BEILSTEIN JOURNAL OF ORGANIC CHEMISTRY

Supporting Information

for

Synthesis and properties of oligonucleotides modified with an *N*-methylguanidine-bridged nucleic acid (GuNA[Me]) bearing adenine, guanine, or 5-methylcytosine nucleobases

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Beilstein J. Org. Chem. **2021,** *17,* 622–629. doi:10.3762/bjoc.17.54

^{1}H, ^{13}C, and ^{32}P NMR spectra for all new compounds, HPLC charts and MALDI–TOF mass data for all new oligonucleotides, UV melting curves of the duplexes formed between GuNA[Me]-modified oligonucleotides and ssDNAs (or ssRNAs), and CD spectra of ON4/ssRNA and ON4/ssDNA

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1. ^{1}H NMR, ^{13}C NMR, and ^{31}P NMR spectra of new compounds

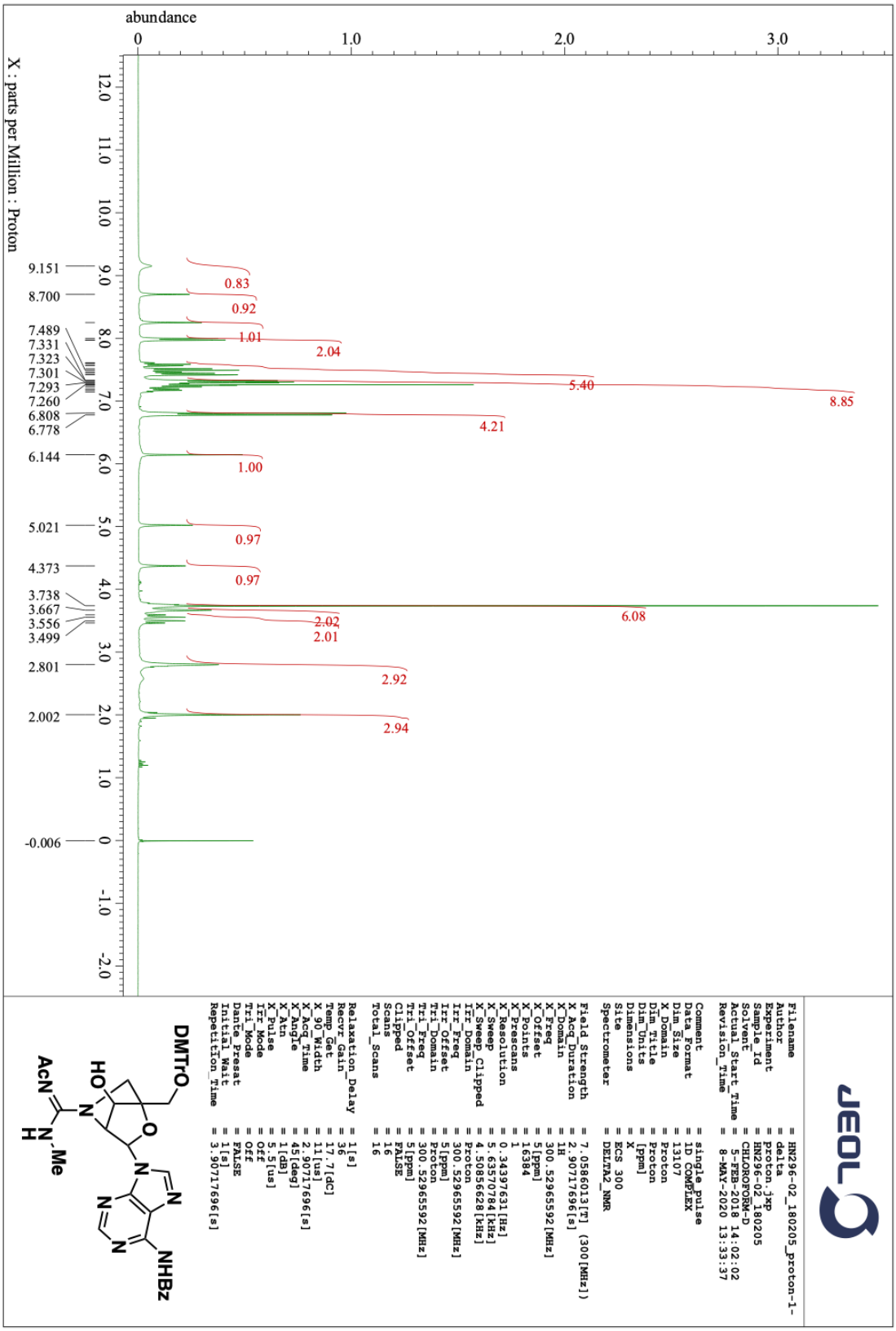


Figure S1: Compound **2a** (^{1}H NMR, $CDCl_3$, 300 MHz)

