

## Supplementary information

# Synthesis and properties of oligodiaminogalactoses that bind to A-type oligonucleotide duplexes

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<sup>b</sup> Department of Neurology and Neurological Science, Graduate School of Medical and Dental Sciences, Tokyo Medical and Dental University, 1-5-45, Yushima, Bunkyo-Ku, Tokyo 113-8519, Japan

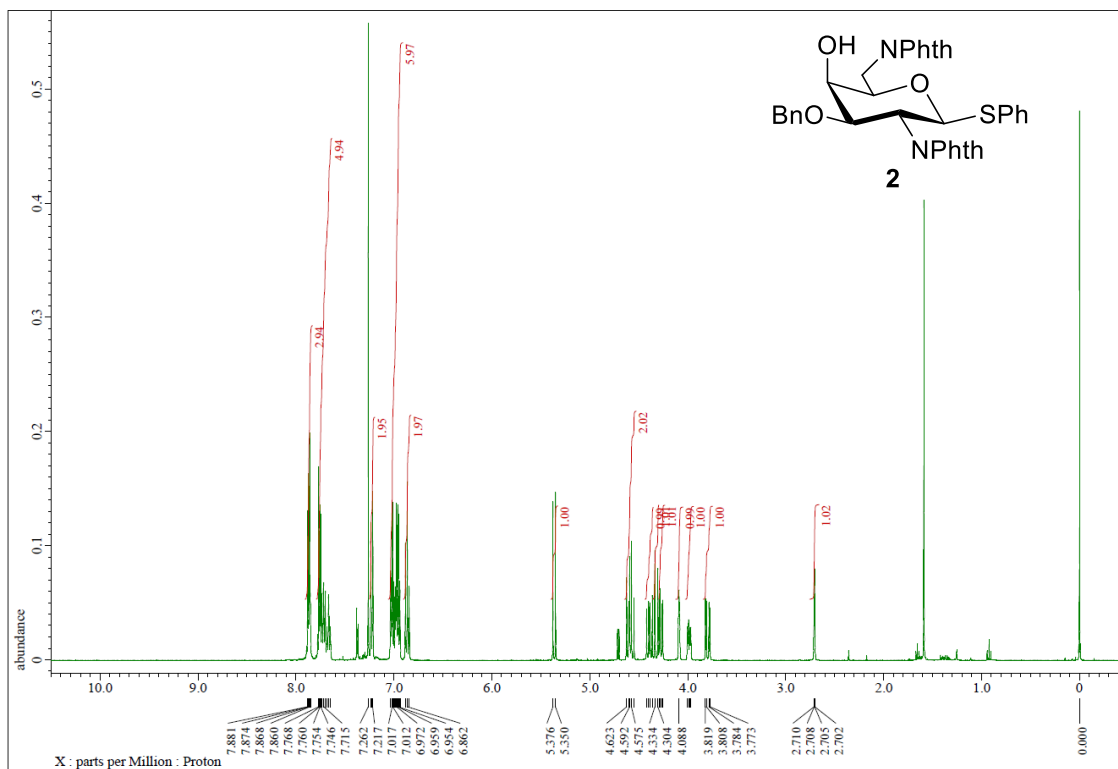
\*twada@rs.tus.ac.jp

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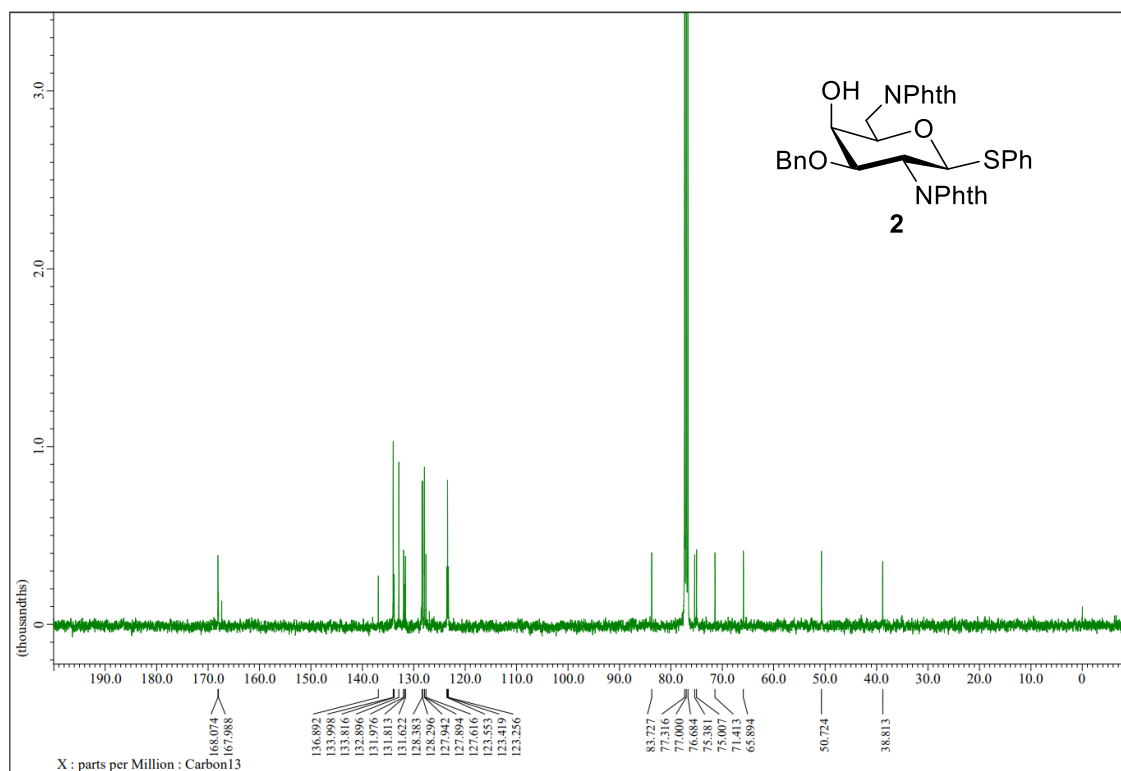
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# 1. <sup>1</sup>H, <sup>13</sup>C NMR spectra

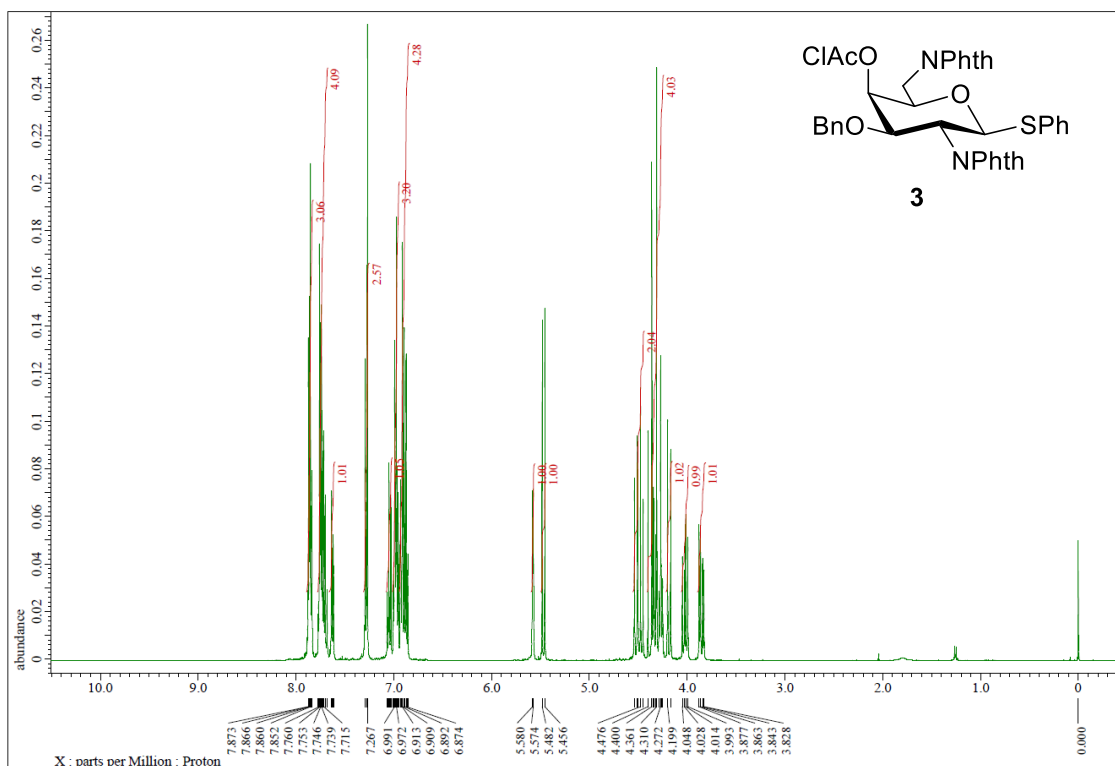
<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)



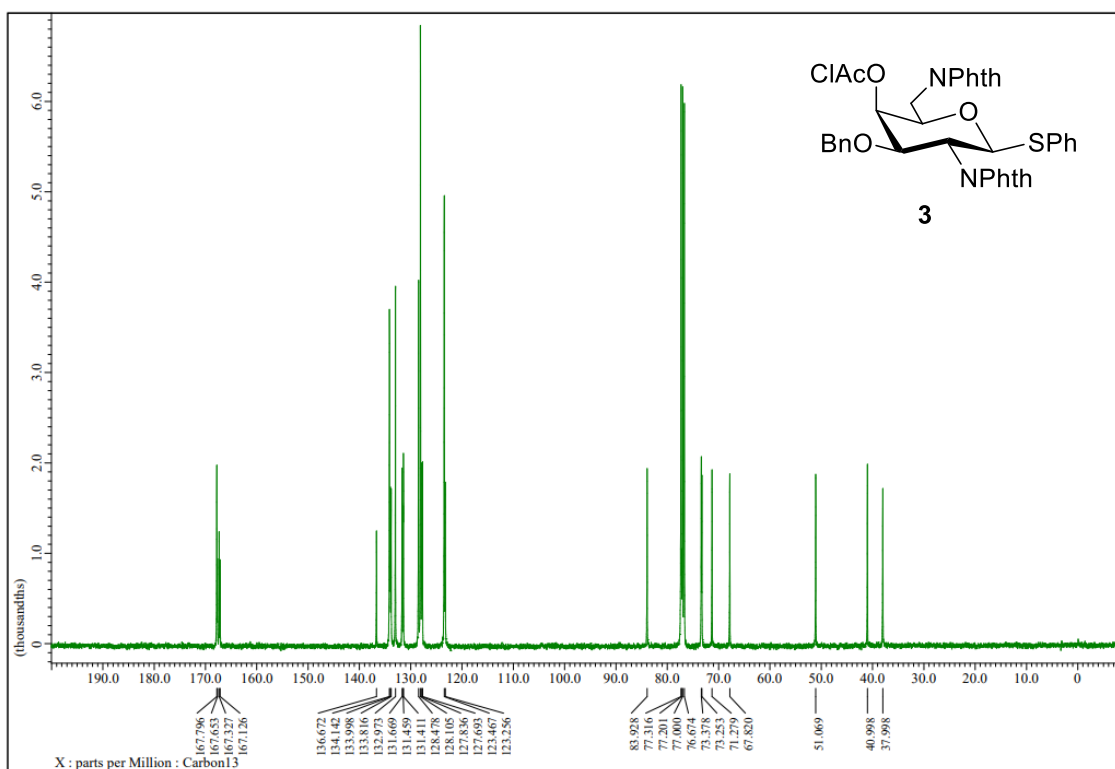
<sup>13</sup>C {<sup>1</sup>H} NMR (101 MHz, CDCl<sub>3</sub>)



