

## **Supporting Information**

### **Oligonucleotides with “Clickable” Sugar Residues: Synthesis, Duplex Stability and Terminal versus Central Interstrand Cross-Linking of 2’-O-Propargylated 2-Aminoadenosine with a Bifunctional Azide**

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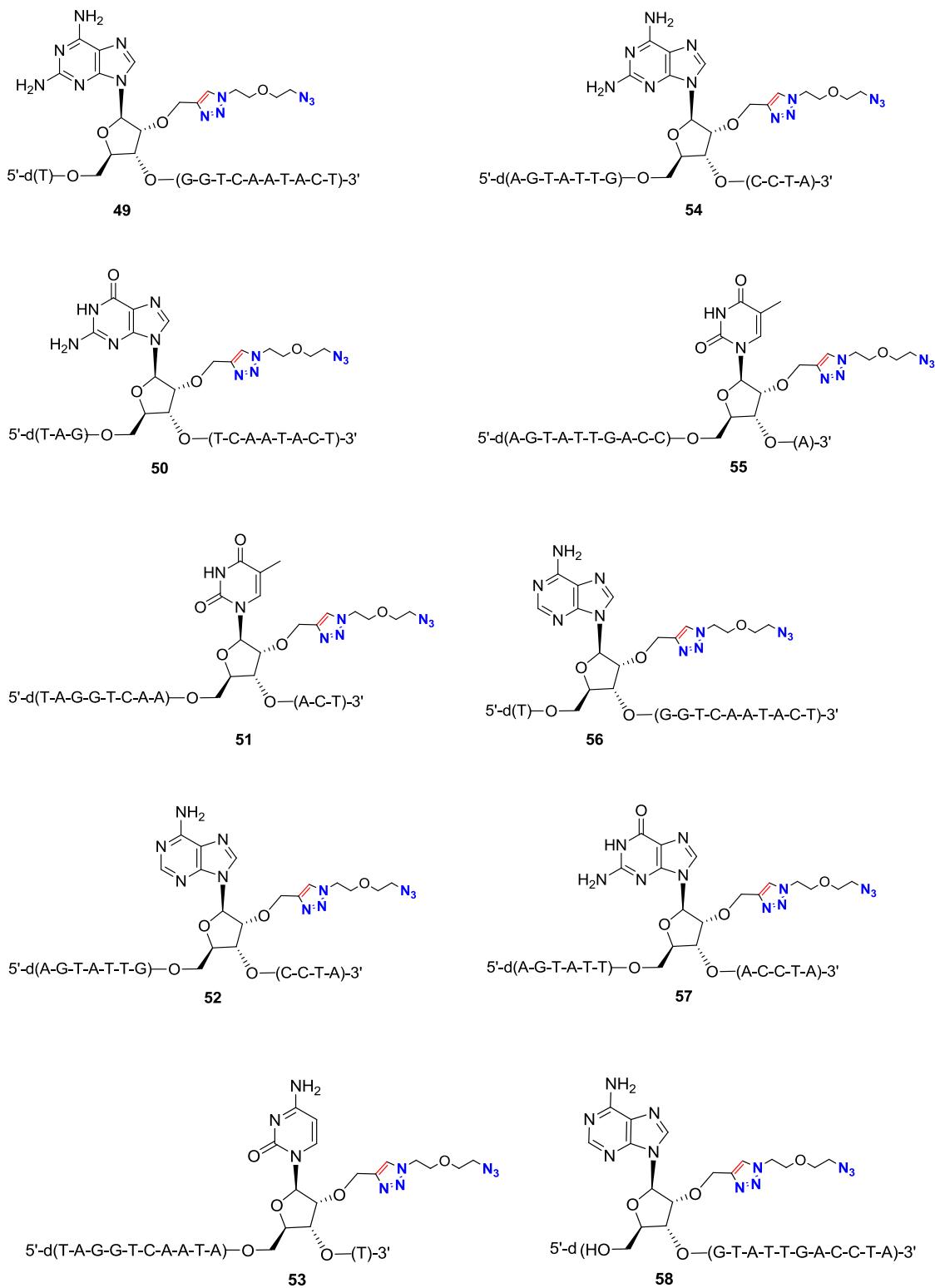
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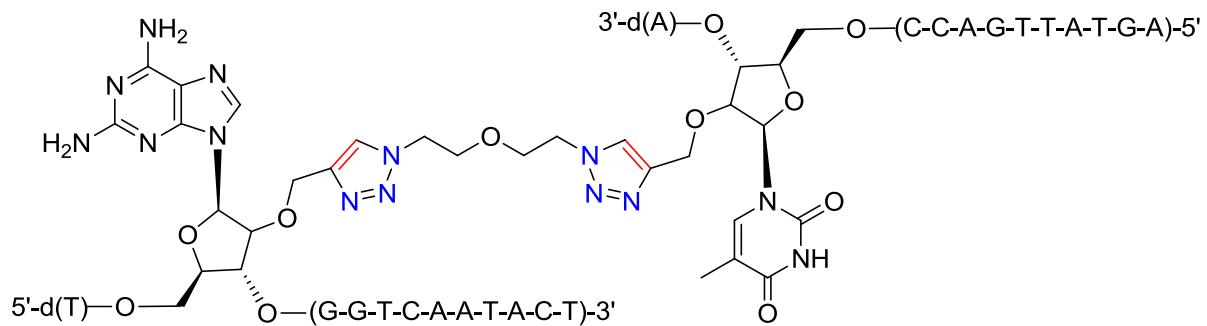
**Table S1.  $^1\text{H}$ - $^{13}\text{C}$  Coupling Constants (Hz) of 2,6-Diaminopurine Nucleosides and Their Derivatives.<sup>a,b</sup>**