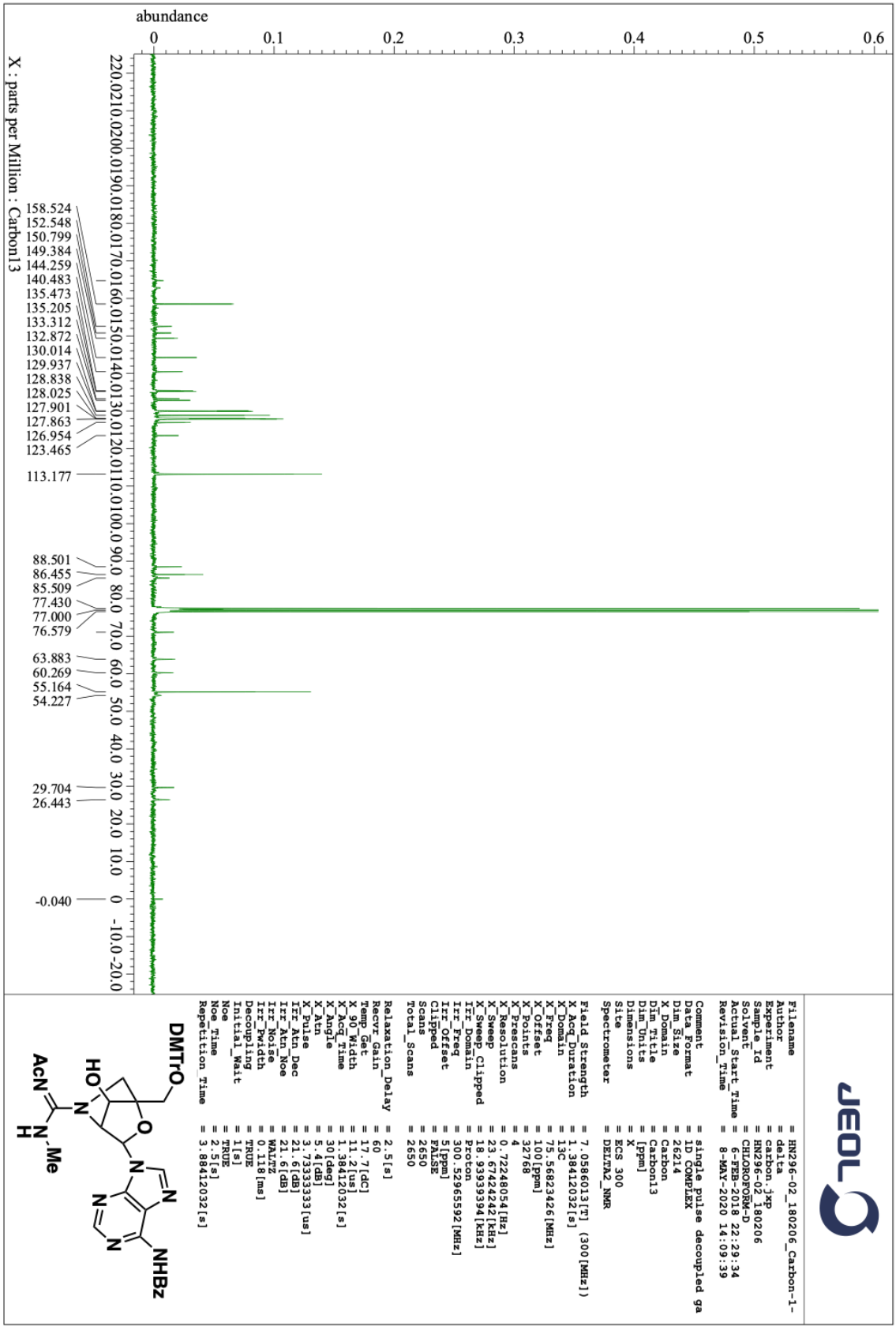


Figure S2: Compound **2a** (^{13}C NMR, $CDCl_3$, 76 MHz)

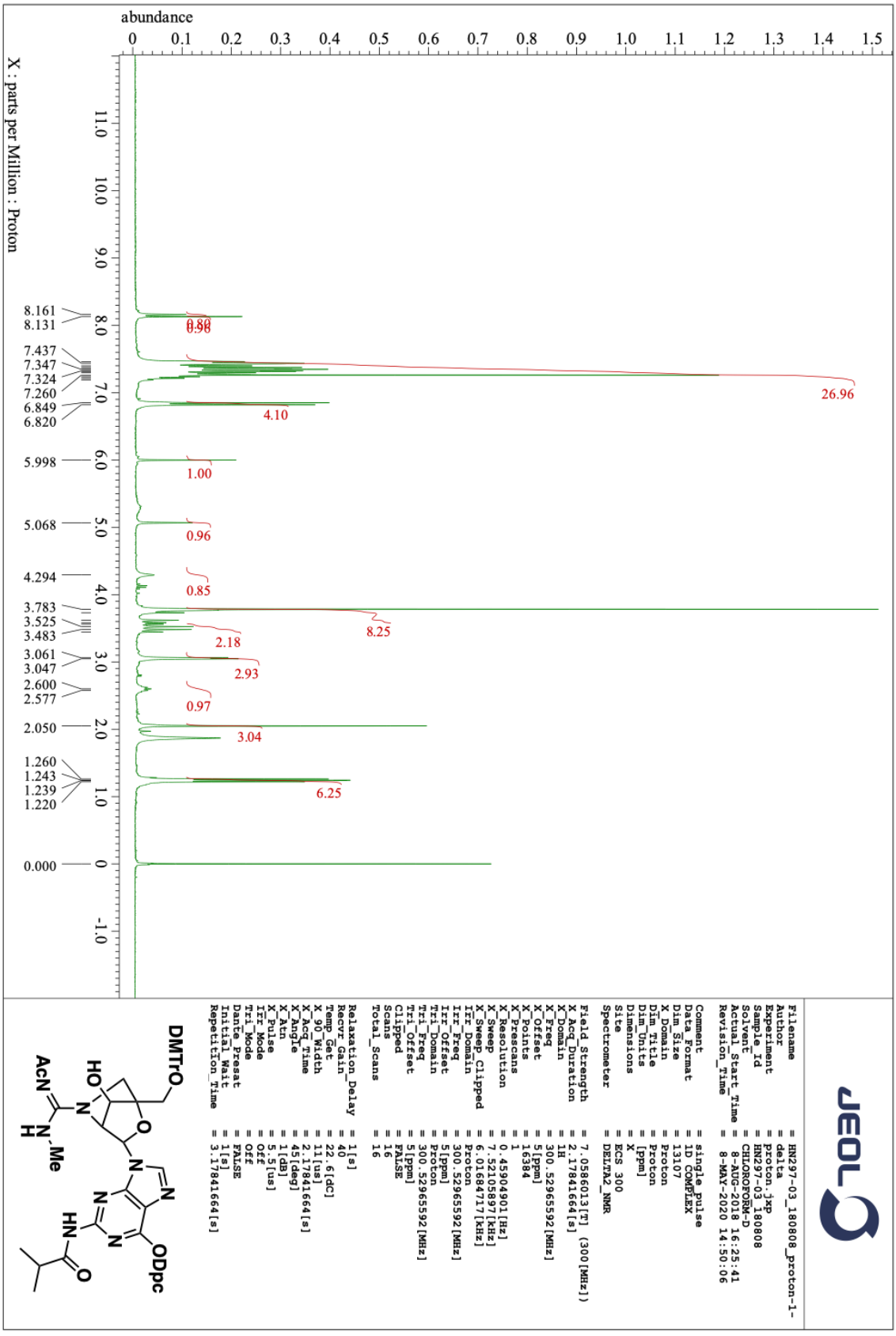


Figure S3: Compound **2b** (^{1}H NMR, $CDCl_3$, 300 MHz)

