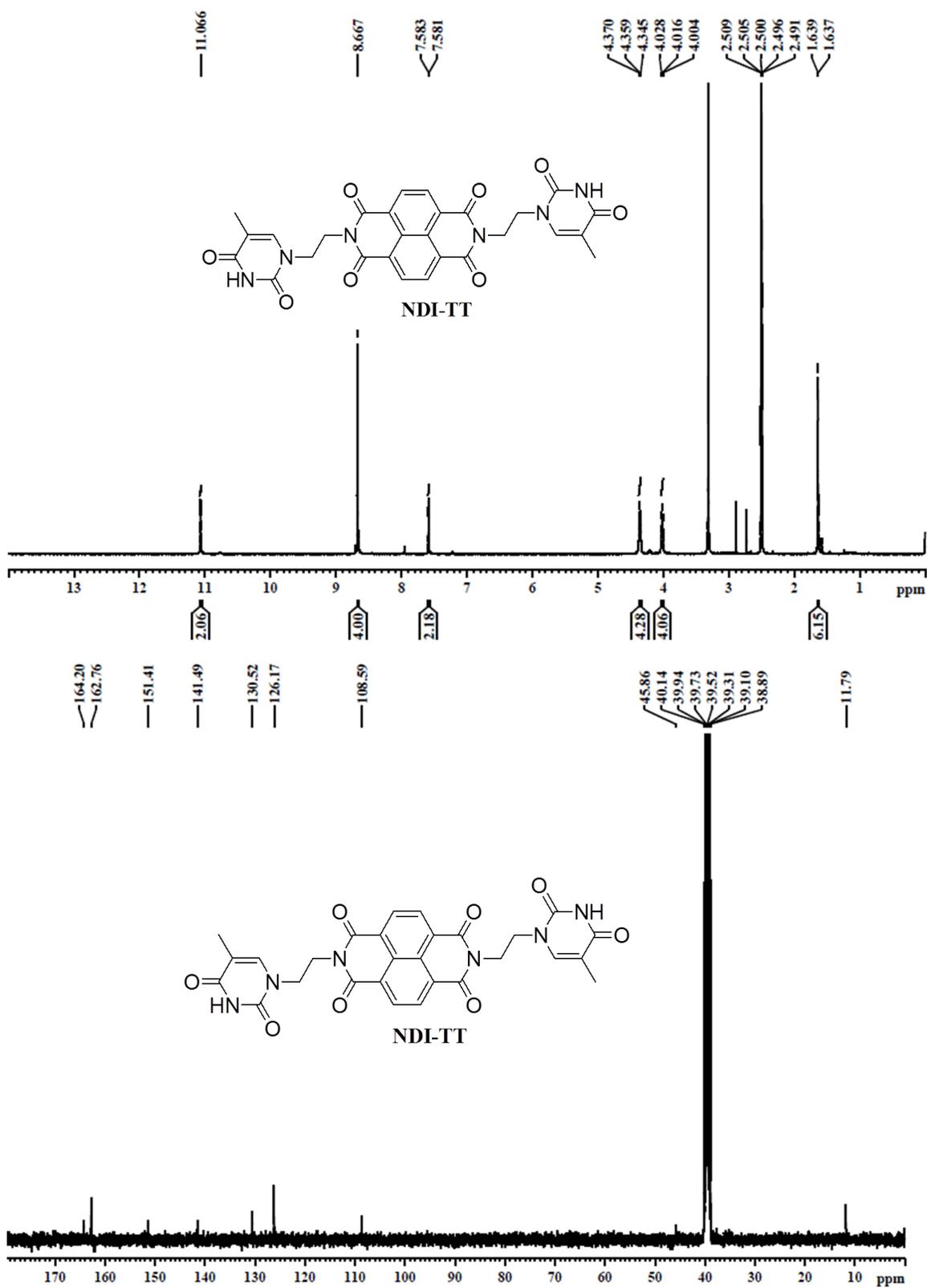


Fig. S5 Mode of hydrogen bonding interactions among adenine and adenine-thymine nucleobases in NDI and PNA conjugates. (A) Intermolecular adenine-adenine hydrogen bonding among **NDI-AA** molecules. (B) Intermolecular thymine-thymine hydrogen bonding among **NDI-TT** molecules. (C) Intermolecular adenine-thymine hydrogen bonding interactions in **PNA-TT** templated **NDI-AA** complex.