$J$ (Hz)												
$^1\text{H}$ - $^{13}\text{C}$ -Coupling Constants	<b>1<sup>c</sup></b>	<b>7</b>	<b>8</b>	<b>9</b>	<b>11</b>	<b>12</b>	<b>13</b>	<b>15</b>	<b>16</b>	<b>17</b>	<b>19</b>	<b>20</b>
$^1J(\text{C8}, \text{H-C8})$	212.5	213.0	214.0	214.0	213.4	215.8	214.5	213.0	215.3	215.0	212.6	216.6
$^3J(\text{C8}, \text{H-C1'})$	4.2	4.4	4.1	3.8	4.3	4.2	3.7	4.0	4.0	--	3.9	--
$^1J(\text{C1'}, \text{H-C1'})$	163.8	163.0	167.8	166.7	164.7	165.5	164.3	164.6	165.5	166.0	164.2	163.1
$^1J(\text{C2'}, \text{H-C2'})$	151.4	147.4	--	--	149.7	149.0	149.5	149.6	147.8	148.0	145.7	146.9
$^1J(\text{C3'}, \text{H-C3'})$	149.2	150.0	149.0	147.5	151.2	149.8	149.7	149.0	149.4	148.1	150.8	148.4
$^1J(\text{C4'}, \text{H-C4'})$	147.4	148.0	147.2	137.0	148.7	148.3	149.4	149.6	148.2	149.5	147.6	148.4
$^1J(\text{C5'}, \text{H-C5'})$	140.4	140.0	141.0	143.6	140.4	140.7	141.9	141.0	141.0	141.5	141.6	140.0
$^1J(\text{C3}'', \text{H-C3}'')$	251.3	251.1	--	--	251.3	251.3	251.3	251.3	251.3	251.0	--	--

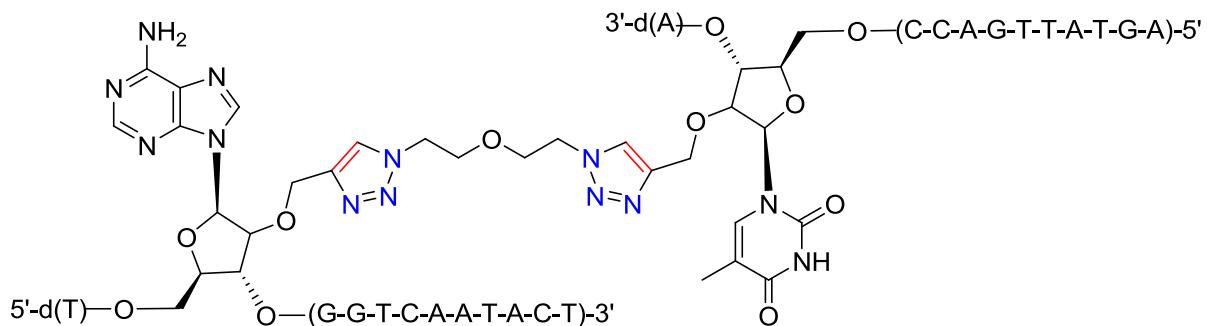
<sup>a</sup> Measured in DMSO ( $d_6$ ) at 298 K. <sup>b</sup> Purine numbering. <sup>c</sup> *J. Org. Chem.* **2013**, *78*, 8545-8561.



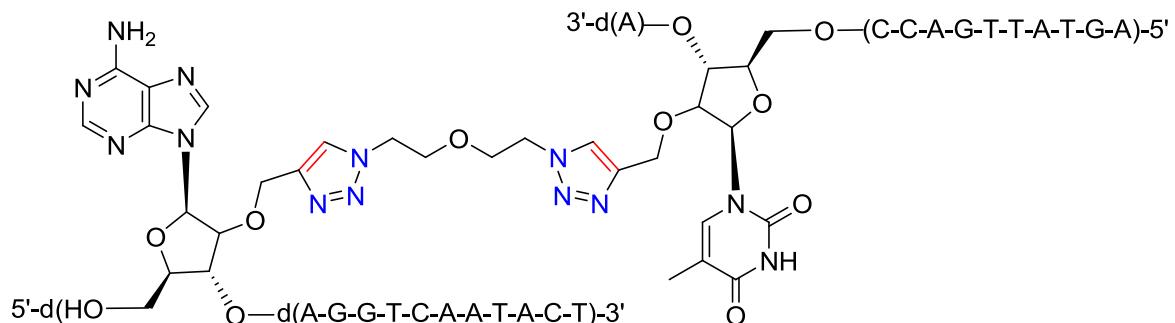
**Figure S1.** Structures of monofunctionalized oligonucleotides prepared by stepwise click reaction.