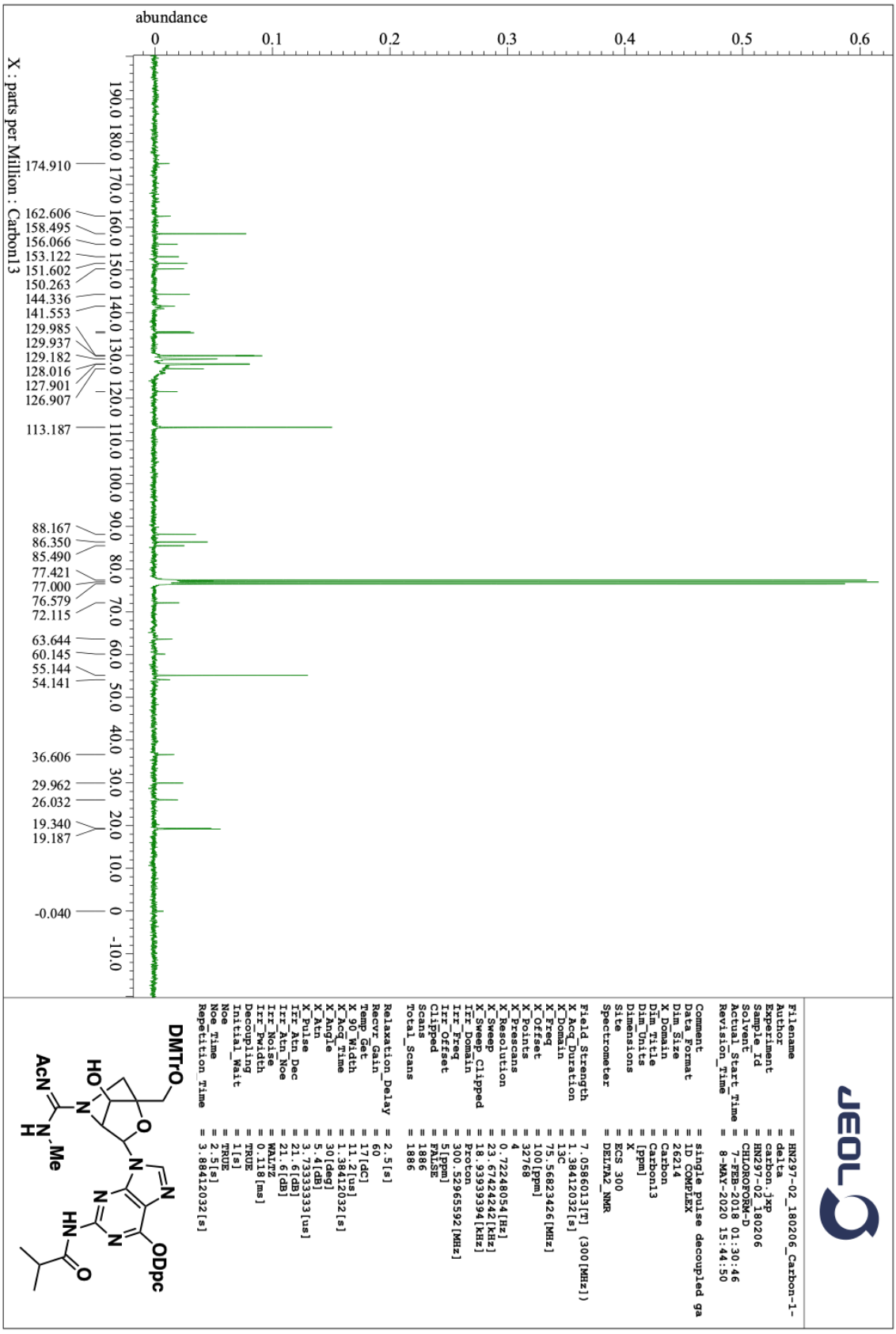


Figure S4: Compound **2b** (^{13}C NMR, $CDCl_3$, 76 MHz)

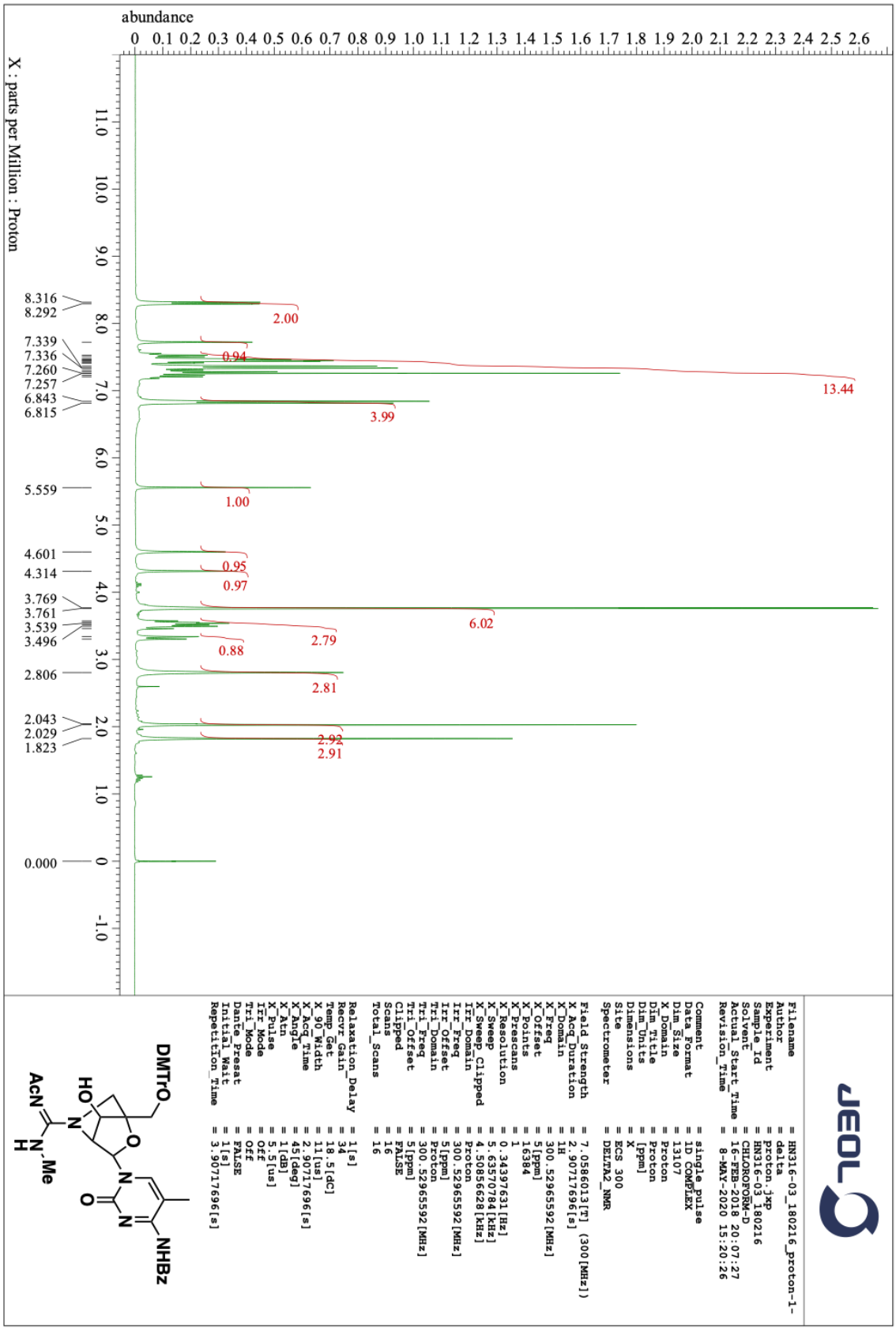


Figure S5: Compound **2c** (^{1}H NMR, $CDCl_3$, 300 MHz)