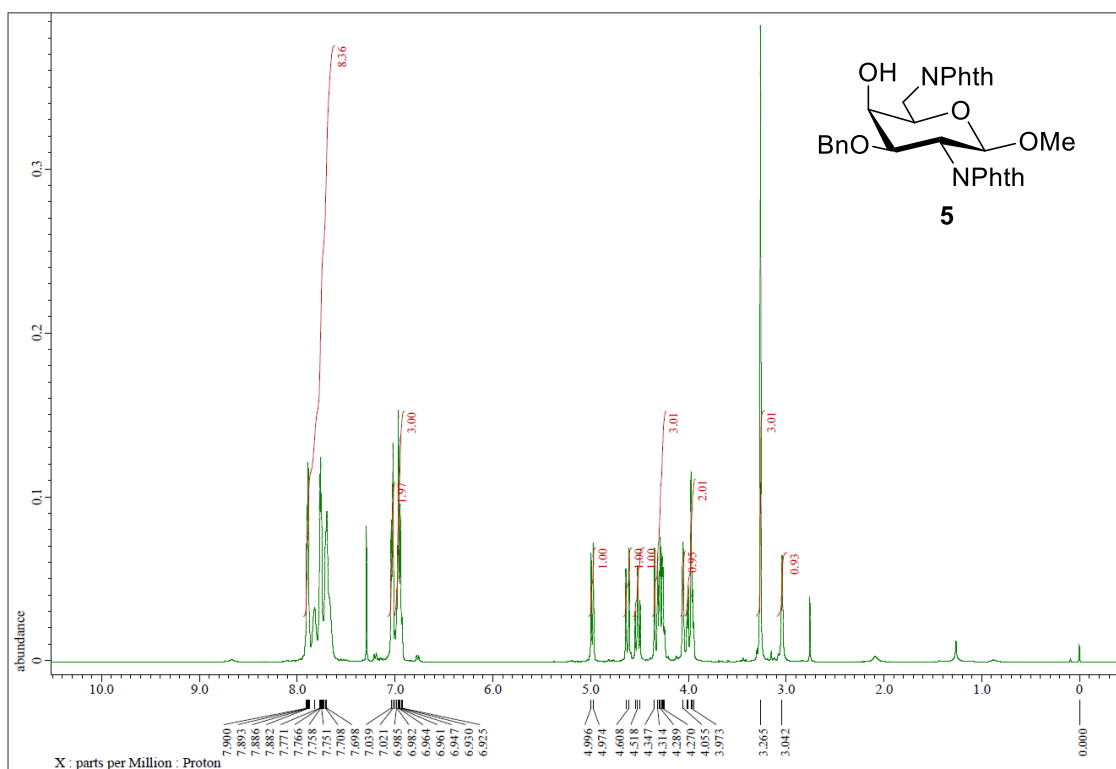
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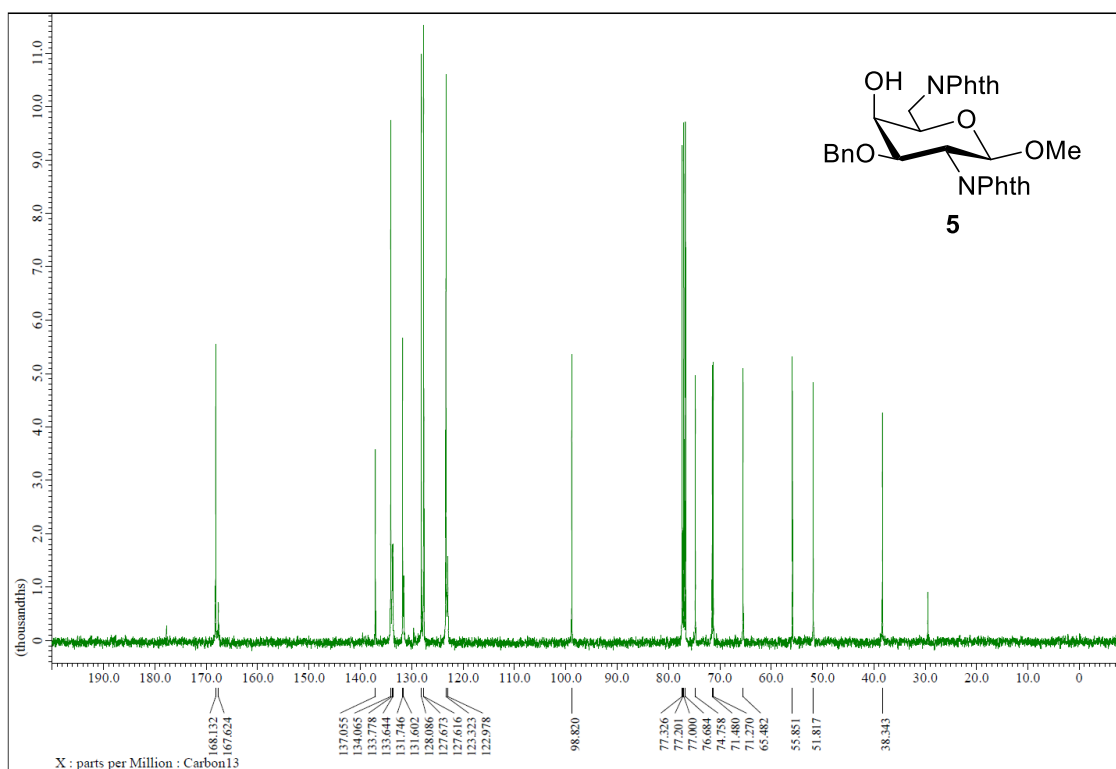
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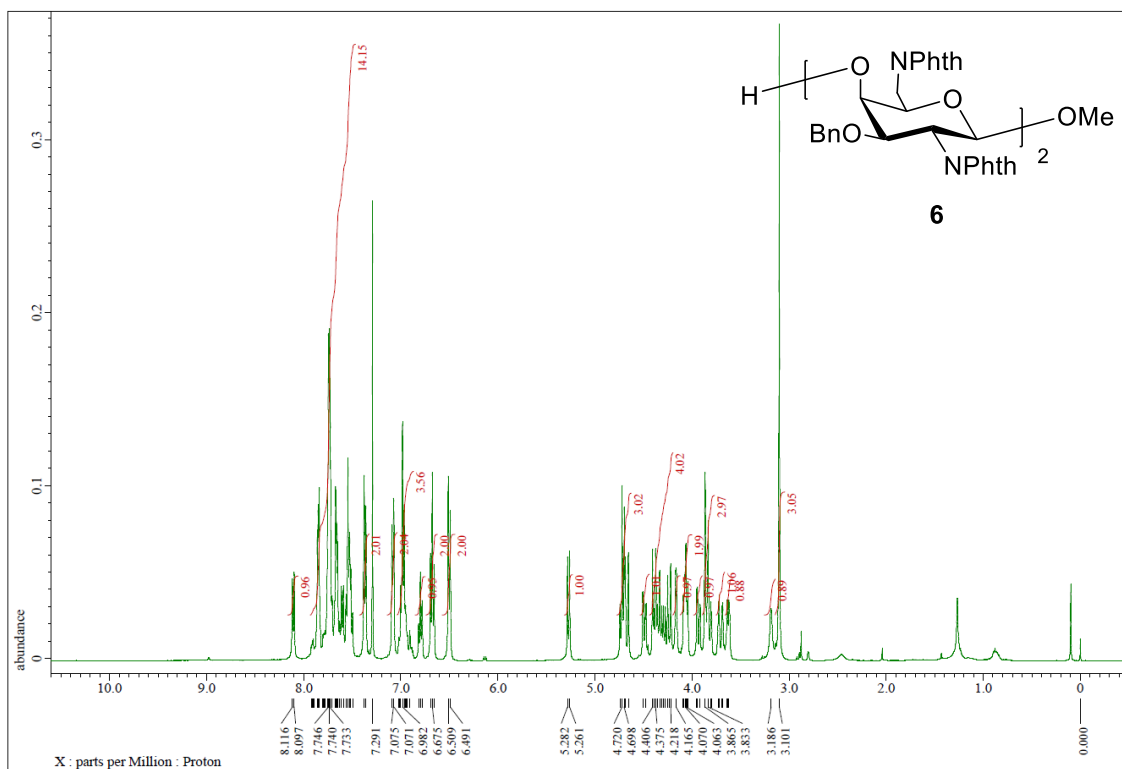
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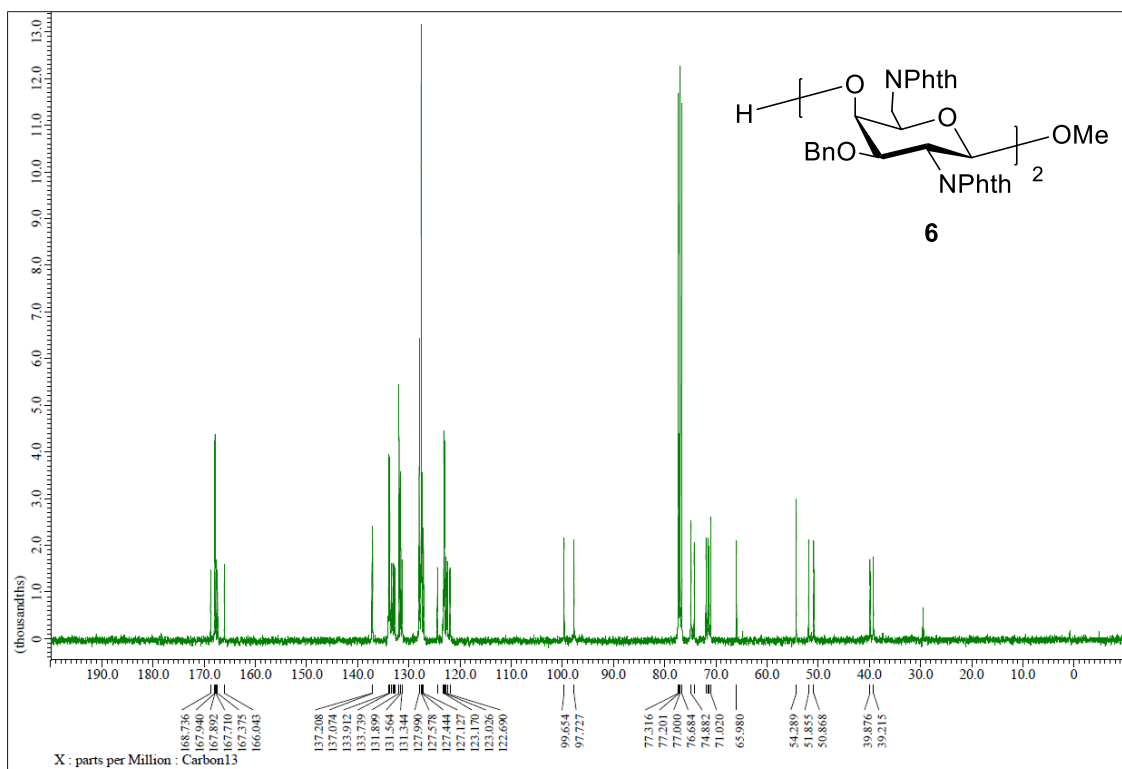
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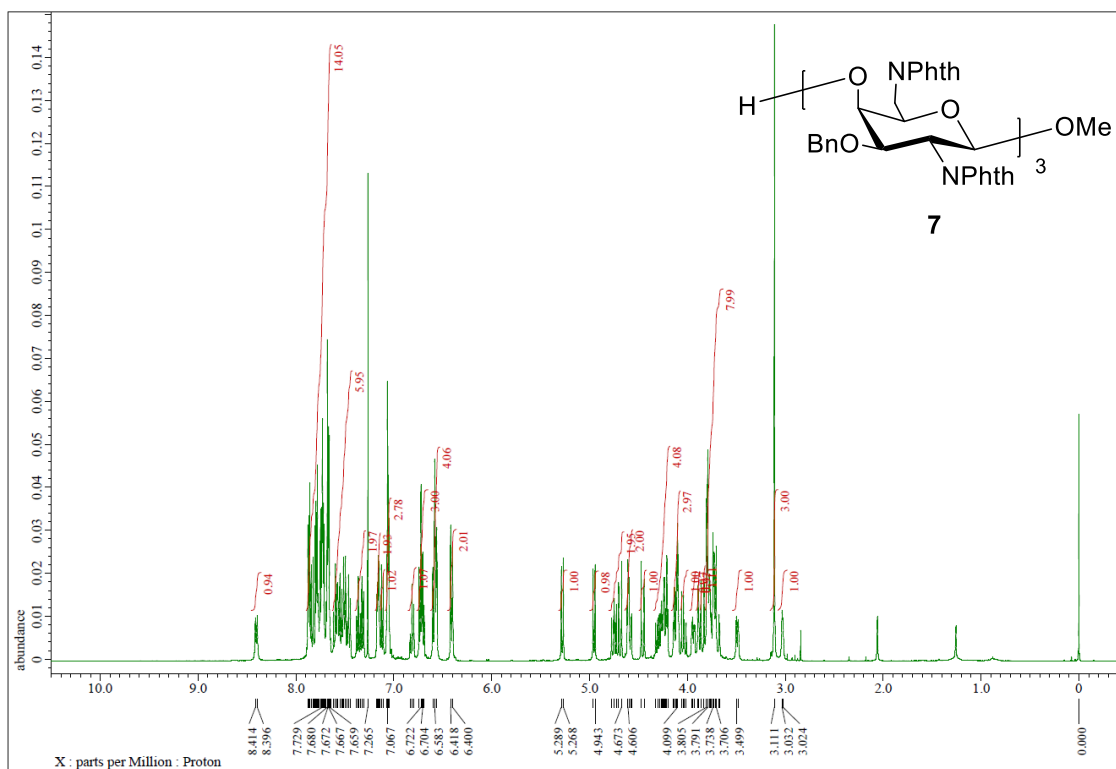
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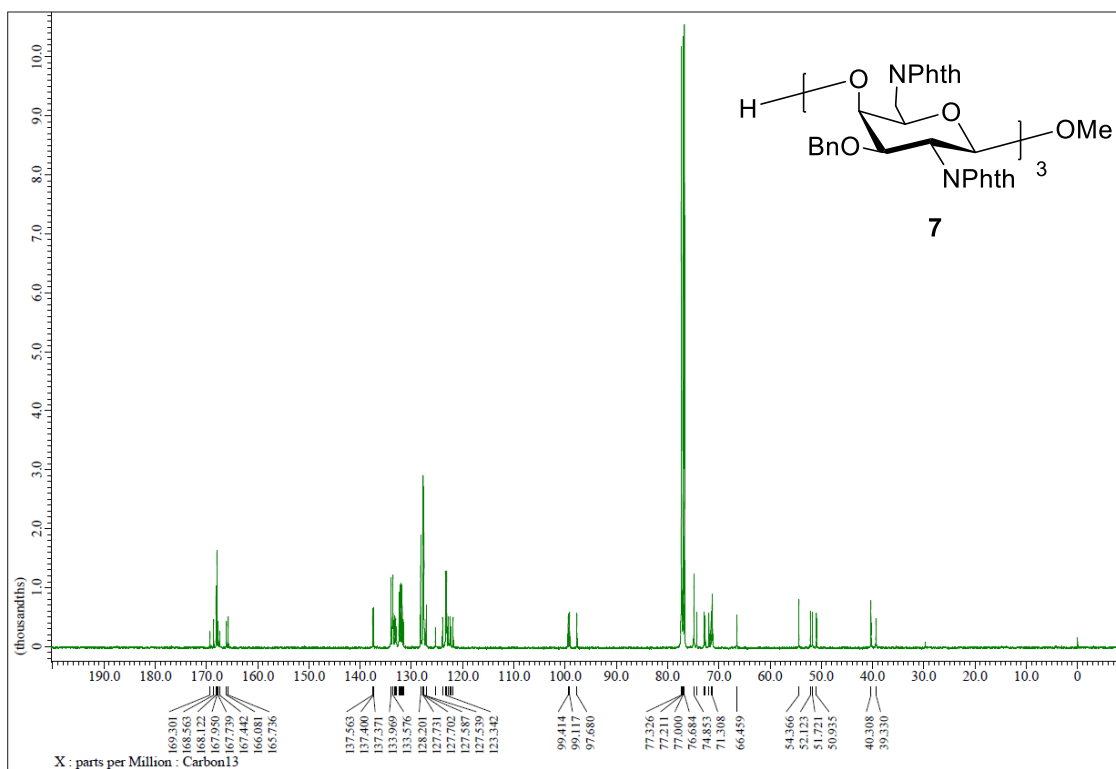
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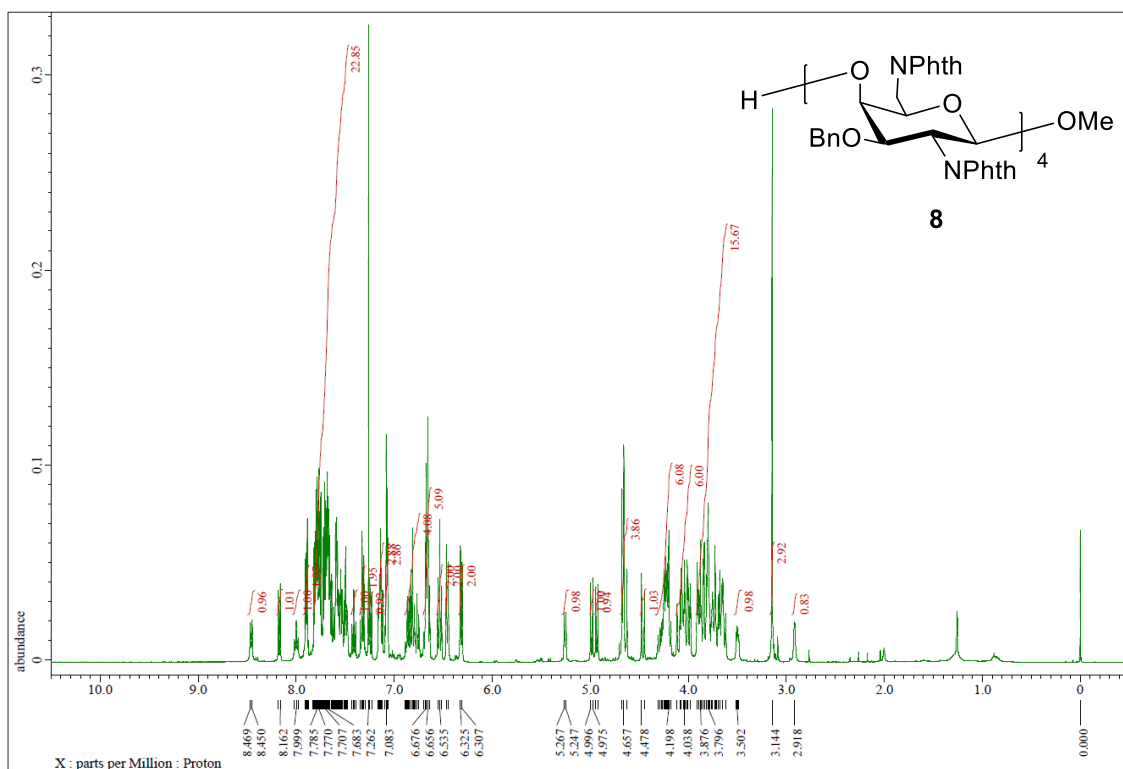
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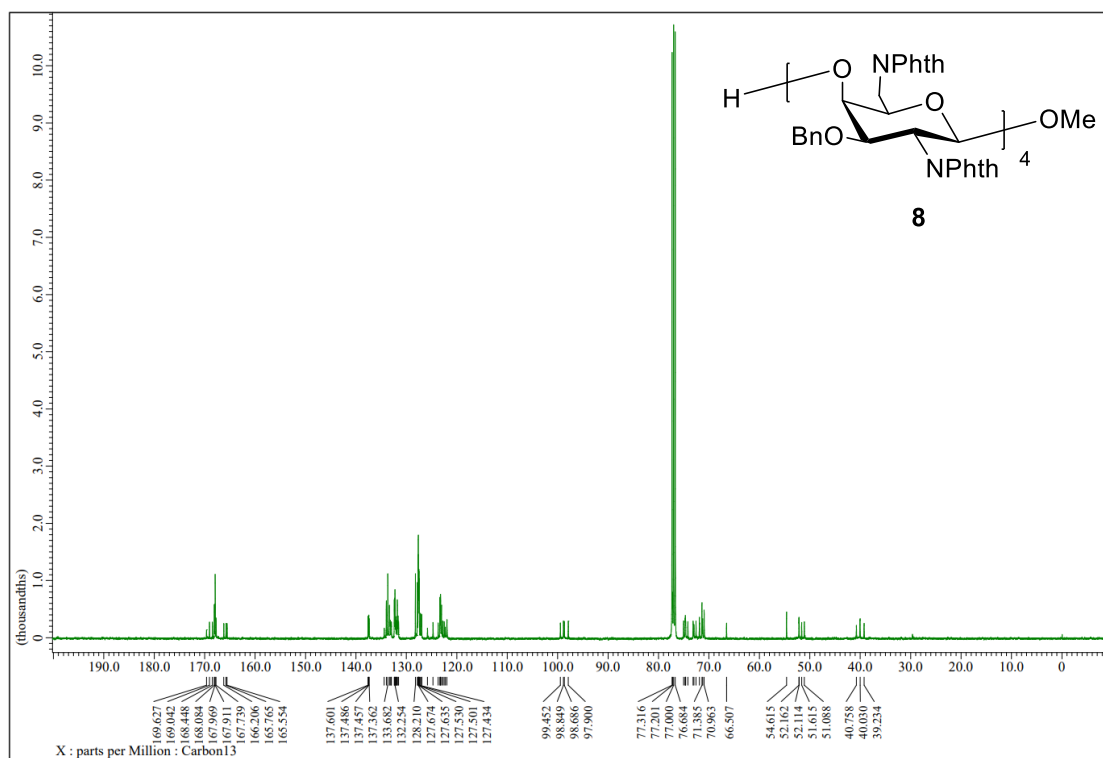
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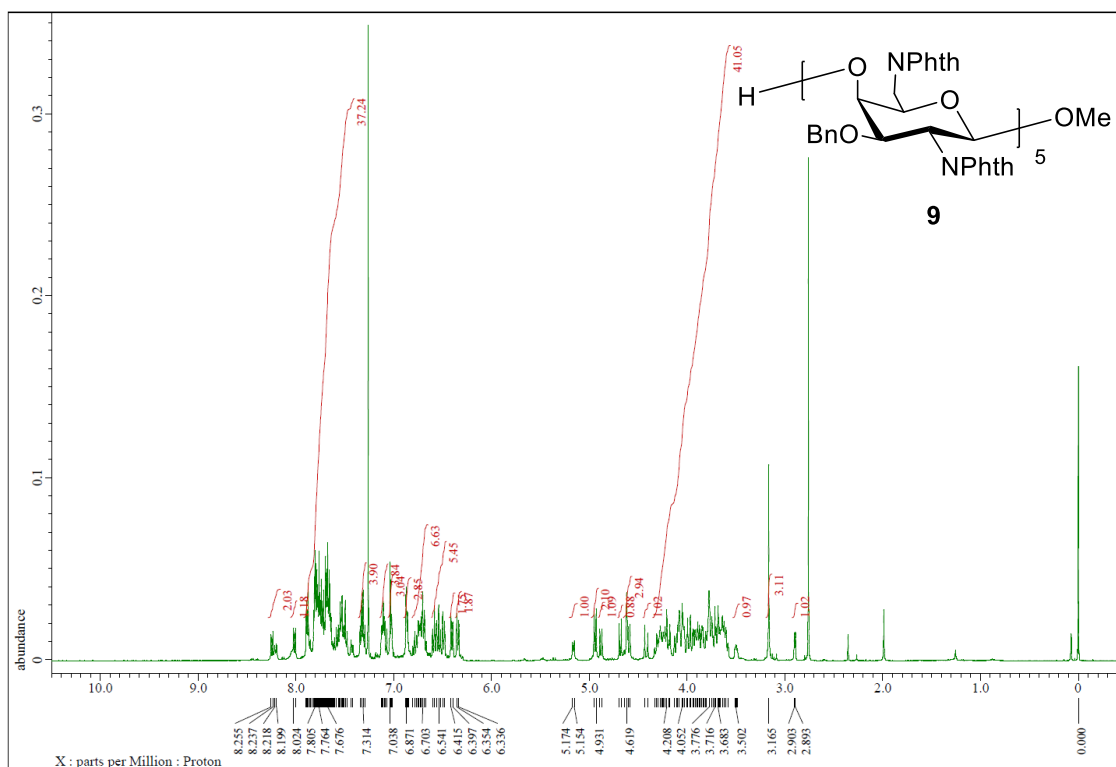