^1H and ^{13}C NMR-spectra of NDI-AA

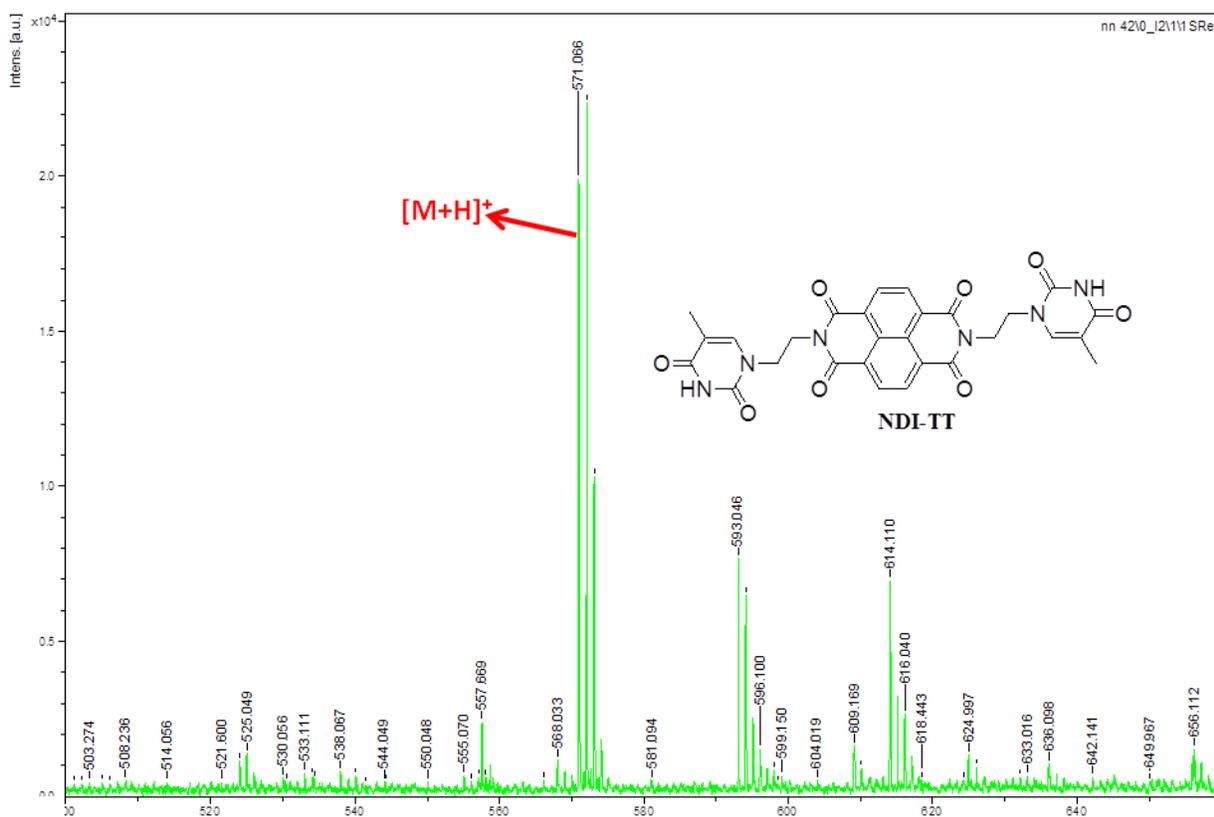
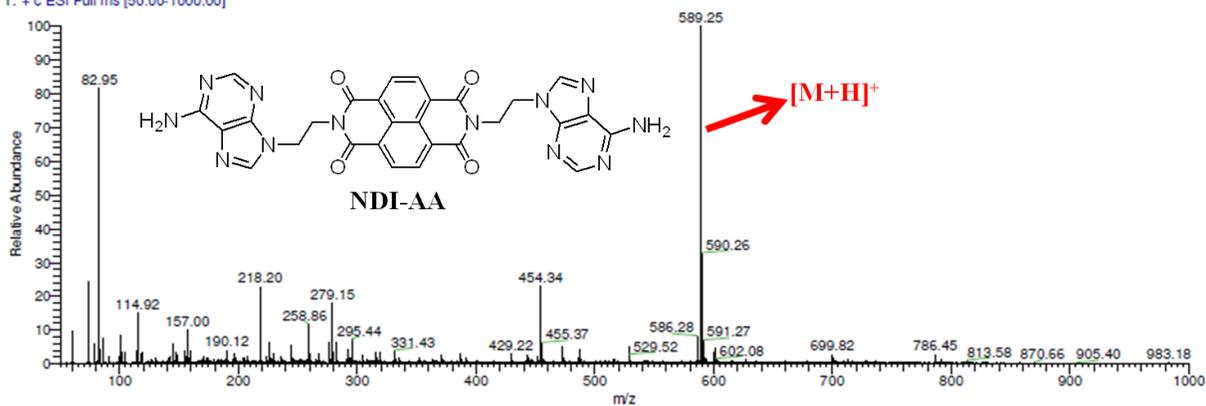


^1H and ^{13}C NMR-spectra of NDI-TT



Mass spectra of NDI-AA and NDI-TT

Nagarjun #1010-1279 RT: 16.87-21.42 AV: 270 NL: 2.23E6
T: + c ESI Full ms [50.00-1000.00]



References

- 1 K. L. Dueholm, M. Egholm, C. Behrens, L. Christensen, H. F. Hansen, T. Vulpius, K. H. Petersen, R. H. Berg, P. E. Nielsen and O. Buchardt, *J. Org. Chem.*, 1994, **59**, 5767–5773.