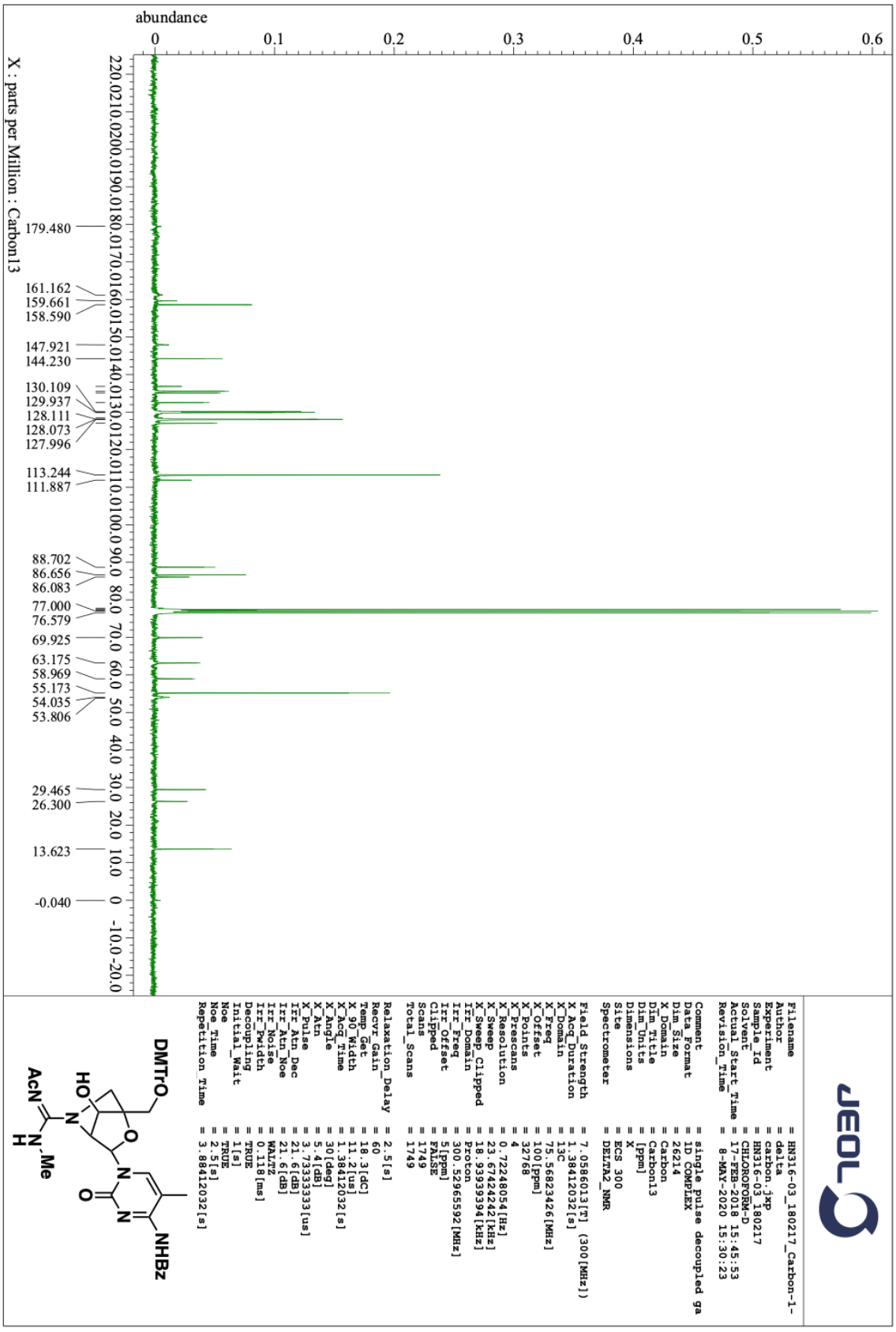


Figure S6: Compound **2c** (^{13}C NMR, $CDCl_3$, 76 MHz)

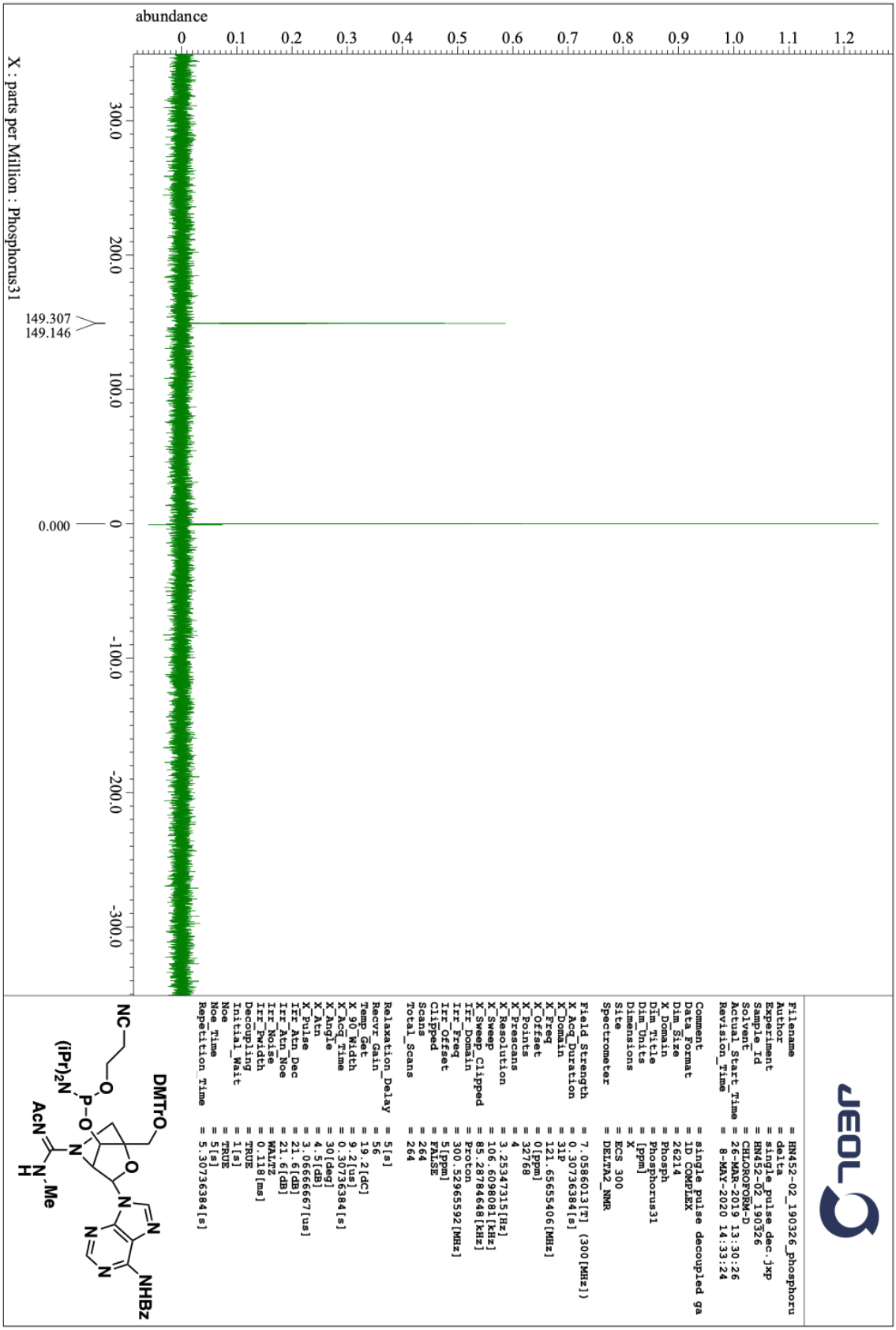


Figure S7: Compound **3a** (^{31}P NMR, $CDCl_3$, 122 MHz)