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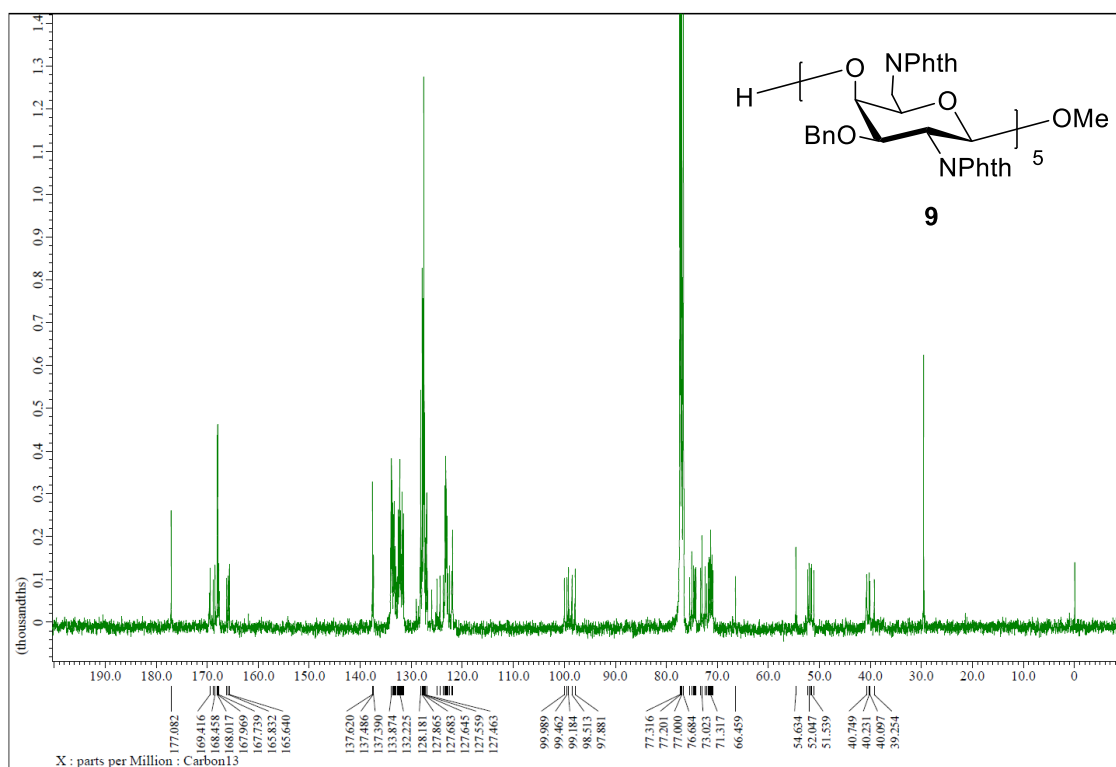