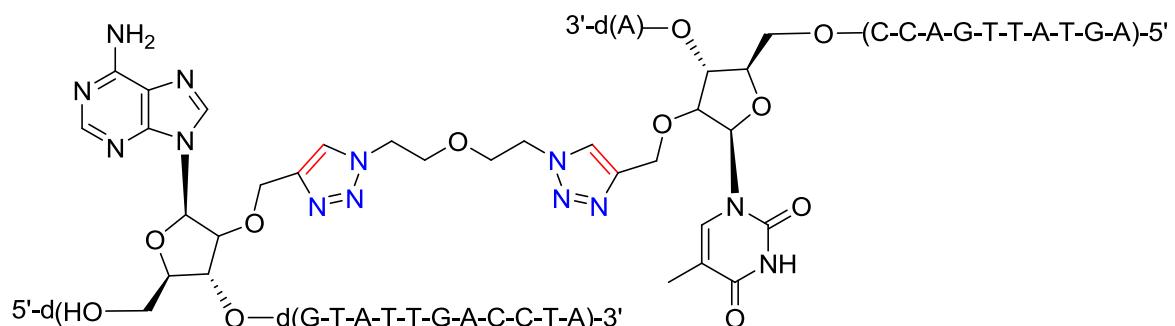
**ICL-1**



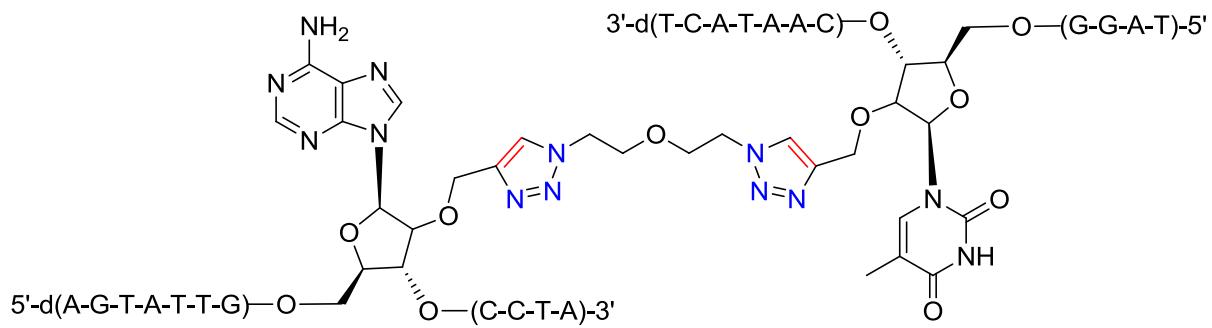
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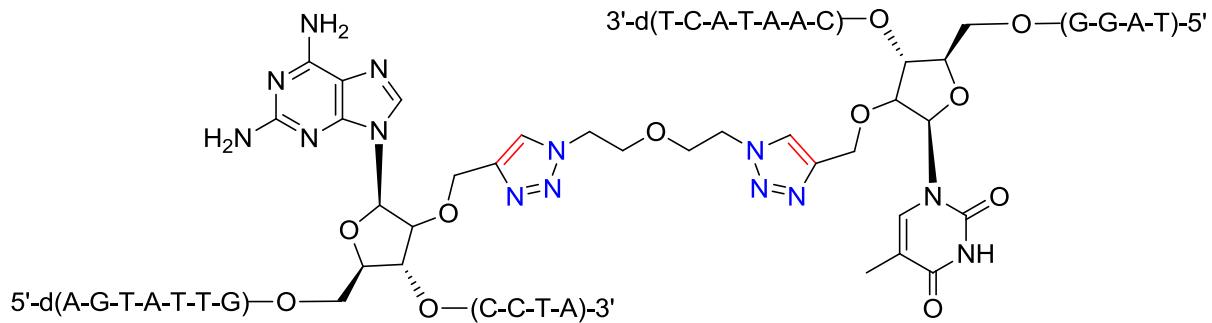
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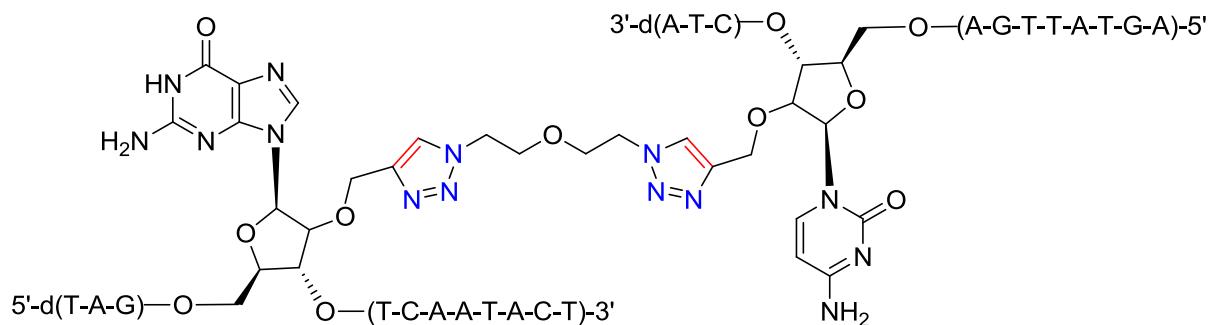
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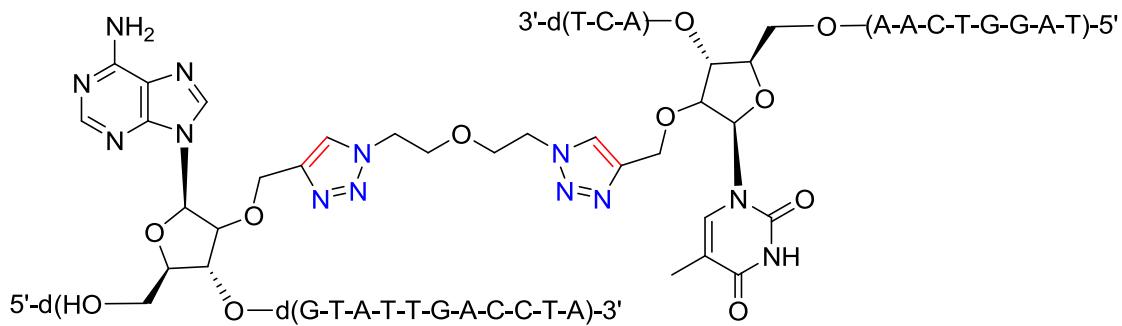
**ICL-5**