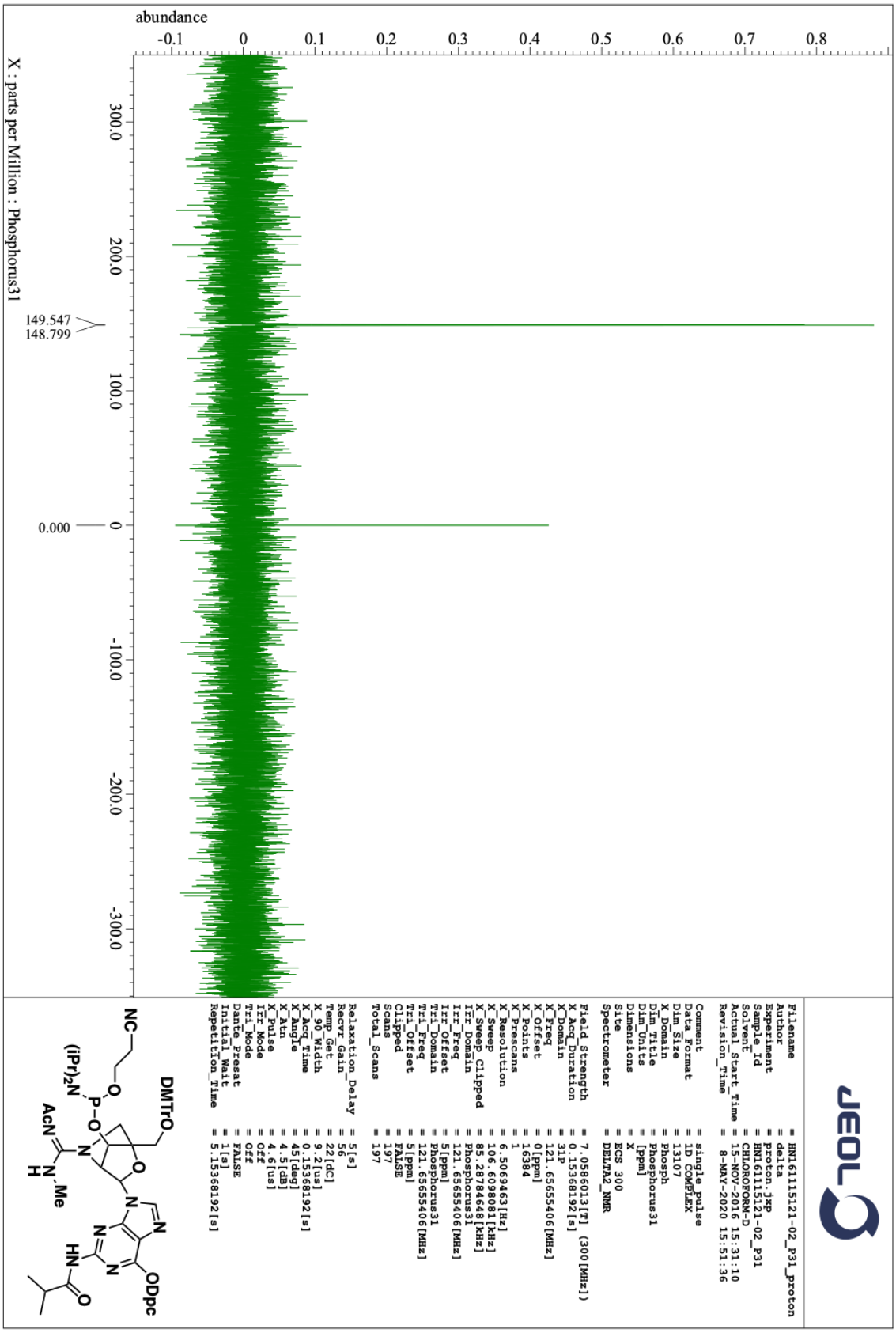


Figure S8: Compound **3b** (^{31}P NMR, $CDCl_3$, 122 MHz)

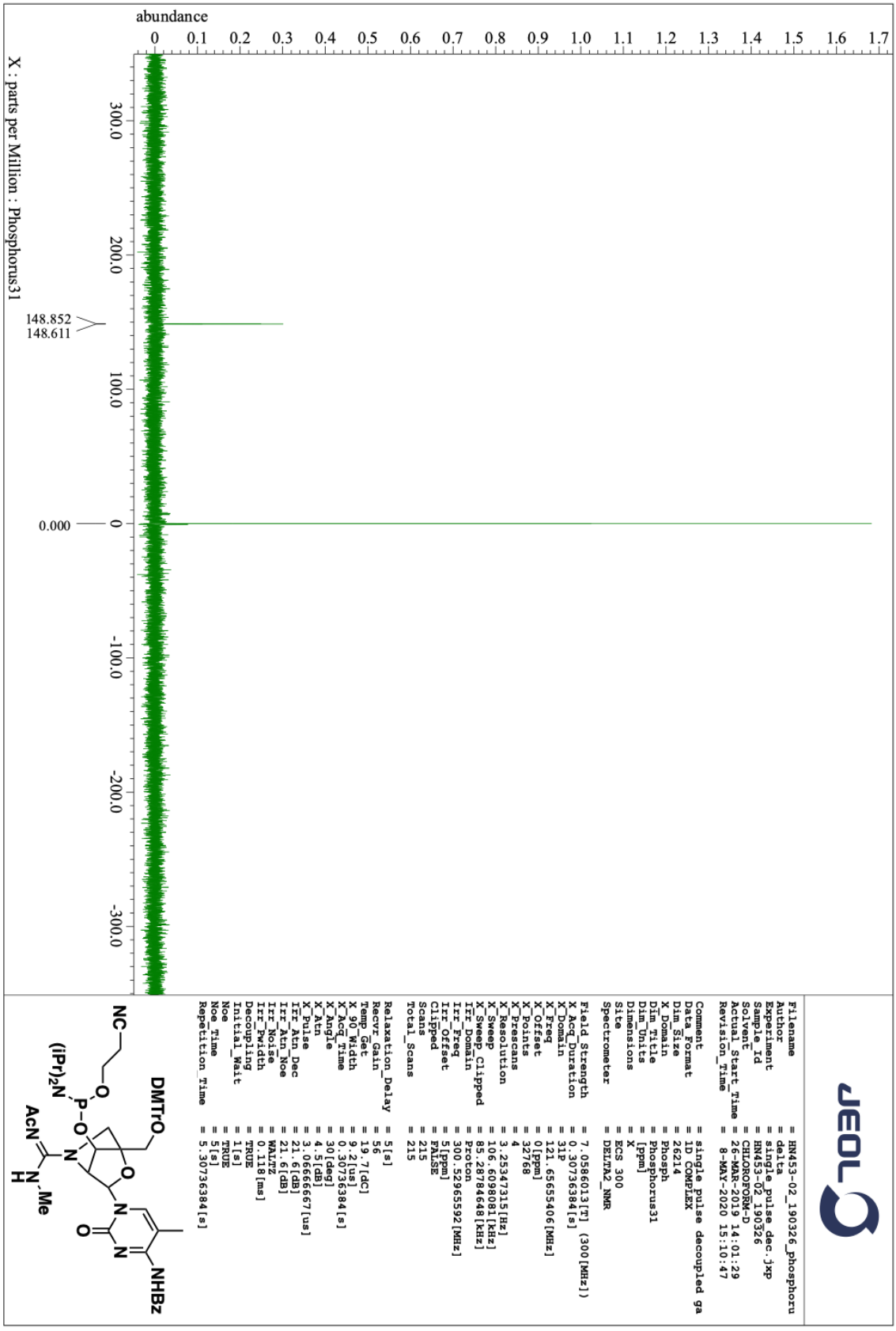
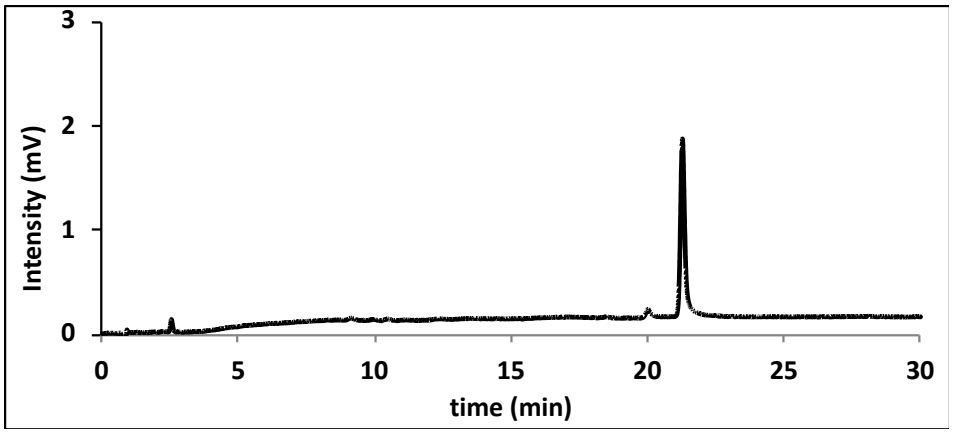