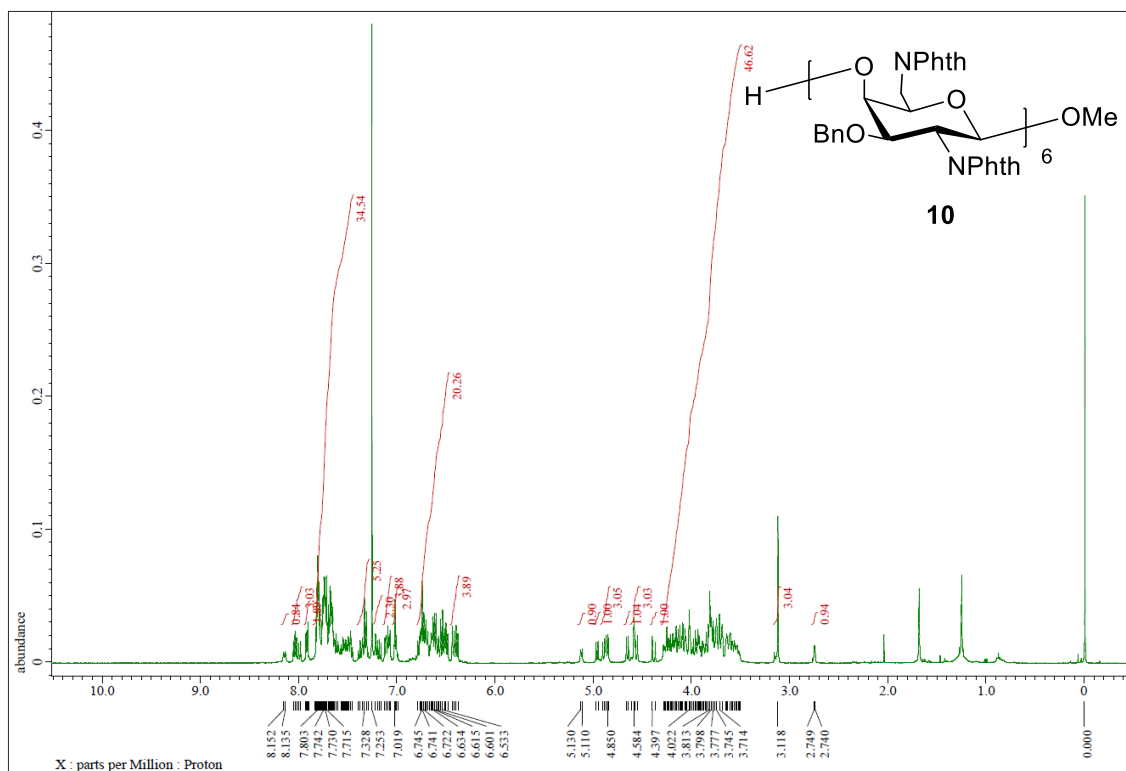
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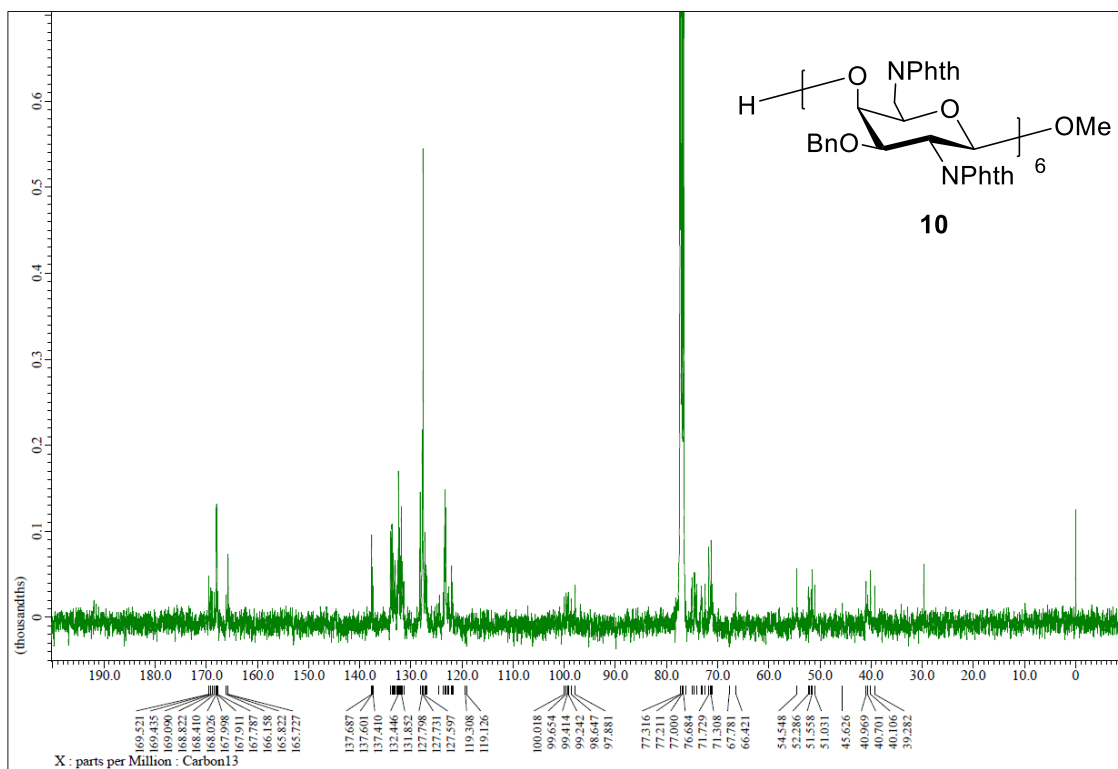
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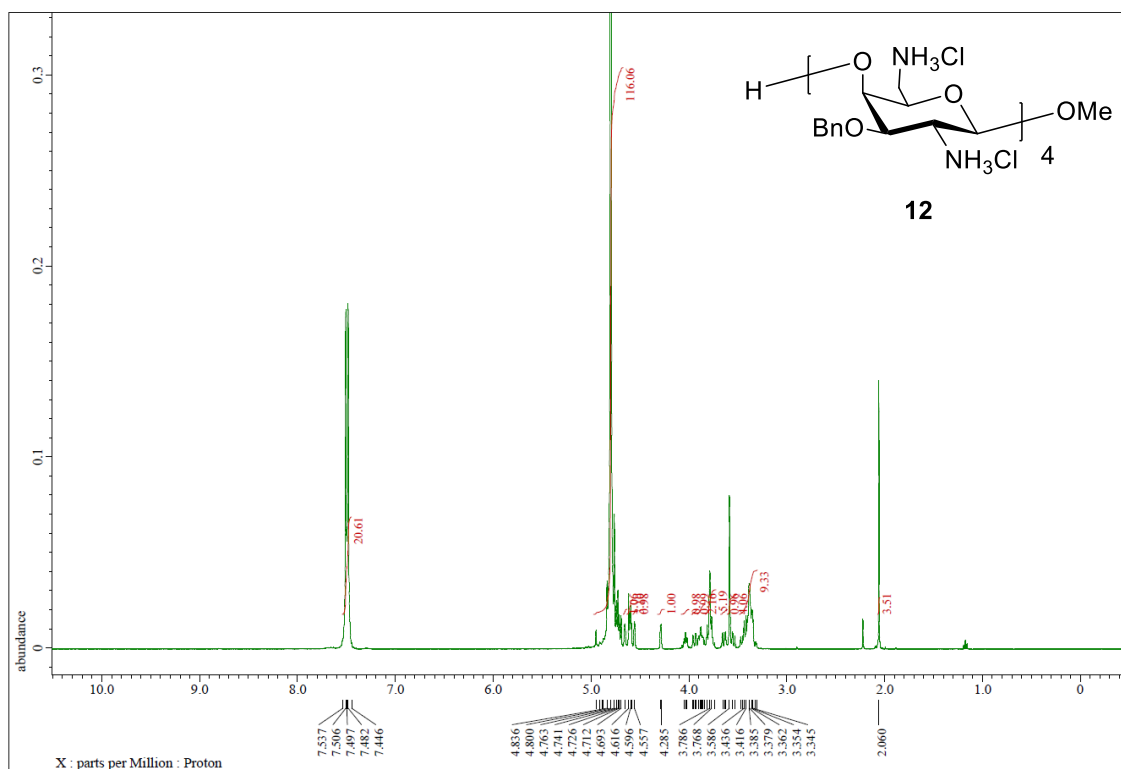
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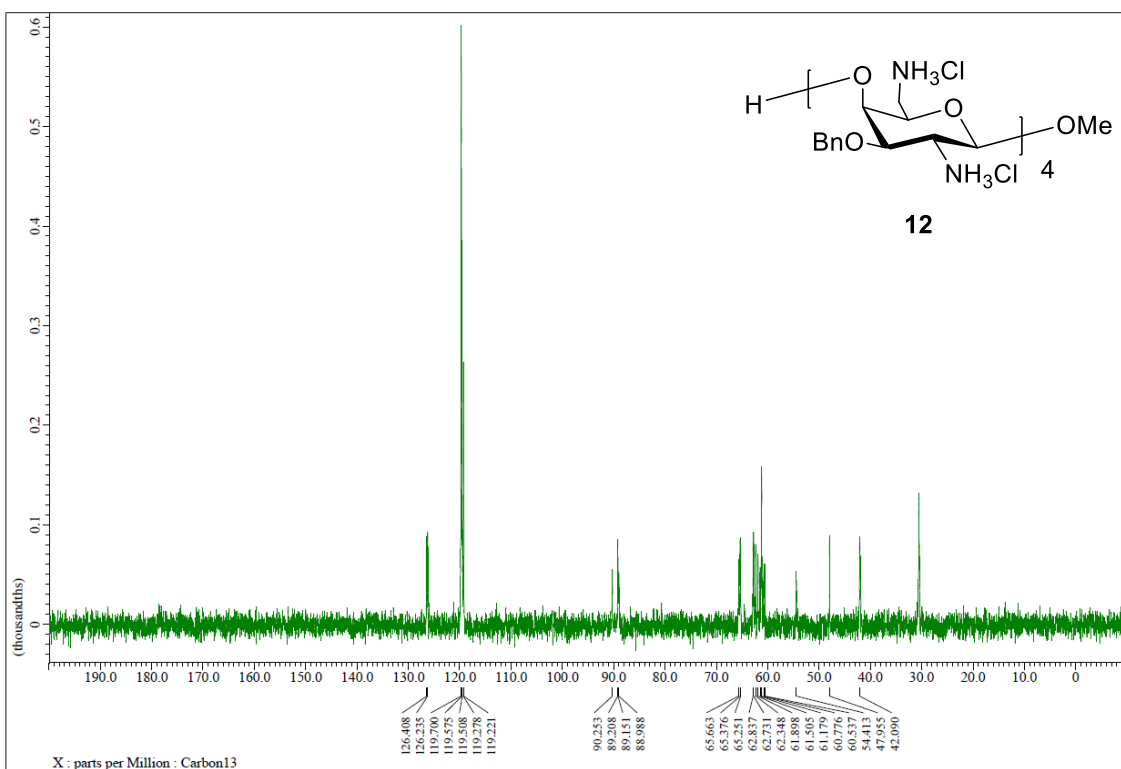
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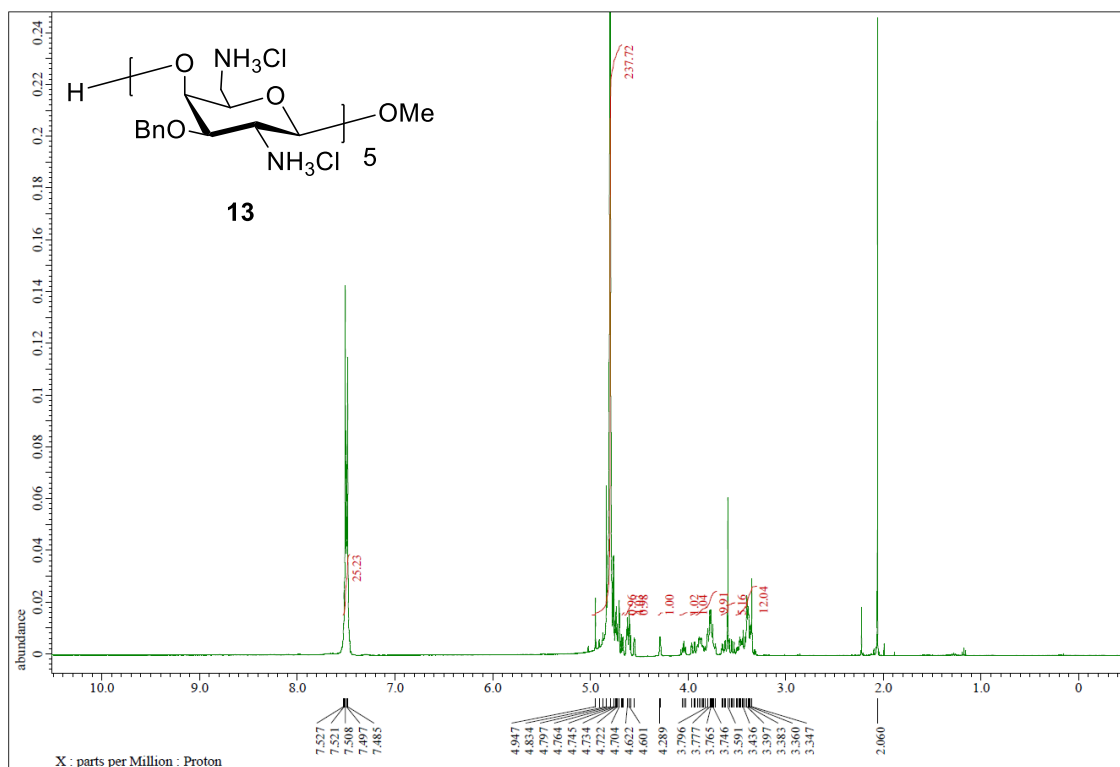
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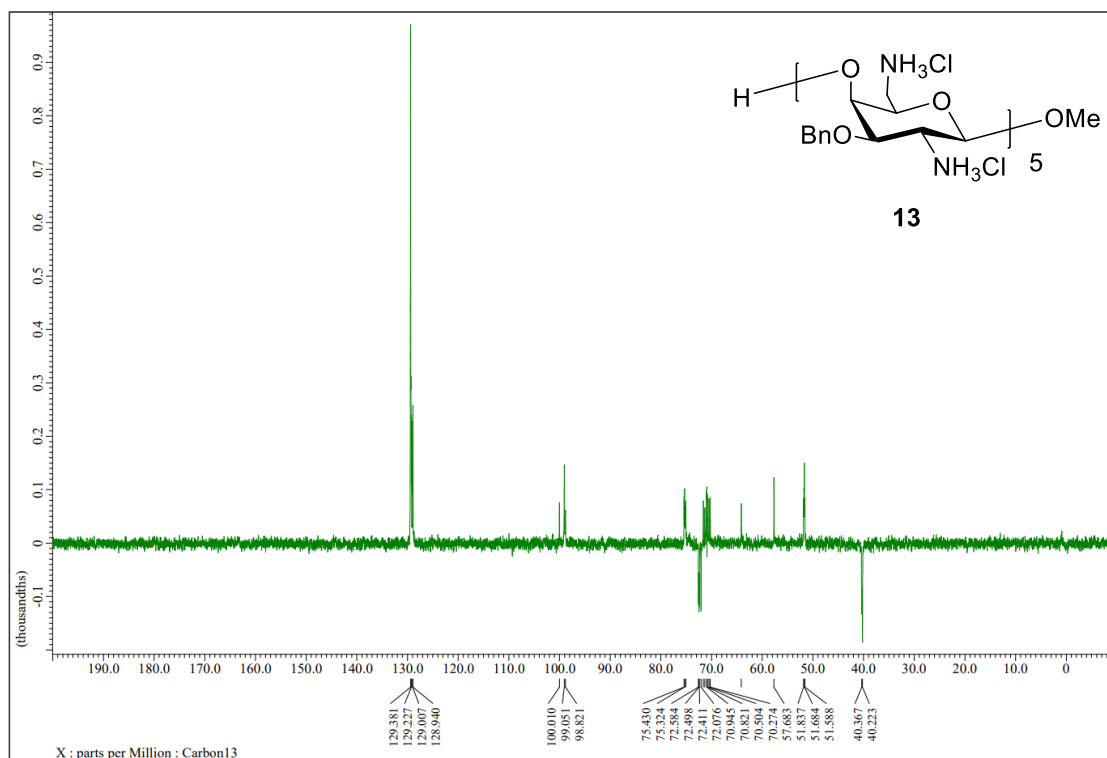
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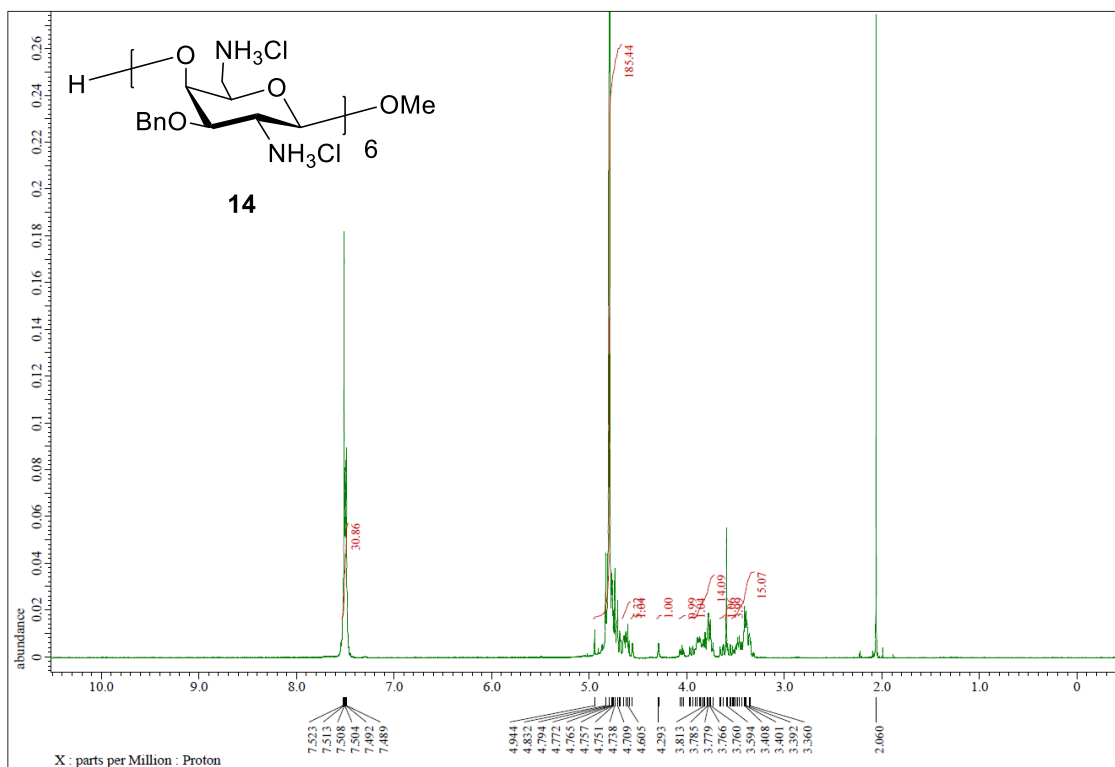
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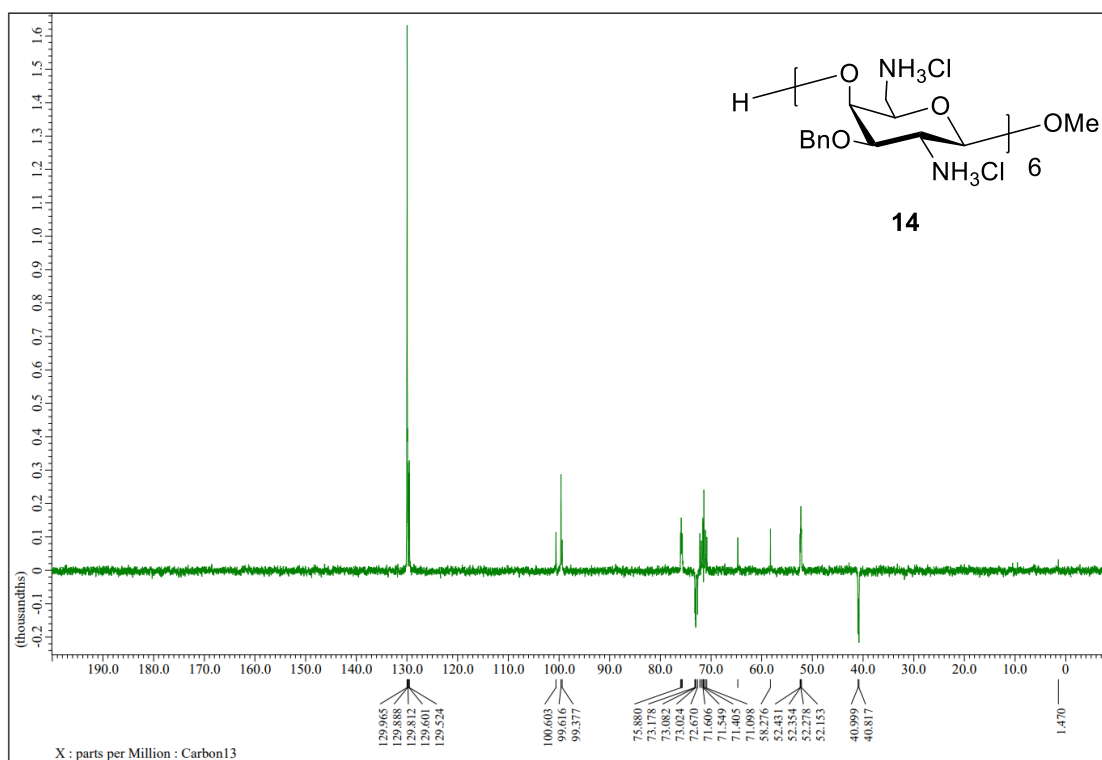
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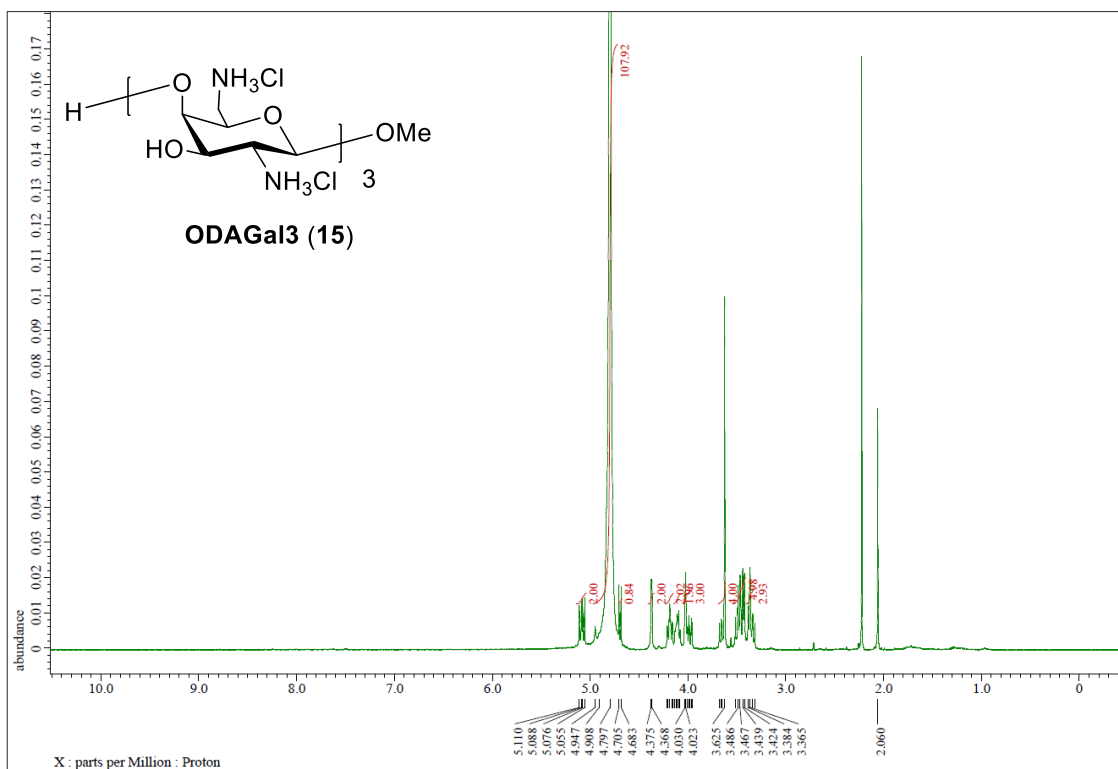
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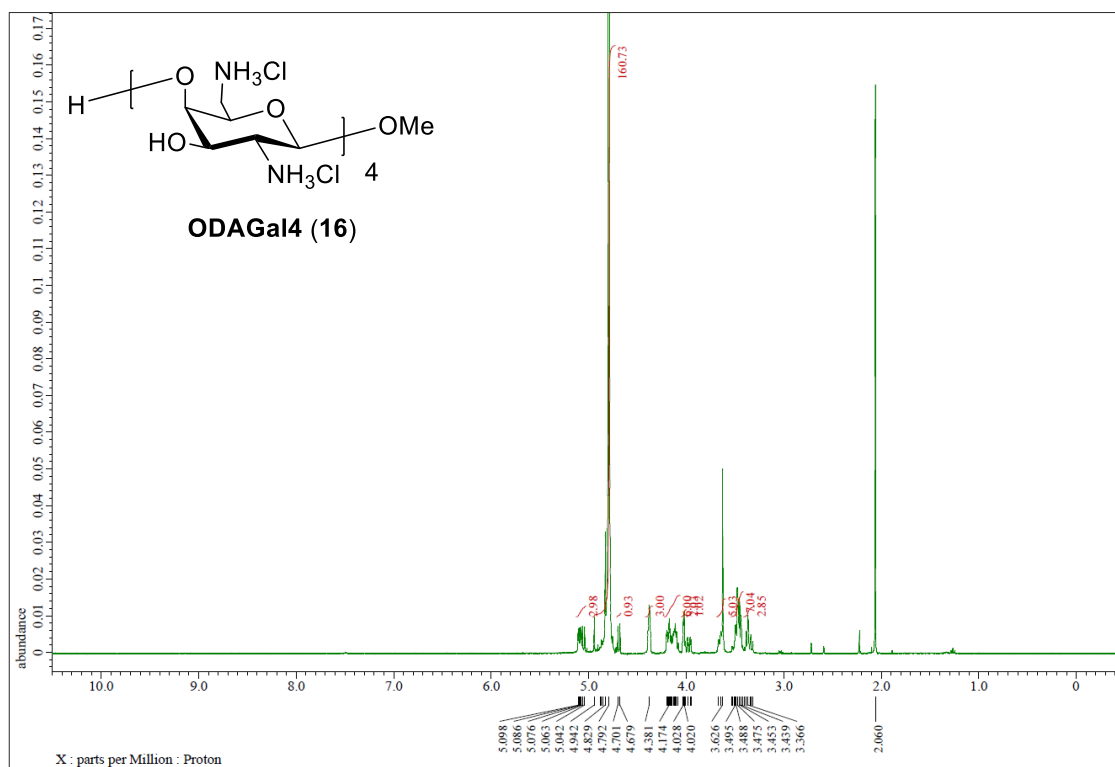
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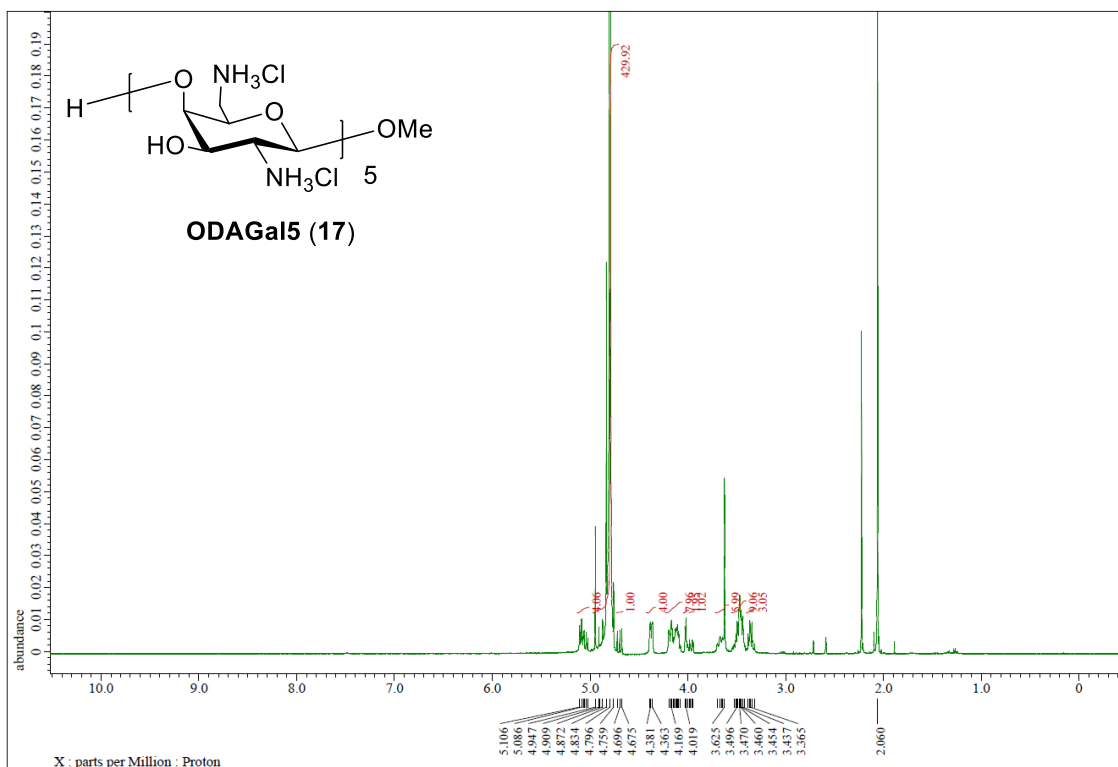
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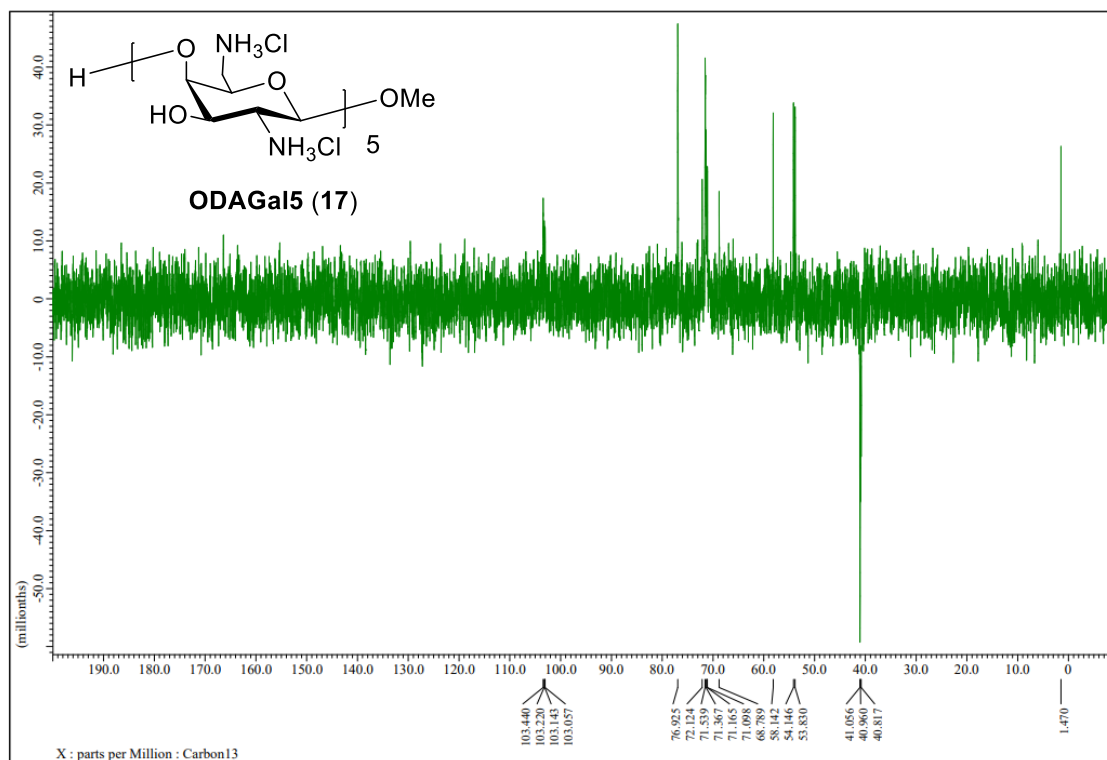
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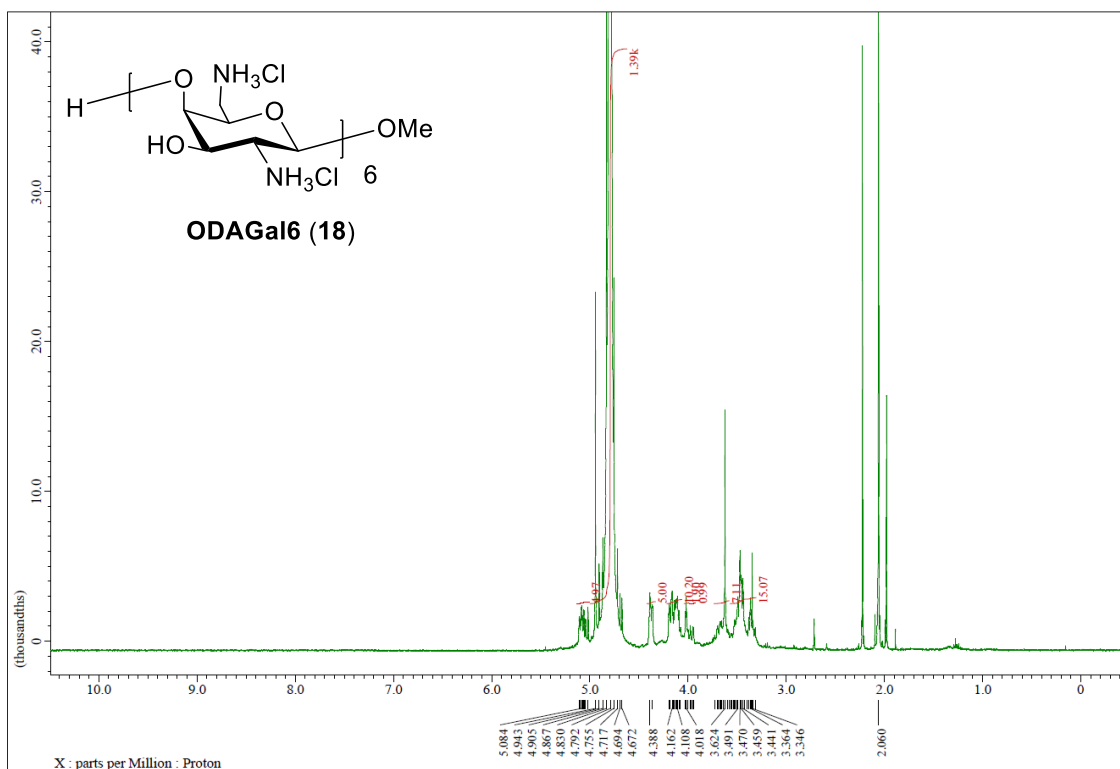
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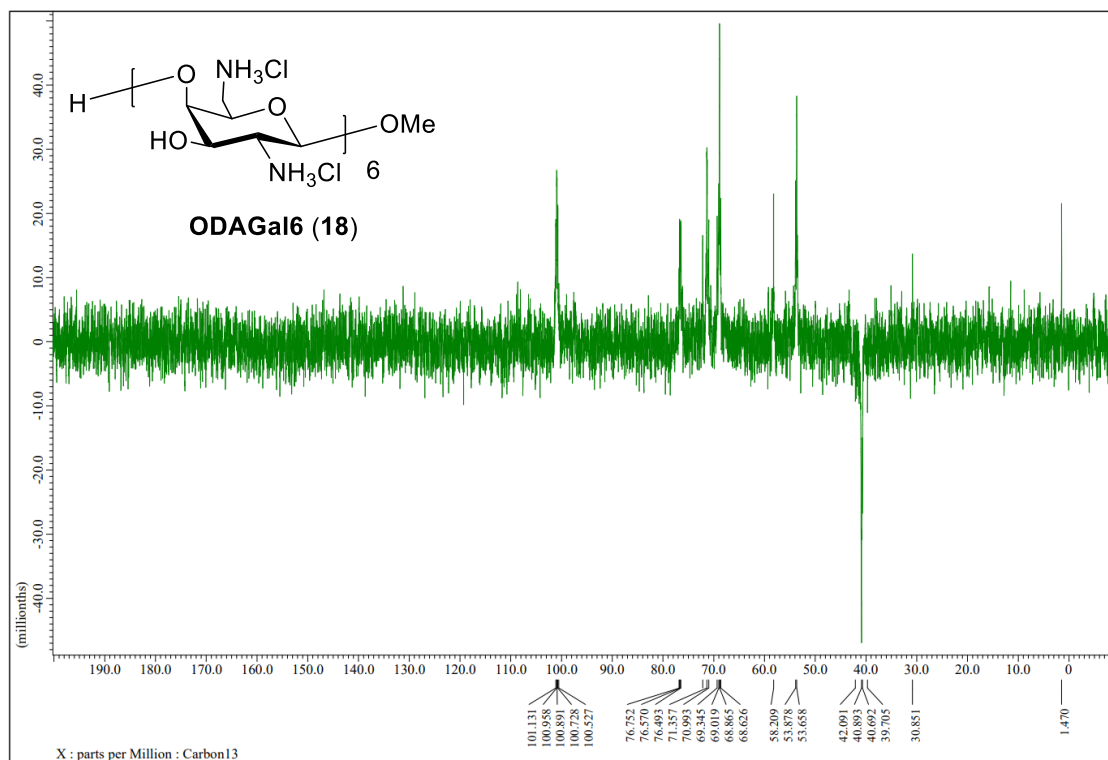
$^{13}\text{C}$   $\{^1\text{H}\}$  (DEPT 135) NMR (101 MHz,  $\text{D}_2\text{O}$ )