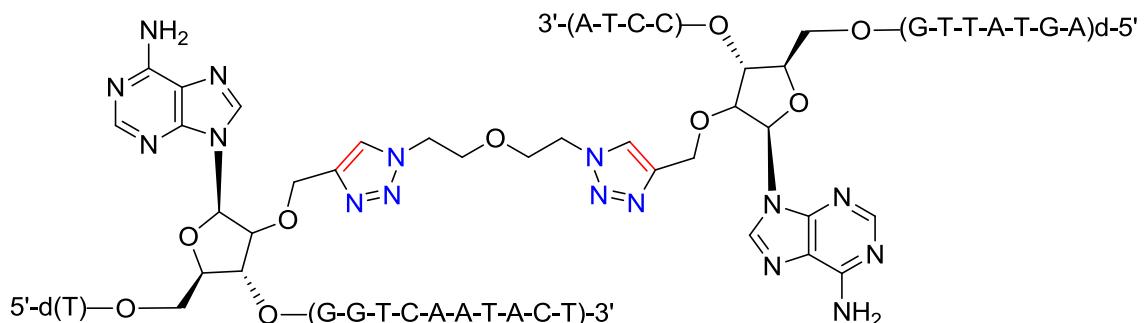
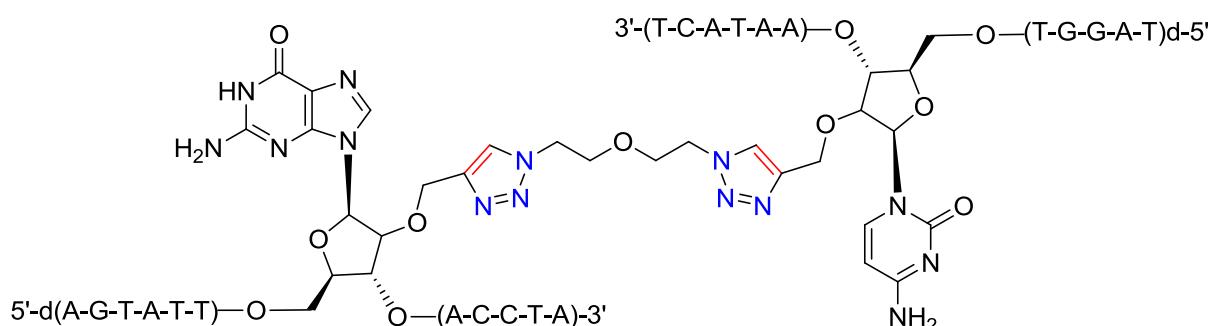
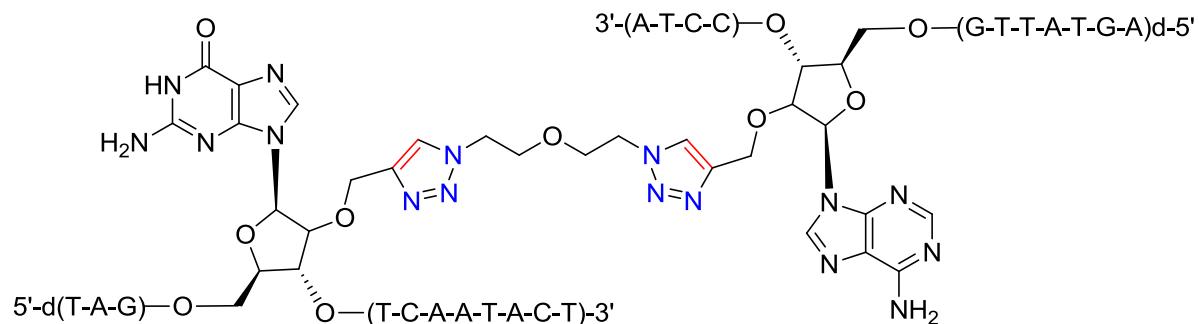