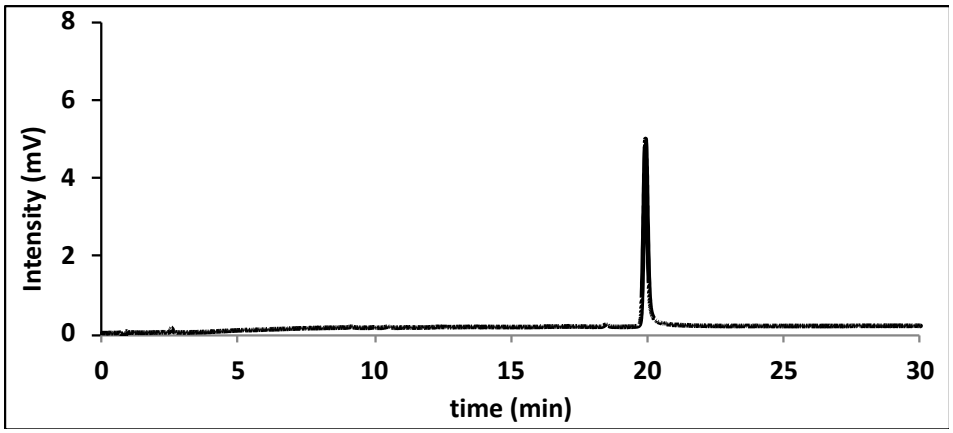
Figure S9: Compound **3c** (^{31}P NMR, $CDCl_3$, 122 MHz)

2. Characterisation of oligonucleotides

ON1 5'-d(GCG TTA TTT GCT)-3'



ON2 5'-d(GCG TTG TTT GCT)-3'



ON3 5'-d(GCG TT^{m}C TTT GCT)-3'

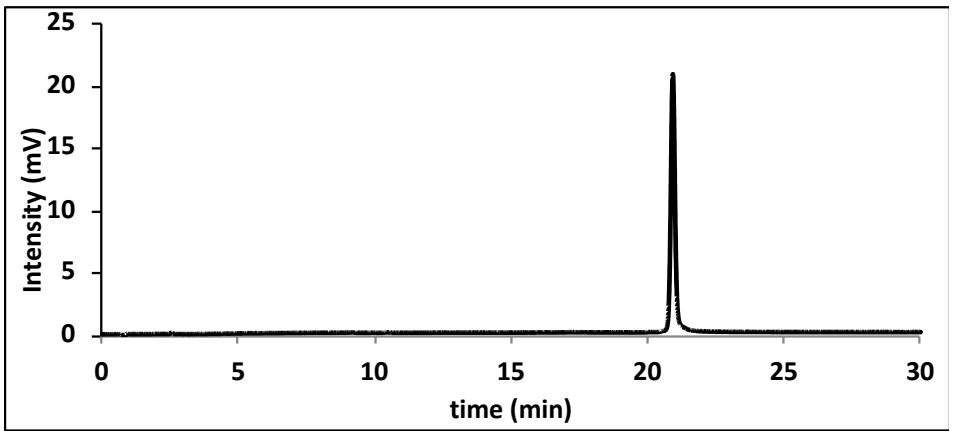
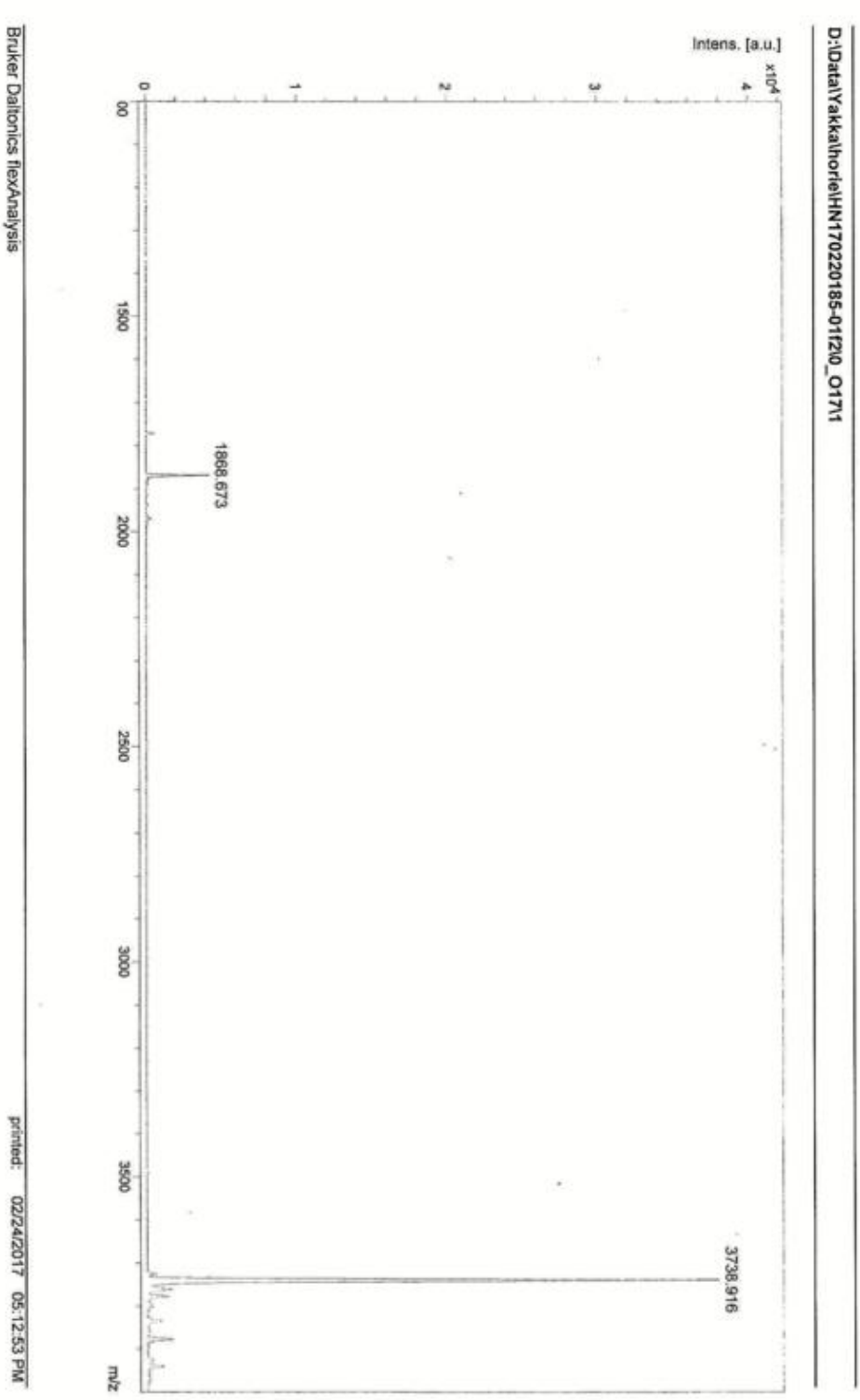