$^1\text{H}$  NMR (400 MHz,  $\text{D}_2\text{O}$ )



$^{13}\text{C}$   $\{^1\text{H}\}$  (DEPT 135) NMR (101 MHz,  $\text{D}_2\text{O}$ )





## **2. Information of sequences of nucleic acid oligomers**

### **RNA/RNA**

CAGURR	RNA	5'- CAGUCAGUCAGU	-3'
	cRNA	5'- ACUGACUGACUG	-3'
AP12RR	RNA	5'- GCAUUGGUAUUC	-3'
	cRNA	5'- GAAUACCAAUGC	-3'
AP16RR	RNA	5'- CAGCAUUGGUAUUCAG	-3'
	cRNA	5'- CUGAAUACCAAUGCUG	-3'
ML16RR	RNA	5'- CUAGUUCACUGAAUGC	-3'
	cRNA	5'- GCAUUCAGUGAACUAG	-3'
AP20RR	RNA	5'- GCCUCAGUCUGCUUCGCACC	-3'
	cRNA	5'- GGUGCGAAGCAGACUGAGGC	-3'
MP20RR	RNA	5'- UUCAGCAUUGGUAUUCAGUG	-3'
	cRNA	5'- CACUGAAUACCAAUGCUGAA	-3'

Capital letter: RNA.

### **DNA/RNA**

CAGTDR	DNA	5'- cagtcagtcagt	-3'
	cRNA	5'- ACUGACUGACUG	-3'
AP12DR	DNA	5'- gcattggtattc	-3'
	cRNA	5'- GAAUACCAAUGC	-3'
AP16DR	DNA	5'- cagcattggtattcag	-3'
	cRNA	5'- CUGAAUACCAAUGCUG	-3'
ML16DR	DNA	5'- ctagttcactgaatgc	-3'
	cRNA	5'- GCAUUCAGUGAACUAG	-3'
AP20DR	DNA	5'- ttcagcattggtattcagtg	-3'
	cRNA	5'- CACUGAAUACCAAUGCUGAA	-3'
MP20DR	DNA	5'- gcctcagtctgcttcgacc	-3'
	cRNA	5'- GGUGCGAAGCAGACUGAGGC	-3'

Small letter: DNA; capital letter: RNA.

**DNA/DNA**

AP16DD	DNA	5'- cagcattggattcag	-3'
	DNA	5'- ctgaataccaatgctg	-3'
MP20DD	DNA	5'- gcctcagtctgcttcgcacc	-3'
	DNA	5'- cgtgcgaagcagactgagcc	-3'

Small letter: DNA.

**gapmer/RNA and 2'-OMe RNA/RNA**

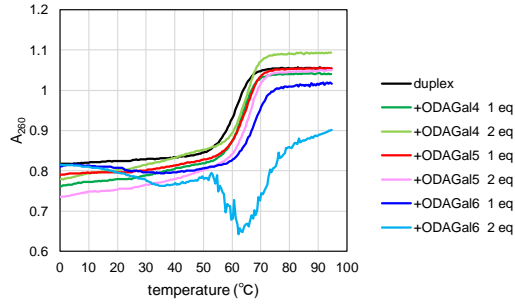
AP12G2	gapmer	5'- G <sup>M</sup> C <sup>M</sup> attggatU <sup>M</sup> C <sup>M</sup>	-3'
	cRNA	5'- GAAUACCAAUGC	-3'
AP12G4	gapmer	5'- G <sup>M</sup> C <sup>M</sup> A <sup>M</sup> U <sup>M</sup> tggtA <sup>M</sup> U <sup>M</sup> U <sup>M</sup> C <sup>M</sup>	-3'
	cRNA	5'- GAAUACCAAUGC	-3'
AP12MR	2'-OMe RNA	5'- G <sup>M</sup> C <sup>M</sup> A <sup>M</sup> U <sup>M</sup> U <sup>M</sup> G <sup>M</sup> G <sup>M</sup> U <sup>M</sup> A <sup>M</sup> U <sup>M</sup> U <sup>M</sup> C <sup>M</sup>	-3'
	cRNA	5'- GAAUACCAAUGC	-3'
AP16G2	gapmer	5'- C <sup>M</sup> A <sup>M</sup> gcattggatucA <sup>M</sup> G <sup>M</sup>	-3'
	cRNA	5'- CUGAAUACCAAUGCUG	-3'
ML16G2	gapmer	5'- C <sup>M</sup> U <sup>M</sup> agttcactgaatG <sup>M</sup> C <sup>M</sup>	-3'
	cRNA	5'- GCAUUCAGUGAACUAG	-3'
AP16G4	gapmer	5'- C <sup>M</sup> A <sup>M</sup> G <sup>M</sup> C <sup>M</sup> attggatU <sup>M</sup> C <sup>M</sup> A <sup>M</sup> G <sup>M</sup>	-3'
	cRNA	5'- CUGAAUACCAAUGCUG	-3'
ML16G4	gapmer	5'- C <sup>M</sup> U <sup>M</sup> A <sup>M</sup> G <sup>M</sup> ttcactgaA <sup>M</sup> U <sup>M</sup> G <sup>M</sup> C <sup>M</sup>	-3'
	cRNA	5'- GCAUUCAGUGAACUAG	-3'
AP16MR	2'-OMe RNA	5'- C <sup>M</sup> A <sup>M</sup> G <sup>M</sup> C <sup>M</sup> A <sup>M</sup> T <sup>M</sup> T <sup>M</sup> G <sup>M</sup> G <sup>M</sup> T <sup>M</sup> A <sup>M</sup> T <sup>M</sup> U <sup>M</sup> C <sup>M</sup> A <sup>M</sup> G <sup>M</sup>	-3'
	cRNA	5'- CUGAAUACCAAUGCUG	-3'
ML16MR	2'-OMe RNA	5'- C <sup>M</sup> U <sup>M</sup> A <sup>M</sup> G <sup>M</sup> U <sup>M</sup> U <sup>M</sup> C <sup>M</sup> A <sup>M</sup> C <sup>M</sup> U <sup>M</sup> G <sup>M</sup> A <sup>M</sup> A <sup>M</sup> U <sup>M</sup> G <sup>M</sup> C <sup>M</sup>	-3'
	cRNA	5'- GCAUUCAGUGAACUAG	-3'

Small letter: DNA; capital letter: RNA; X<sup>M</sup>; 2'-OMe RNA.