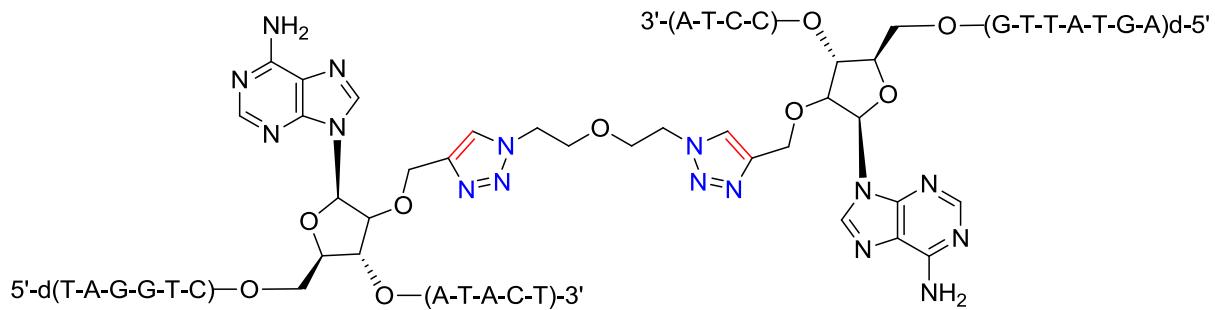
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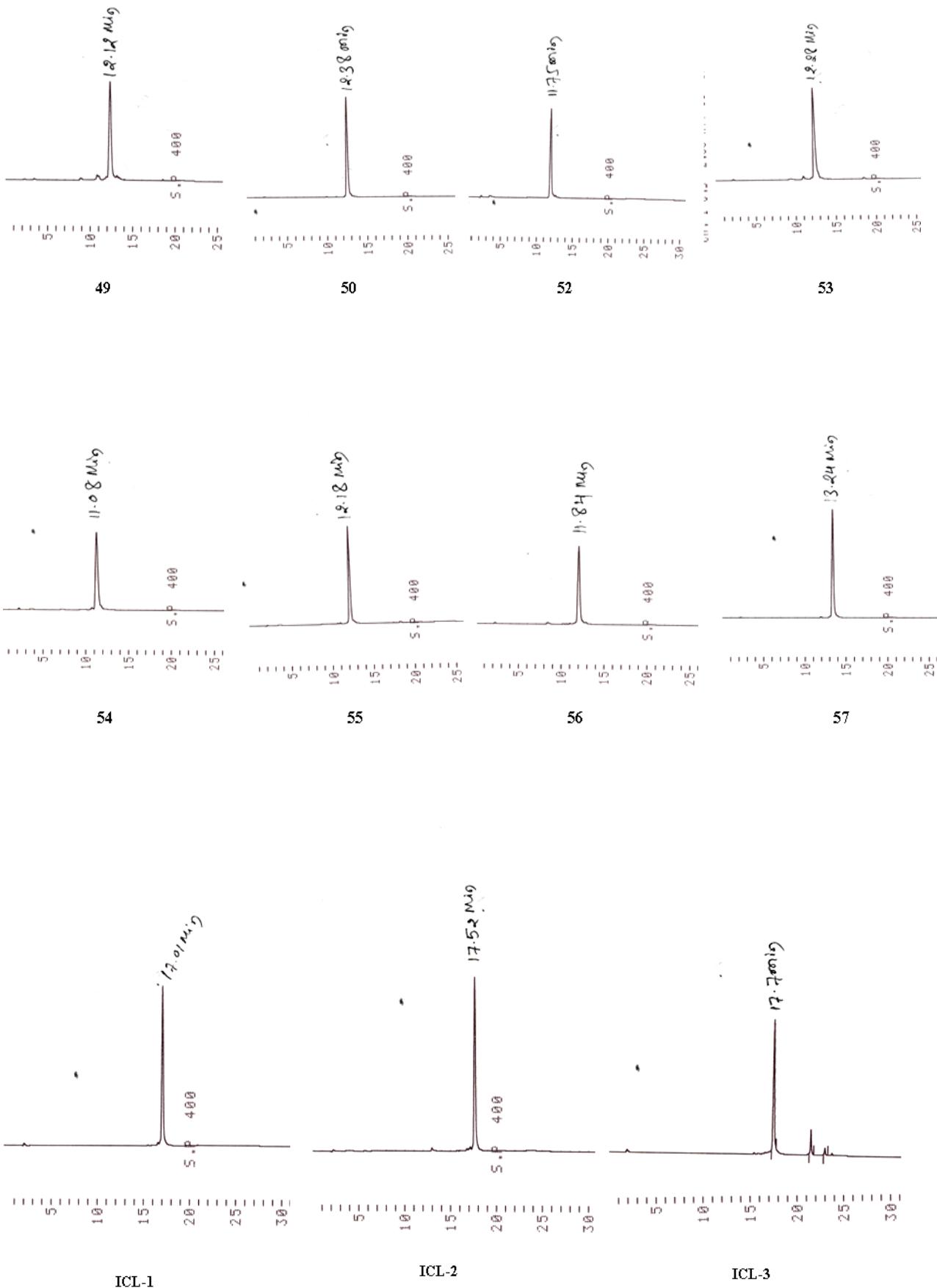
**ICL-7**