Figure S10: HPLC charts of all new oligonucleotides. HPLC conditions: Reversed-phase HPLC (Waters XBridge™ C18 column) with a linear gradient of acetonitrile (5 to 10% over 5 min, then 10 to 15% over 20 min, then 15 to 15%, over 5 min) in 0.1 M triethylammonium acetate buffer (pH 7.0). A, G, and ^{m}C indicate GuNA[Me] modifications.

ON1 5’-d(GCG TTA TTT GCT)-3’ (A)

ON2 5’-(GCG TTG TTT GCT)-3’ (B)



ON3 5’-d(GCG TT^{m}C TTT GCT)-3’ (C)

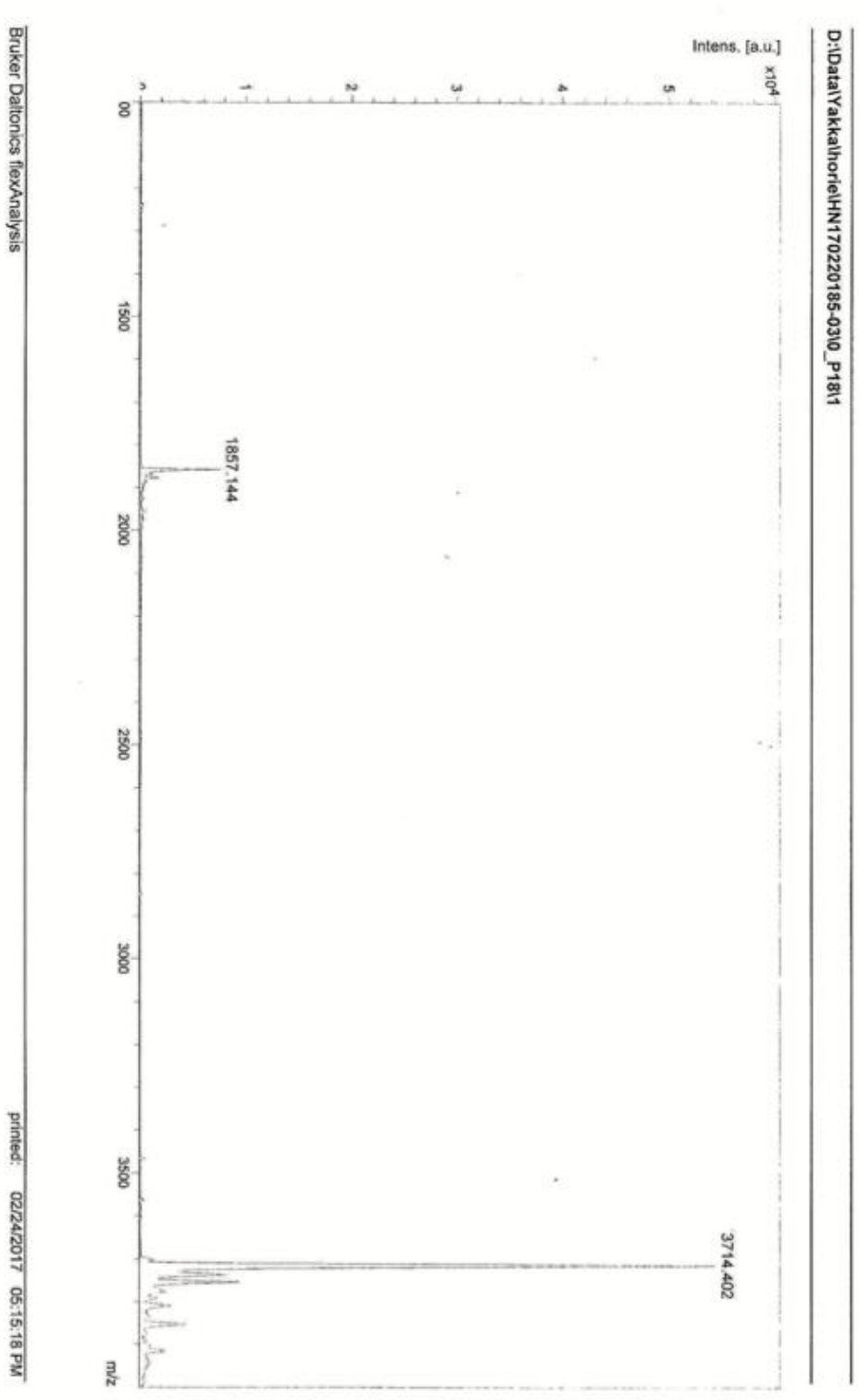


Figure S11: MALDI-TOF-MS charts of all new oligonucleotides. **ON1** 5’-d(GCG TTA TTT GCT)-3’ (A), **ON2** 5’-(GCG TTG TTT GCT)-3’ (B), **ON3** 5’-d(GCG TT^{m}C TTT GCT)-3’ (C); A, G, and ^{m}C indicate GuNA[Me] modifications.

3. UV melting experiments

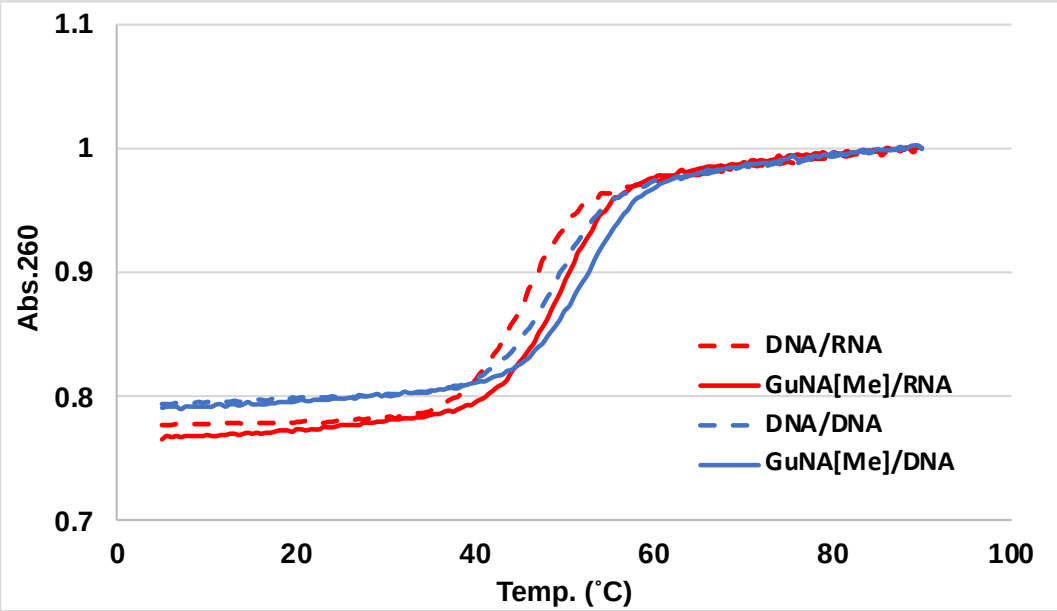


Figure S12: Normalized UV melting curves for the duplexes formed between **ON1**/**ON6** and the complementally DNA or RNA strands. The sequences are 5'-d(GCG TTA TTT GCT)-3' and 5'-r/d(AGC AAA YAA CGC)-3', respectively.