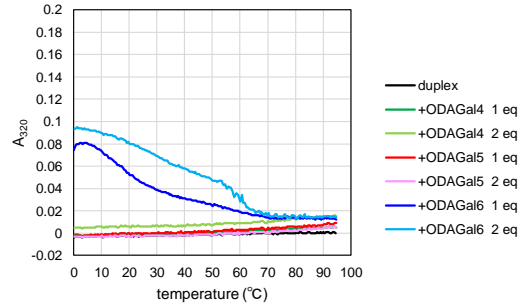
### 3. Melting curves of RNA/RNA (Table 1)

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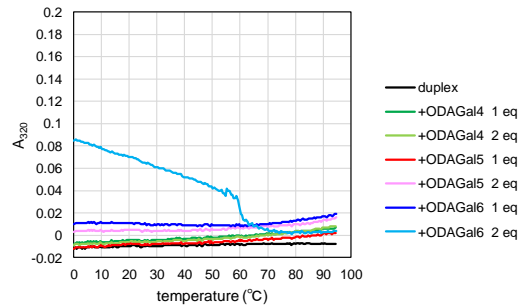
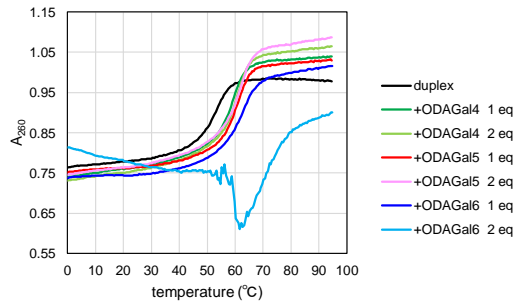
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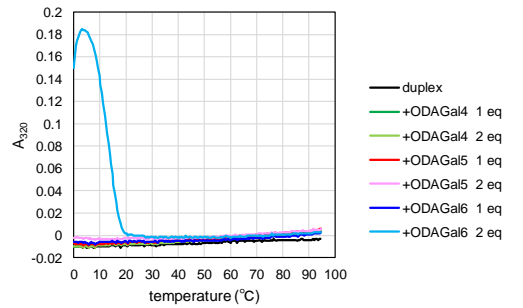
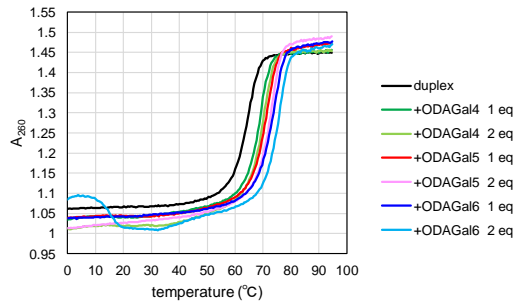
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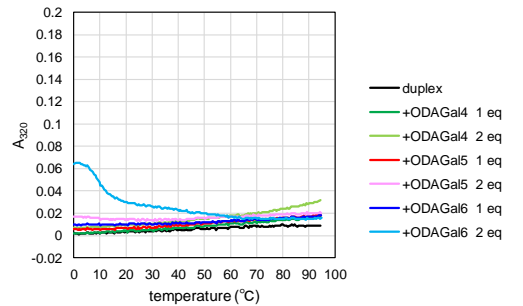
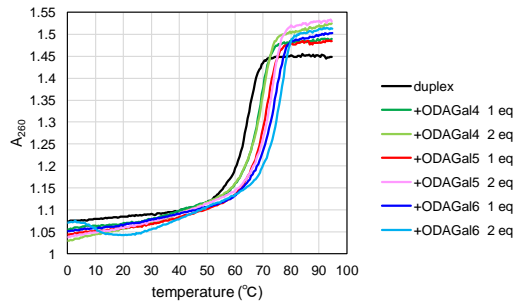
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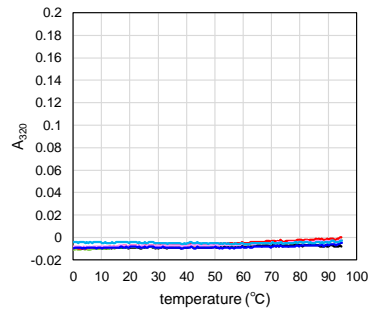
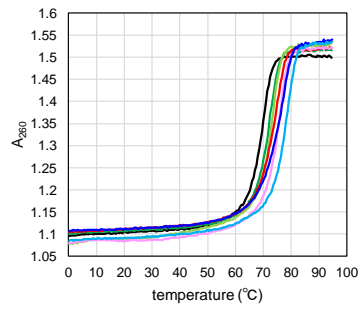
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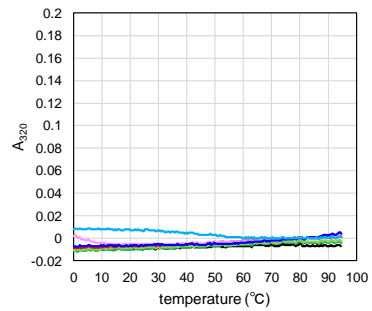
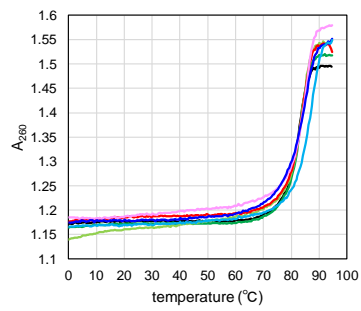
ML16RR



**AP20RR**



**MP20RR**

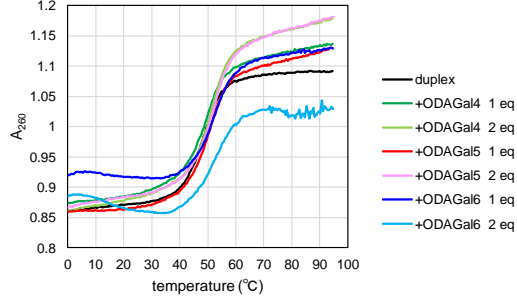


**Figure S1** UV melting curves of RNA/RNA in the absence and presence of ODAGals. (A) at 260 nm (B) at 320 nm.

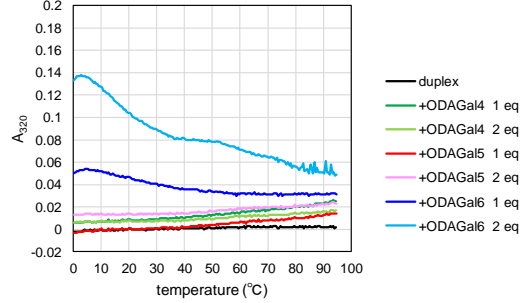
## 4. Melting curves of DNA/RNA (Table 2)

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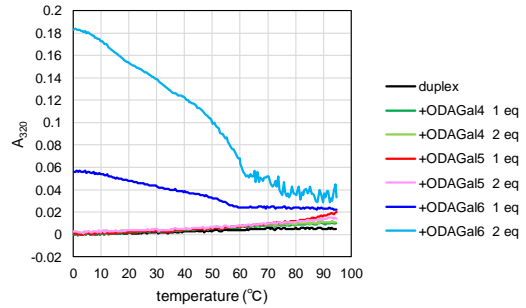
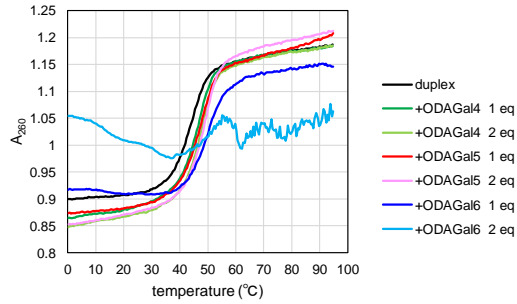
CAGTDR



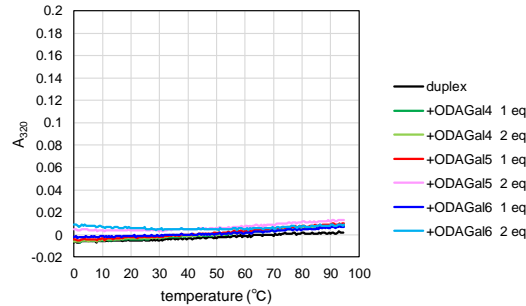
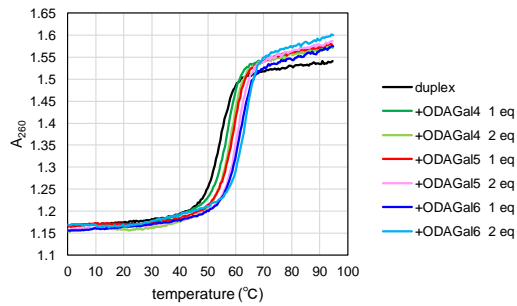
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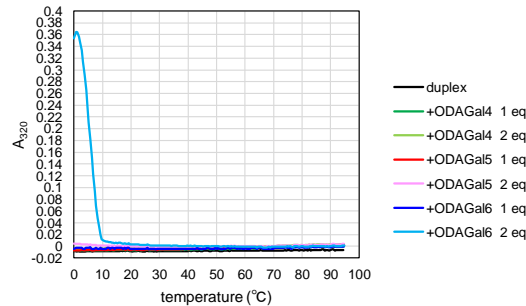
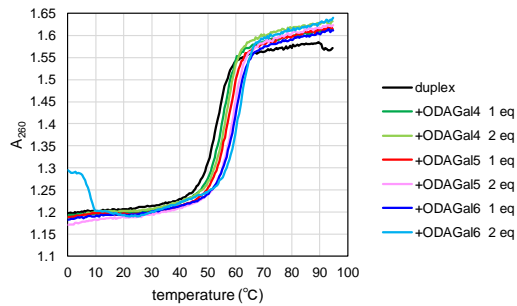
AP12DR

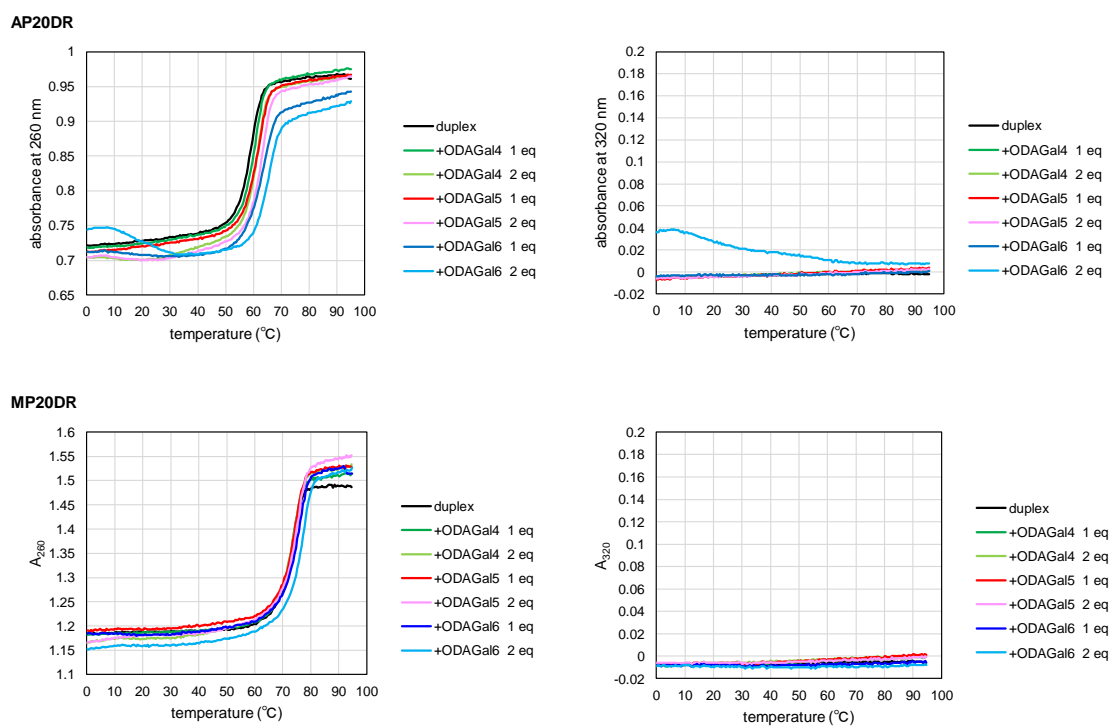


AP16DR



ML16DR



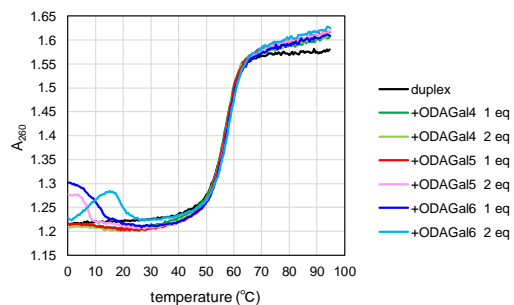


**Figure S2** UV melting curves of DNA/RNA in the absence and presence of ODAGals. (A) at 260 nm (B) at 320 nm.

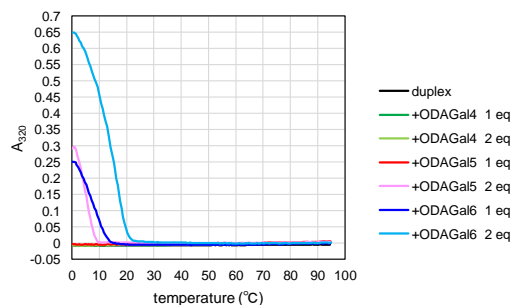
## 5. Melting curves of DNA/DNA (Table 3)

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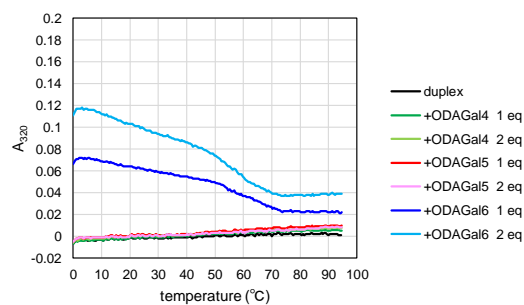
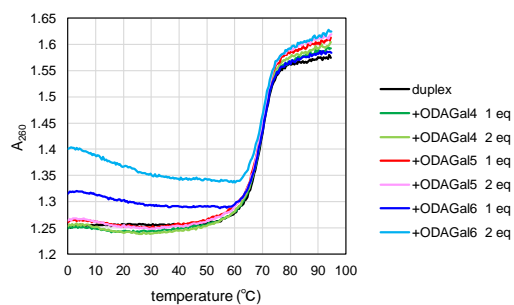
AP16DD



B



MP20DD

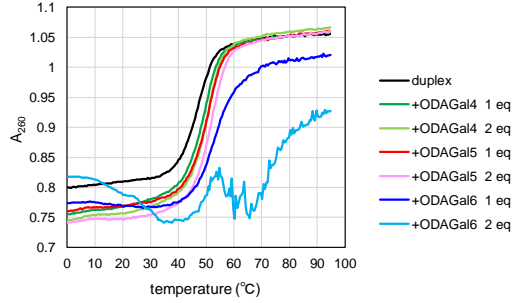


**Figure S3** UV melting curves of DNA/DNA in the absence and presence of ODAGals. (A) at 260 nm (B) at 320 nm.

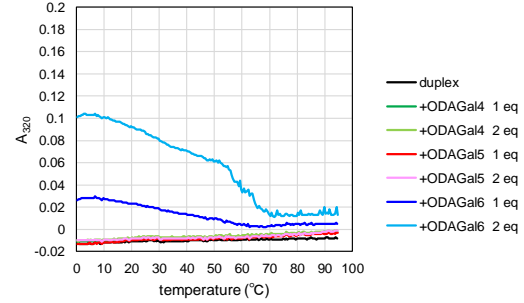
## 6. Melting curves of gapmer/ RNA and 2'-OMe RNA/RNA (Table 4)

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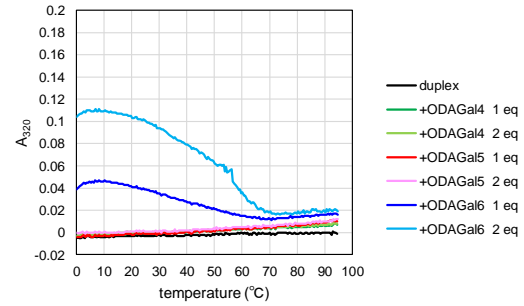
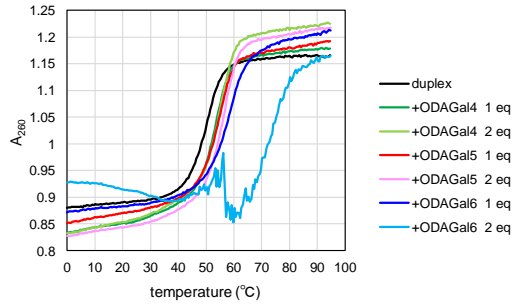
AP12G2



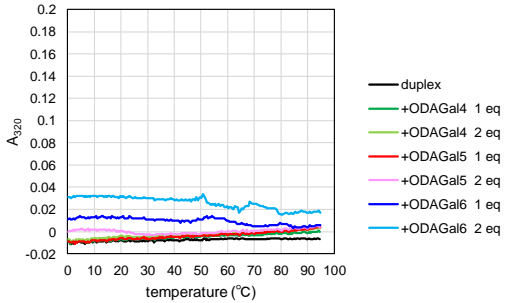
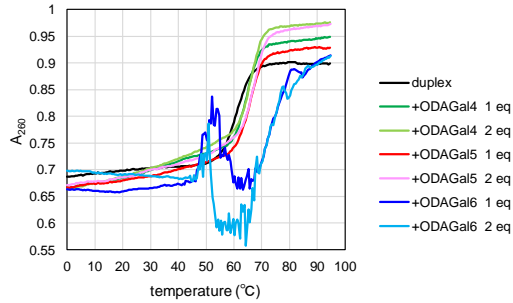
B