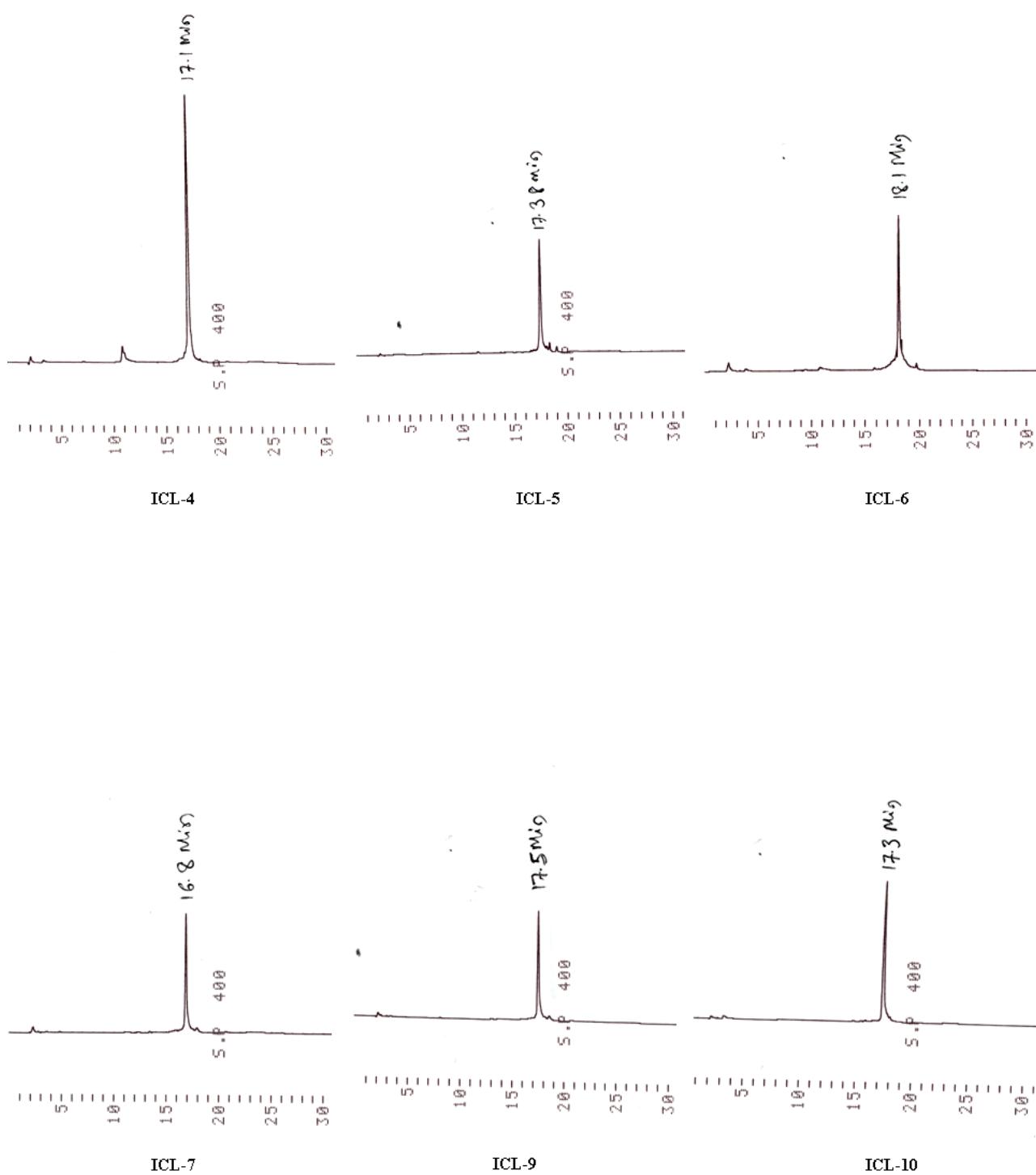


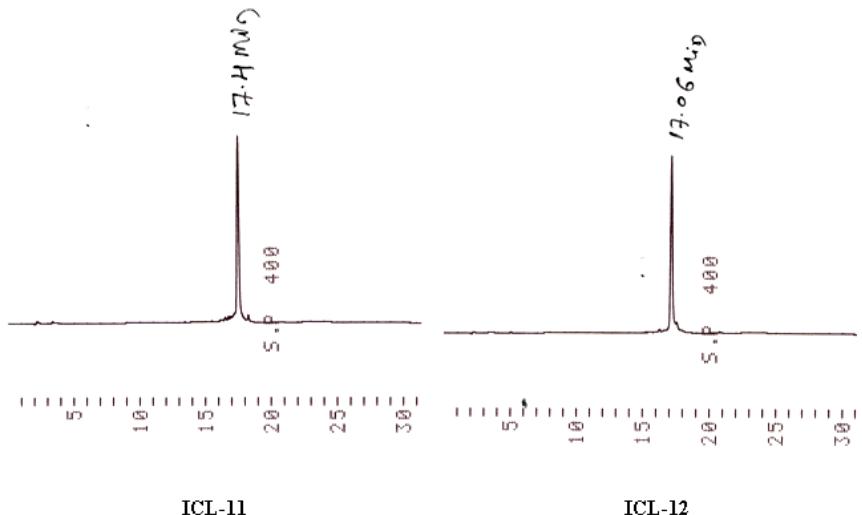
**ICL-8**



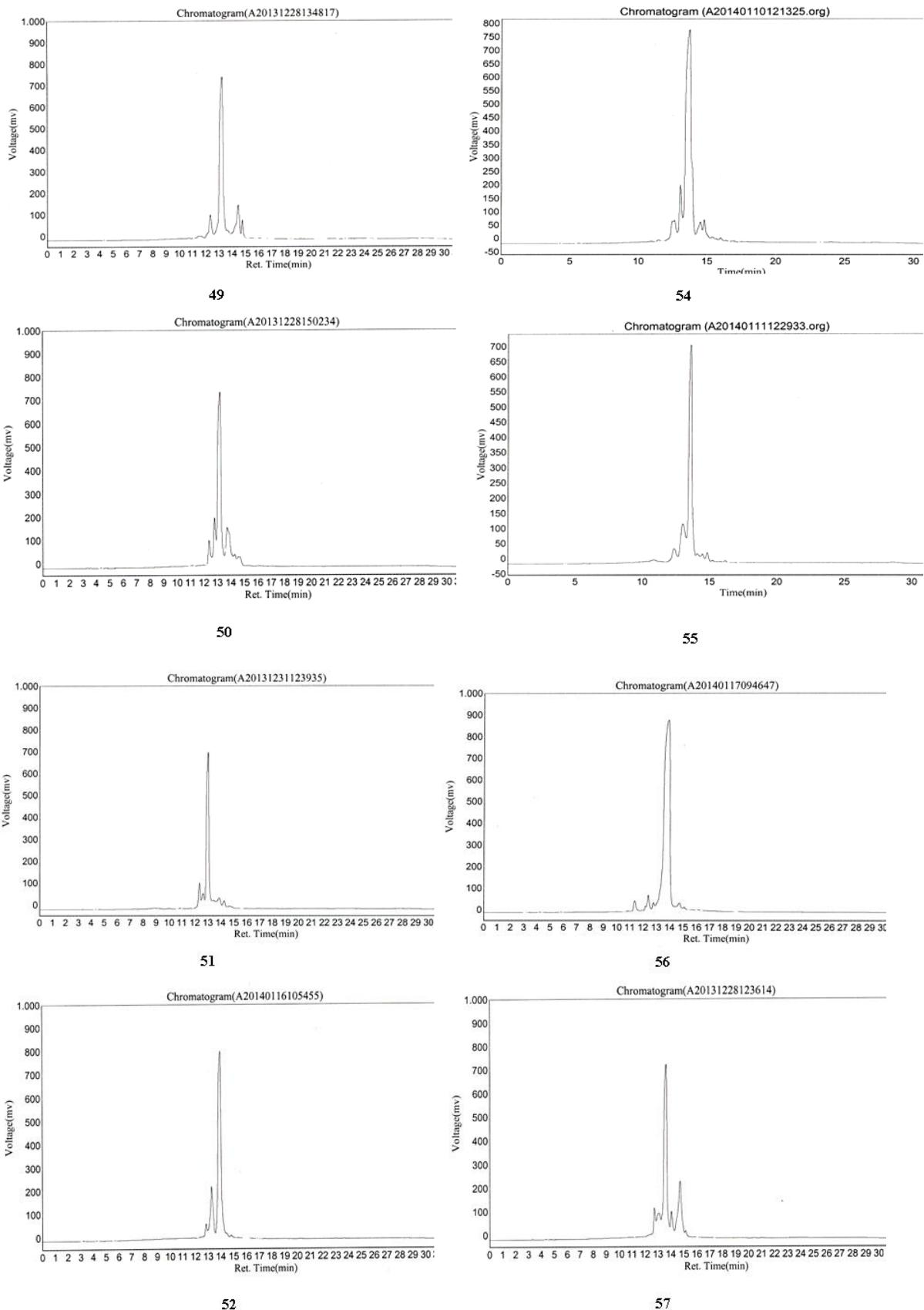
**Figure S2.** Structures of cross-linked oligonucleotides prepared by stepwise click reaction.