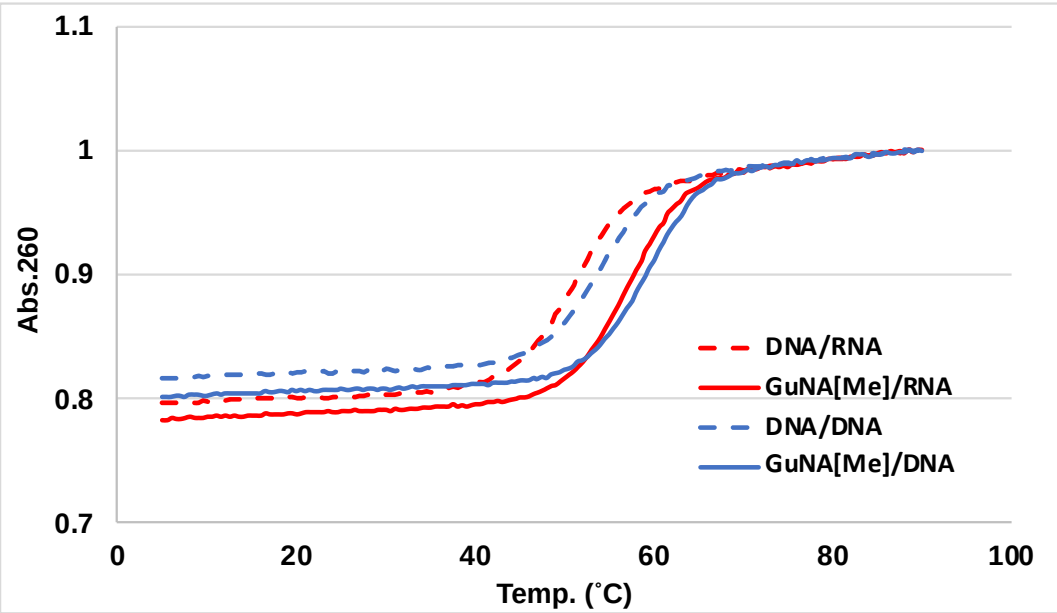


Figure S13: Normalized UV melting curves for the duplexes formed between **ON2**/**ON7** and the complementally DNA or RNA strands. The sequences are 5'-d(GCG TTG TTT GCT)-3' and 5'-r/d(AGC AAA CAA CGC)-3', respectively. Y indicates U for RNA, and T for DNA.

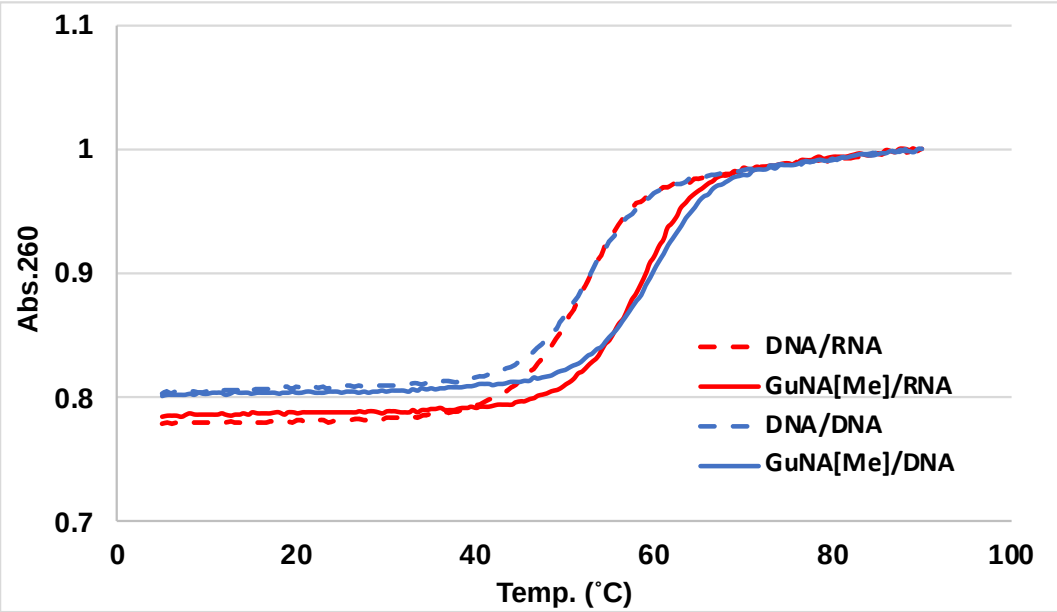


Figure S14: Normalized UV melting curves for the duplexes formed between **ON3**/**ON8** and the complementally DNA or RNA strands. The sequences are 5'-d(GCG TT^{m}CTTT GCT)-3' and 5'-r/d(AGC AAA GAA CGC)-3', respectively.

4. CD spectral analysis

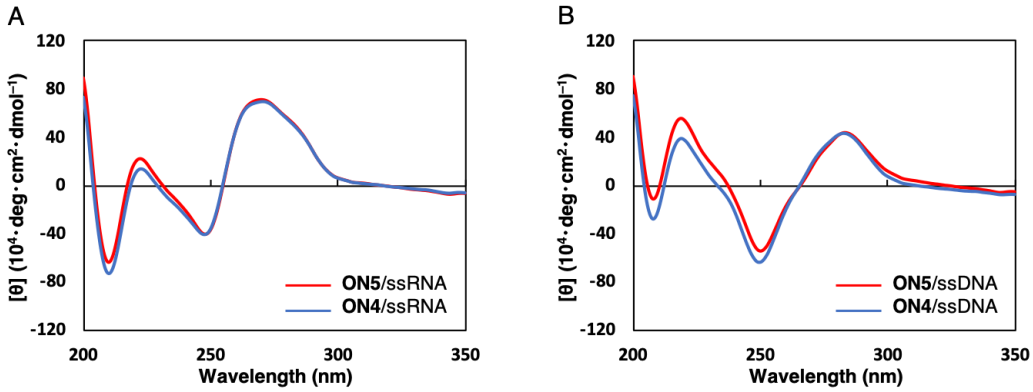


Figure S15: CD spectra of the **ON5**/ssRNA, **ON4**/ssRNA, **ON5**/ssDNA and **ON4**/ssDNA duplexes. Conditions: 10 mM sodium phosphate buffer (pH 7.2), 100 mM NaCl, 4 µM each oligonucleotide. Sequences of the complementary ssRNA and ssDNA are 5'-r(AGC AAA AAA CGC)-3' and 5'-d(AGC AAA AAA CGC)-3', respectively.