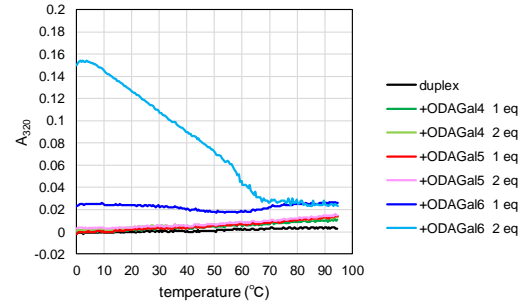
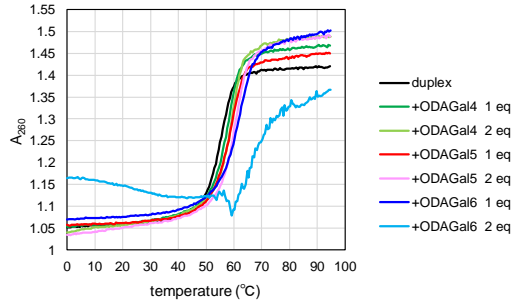
AP12G4



AP12MR

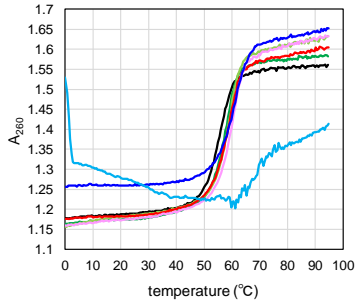


AP16G2

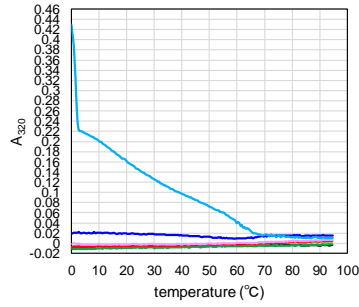




**ML16G2**

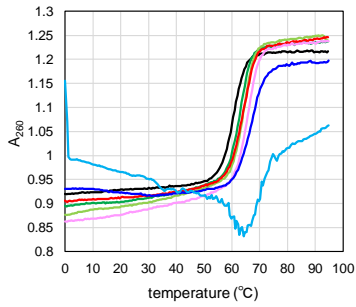


- duplex
- +ODAGal4 1 eq
- +ODAGal4 2 eq
- +ODAGal5 1 eq
- +ODAGal5 2 eq
- +ODAGal6 1 eq
- +ODAGal6 2 eq

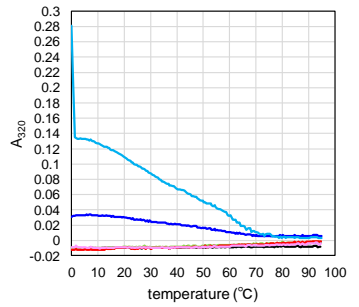


- duplex
- +ODAGal4 1 eq
- +ODAGal4 2 eq
- +ODAGal5 1 eq
- +ODAGal5 2 eq
- +ODAGal6 1 eq
- +ODAGal6 2 eq

**AP16G4**

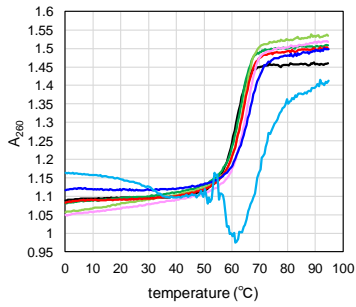


- duplex
- +ODAGal4 1 eq
- +ODAGal4 2 eq
- +ODAGal5 1 eq
- +ODAGal5 2 eq
- +ODAGal6 1 eq
- +ODAGal6 2 eq

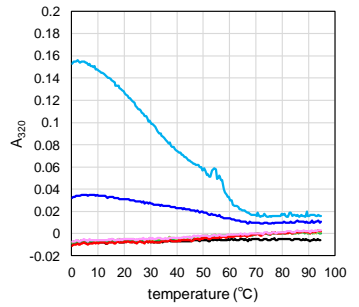


- duplex
- +ODAGal4 1 eq
- +ODAGal4 2 eq
- +ODAGal5 1 eq
- +ODAGal5 2 eq
- +ODAGal6 1 eq
- +ODAGal6 2 eq

**ML16G4**

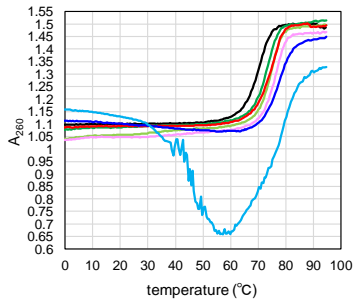


- duplex
- +ODAGal4 1 eq
- +ODAGal4 2 eq
- +ODAGal5 1 eq
- +ODAGal5 2 eq
- +ODAGal6 1 eq
- +ODAGal6 2 eq

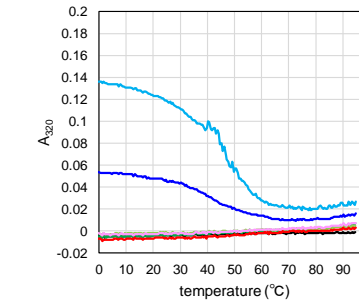


- duplex
- +ODAGal4 1 eq
- +ODAGal4 2 eq
- +ODAGal5 1 eq
- +ODAGal5 2 eq
- +ODAGal6 1 eq
- +ODAGal6 2 eq

**AP16MR**

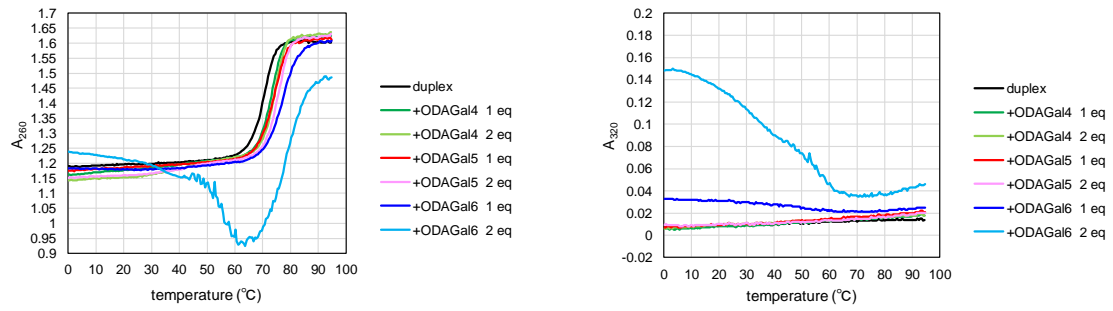


- duplex
- +ODAGal4 1 eq
- +ODAGal4 2 eq
- +ODAGal5 1 eq
- +ODAGal5 2 eq
- +ODAGal6 1 eq
- +ODAGal6 2 eq



- duplex
- +ODAGal4 1 eq
- +ODAGal4 2 eq
- +ODAGal5 1 eq
- +ODAGal5 2 eq
- +ODAGal6 1 eq
- +ODAGal6 2 eq

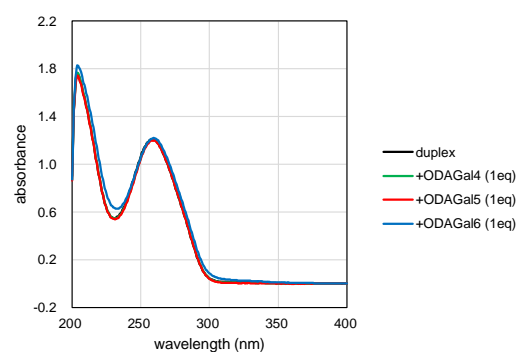
ML16MR



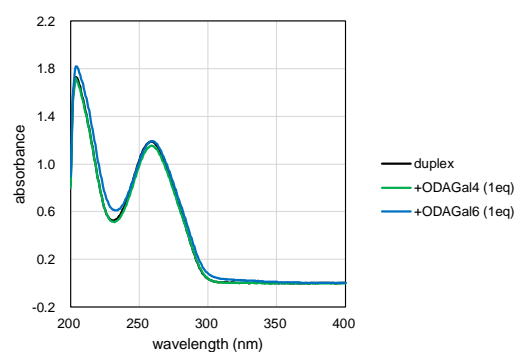
**Figure S4** UV melting curves of gapmer/RNA and 2'-OMe RNA/RNA in the absence and presence of ODAGals. (A) at 260 nm (B) at 320 nm.

## 7. UV spectra of oligomers

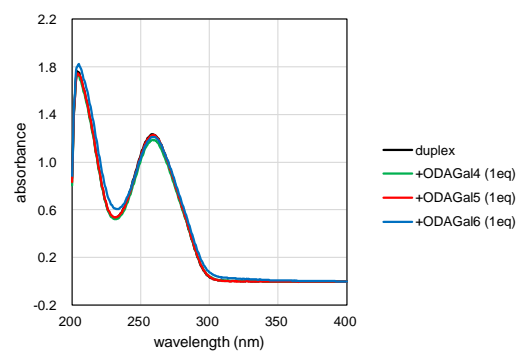
DNA/RNA (AP16DR)



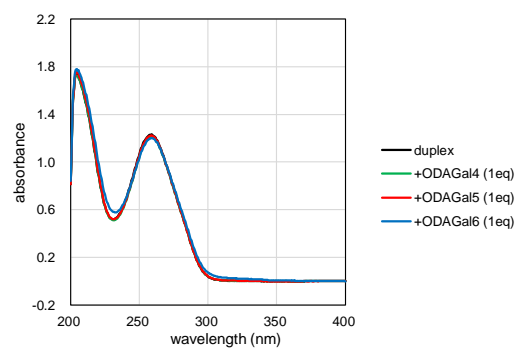
gapmer/RNA (2-12-2) (AP16G2)



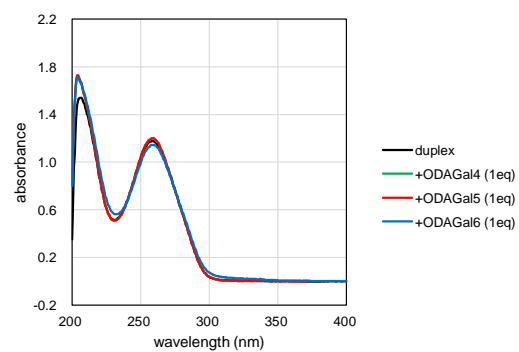
gapmer/RNA (4-8-4) (AP16G4)



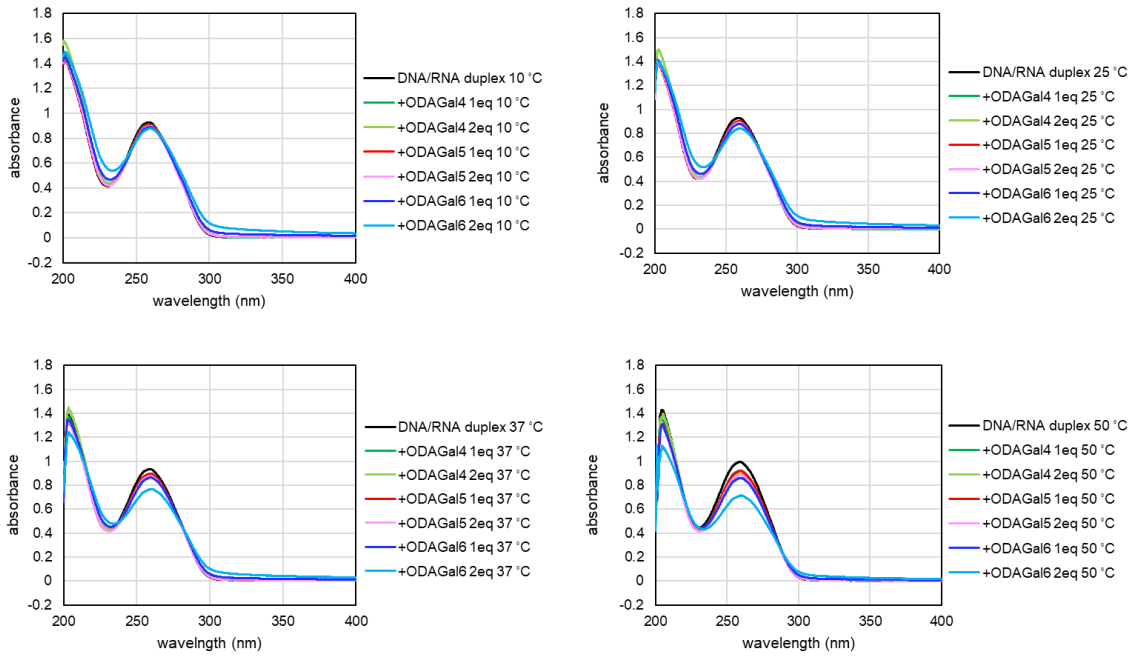
2'-OMe RNA/RNA (AP16MR)



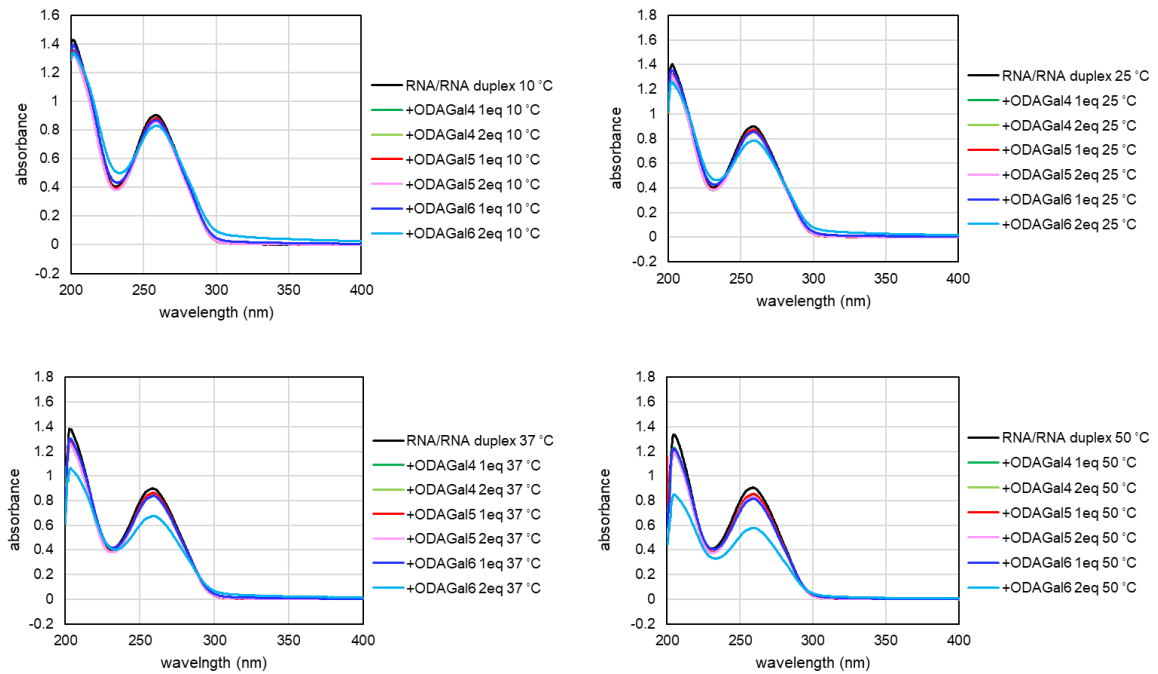
RNA/RNA (AP16RR)



**Figure S5** UV spectra of 5  $\mu$ M duplexes in phosphate buffer at room temperature ( $24 \pm 1$  °C).

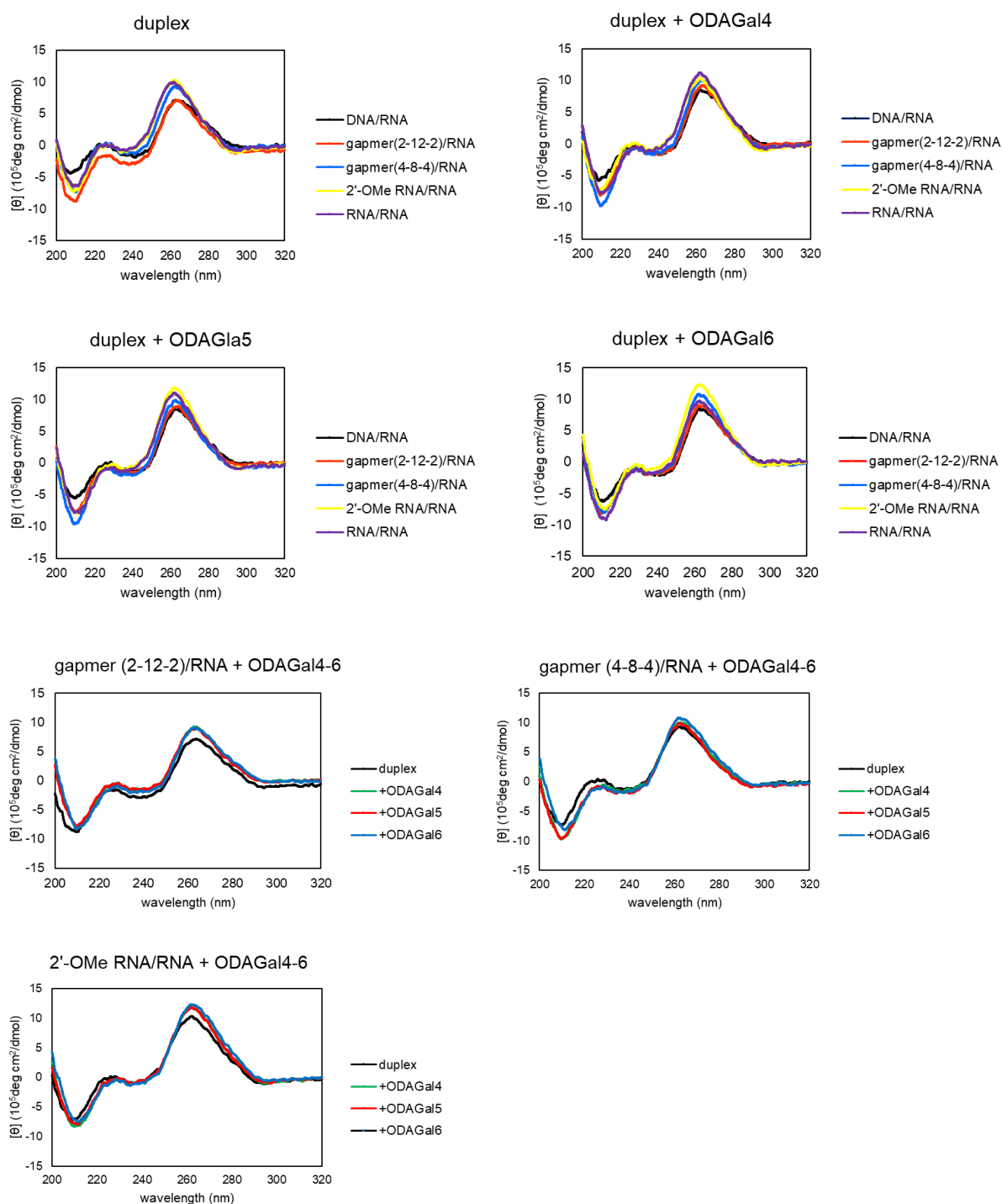


**Figure S6** UV spectra of 4  $\mu$ M DNA/RNA (AP16DR) in phosphate buffer.

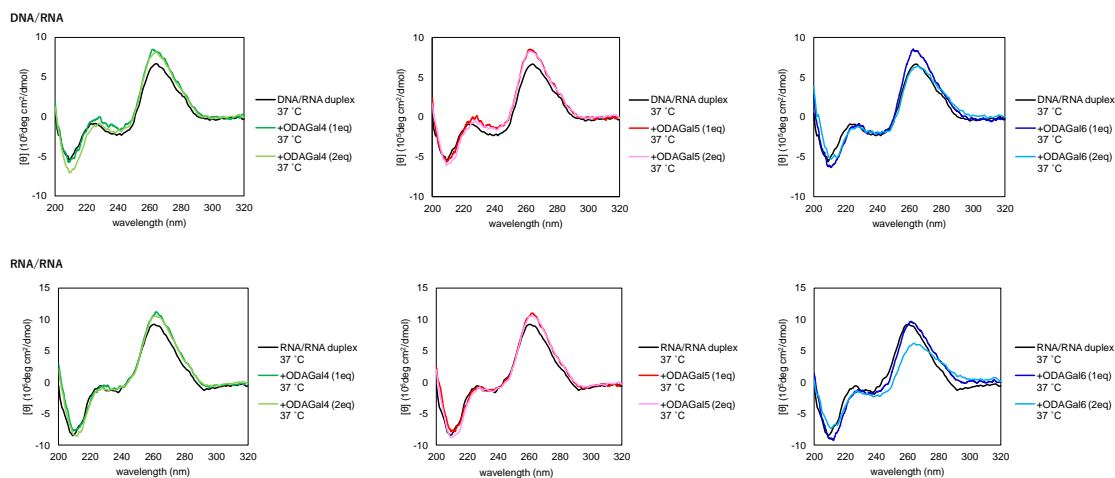


**Figure S7** UV spectra of 4  $\mu$ M RNA/RNA (AP16RR) in phosphate buffer.

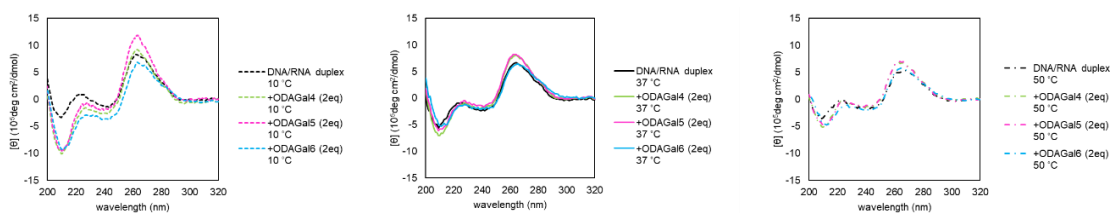
## 8. CD spectra of oligomers



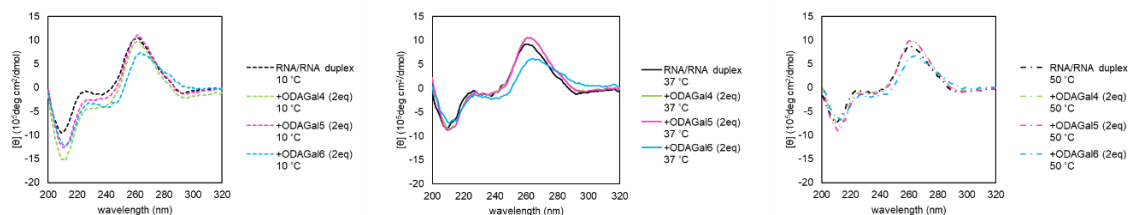
**Figure S8** CD spectra of duplexes in the presence and the absence of 1 equivalent of ODAGals at 37 °C. DNA/RNA: AP16DR; gapmer (2-12-2)/RNA: AP16G2; gapmer (4-8-4)/RNA: AP16G4; 2'-OMe RNA/RNA: AP16MR; RNA/RNA: AP16RR.



**Figure S9** CD spectra of DNA/RNA (AP16DR) and RNA/RNA (AP16RR) in the presence and the absence of 1 or 2 equivalents of ODAGals at 37 °C.

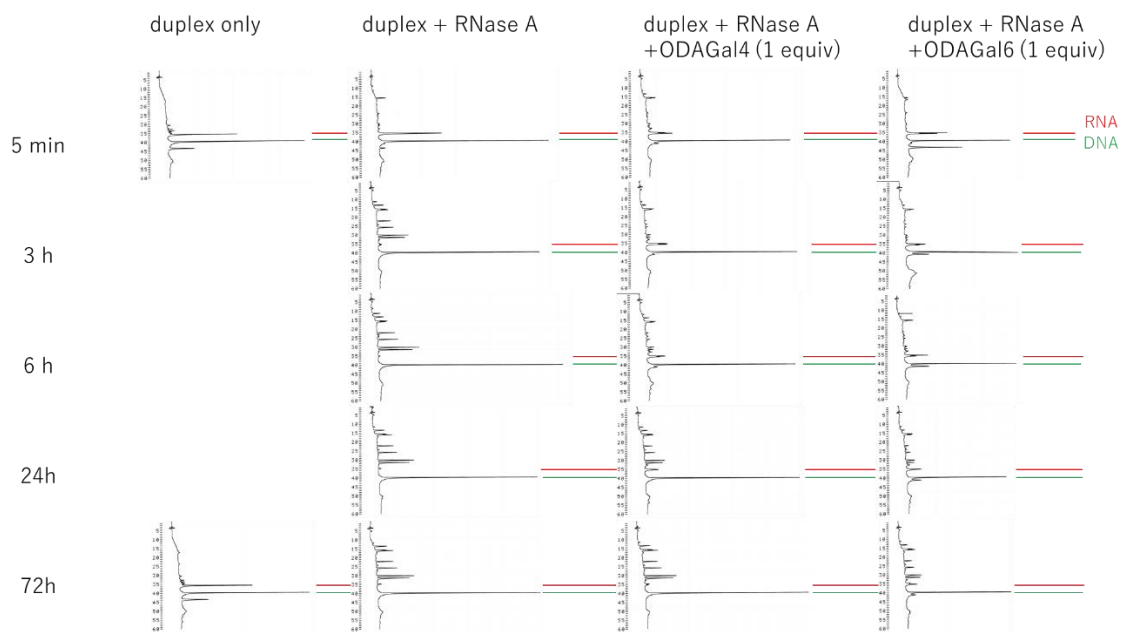


**Figure S10** CD spectra of DNA/RNA (AP16DR) in the presence and the absence of 2 equivalents of ODAGals.

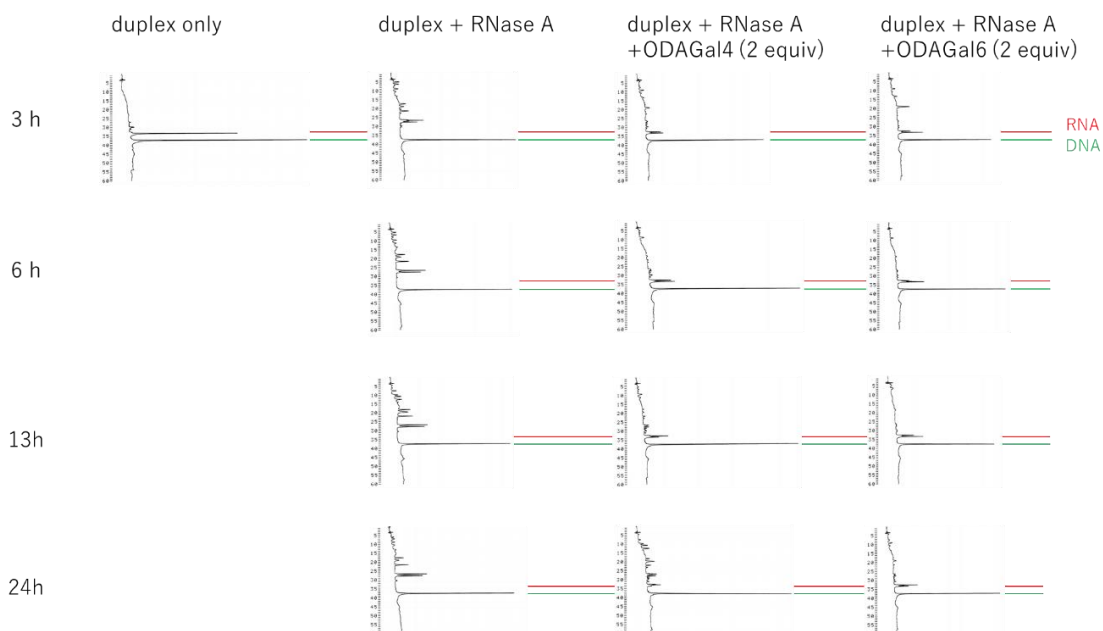


**Figure S11** CD spectra of RNA/RNA (AP16RR) in the presence and the absence of 2 equivalents of ODAGals.

## 9. RP-HPLC profiles of RNase A resistance experiment

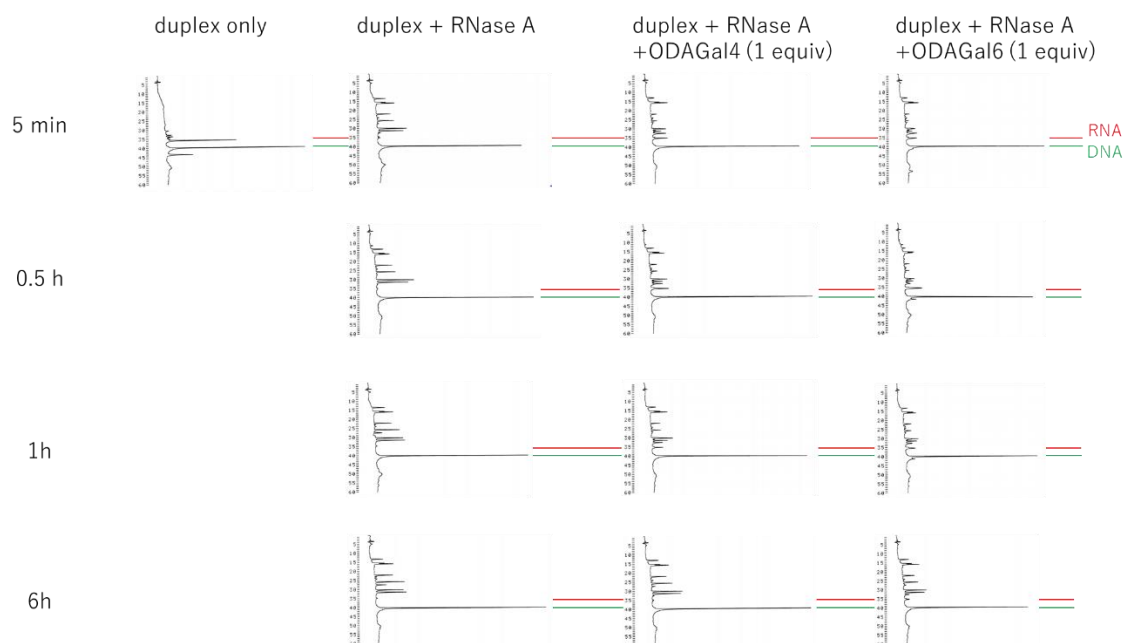


**Figure S12** RP-HPLC profiles of the mixture after treatment of 0.5 µg/mL RNase A. RP-HPLC analyses (UV detection at 260 nm) were performed using a linear gradient of 0%–20% CH<sub>3</sub>CN in 0.1 M triethyl ammonium acetate (TEAA) buffer (pH 7.0) at 30 °C for 60 min with a flow rate of 0.5 mL/min using a C18 column.

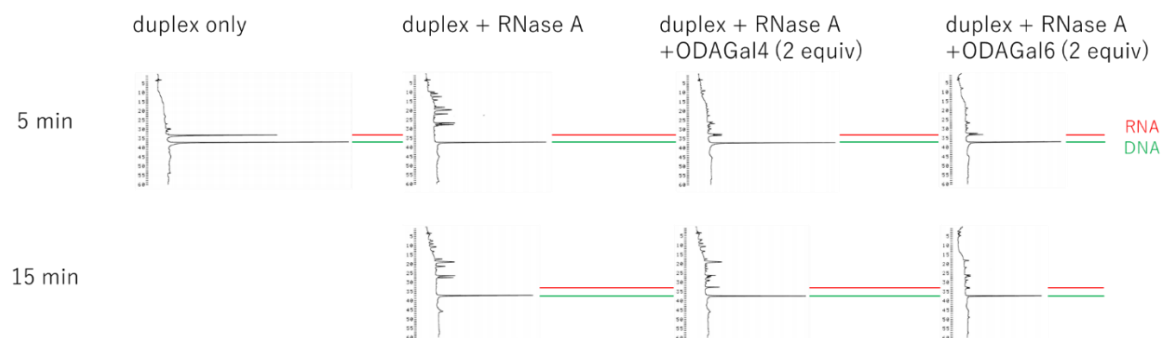


**Figure S13** RP-HPLC profiles of the mixture after treatment of 0.5 µg/mL RNase A. RP-HPLC analyses (UV detection at 260 nm) were performed using a linear gradient of 0%–20% CH<sub>3</sub>CN in

0.1 M TEAA buffer (pH 7.0) at 50 °C for 60 min with a flow rate of 0.5 mL/min using a C18 column.



**Figure S14** RP-HPLC profiles of the mixture after treatment of 10  $\mu\text{g/mL}$  RNase A. RP-HPLC analyses (UV detection at 260 nm) were performed using a linear gradient of 0%–20%  $\text{CH}_3\text{CN}$  in 0.1 M TEAA buffer (pH 7.0) at 30 °C for 60 min with a flow rate of 0.5 mL/min using a C18 column.



**Figure S15** RP-HPLC profiles of the mixture after treatment of 10  $\mu\text{g/mL}$  RNase A. RP-HPLC analyses (UV detection at 260 nm) were performed using a linear gradient of 0%–20%  $\text{CH}_3\text{CN}$  in 0.1 M TEAA buffer (pH 7.0) at 50 °C for 60 min with a flow rate of 0.5 mL/min using a C18 column.