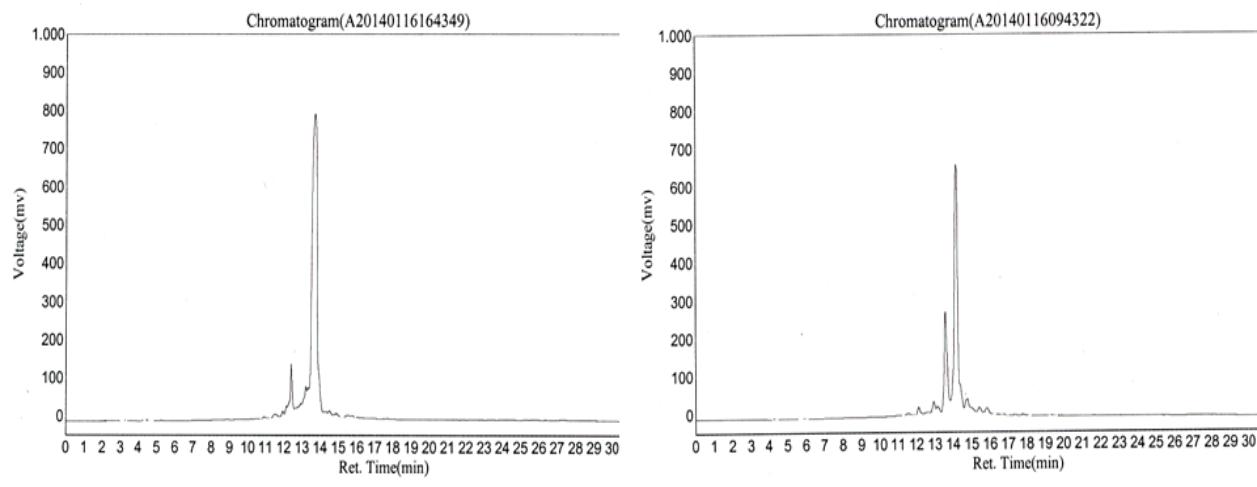






**Figure S3.** Ion exchange HPLC profiles of monofunctionalized and interstrand cross-linked oligonucleotides performed on a RP-18 column (4 x 250nm) at 260 nm. The compounds were eluted using the following gradient: (A) 25 mM Tris-HCl, 10% MeCN, pH 7.0; (B) 25 mM Tris-HCl, 1.0 M NaCl, and 10% MeCN, pH 7.0. Elution gradient: 0-30 min 20-80% B in A with a flow rate of 0.75 mL min<sup>-1</sup>. X-axis corresponds to retention time [min] and y-axis corresponds to absorbance.

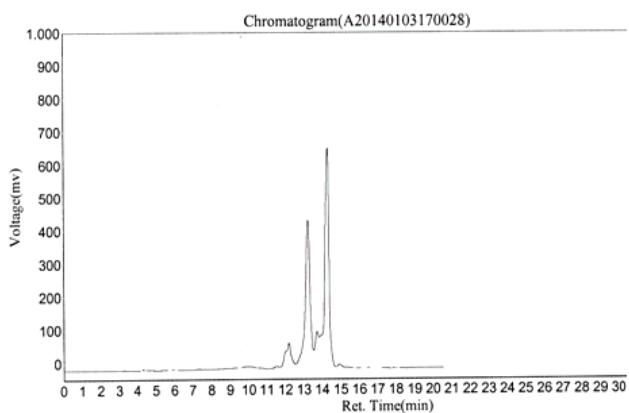




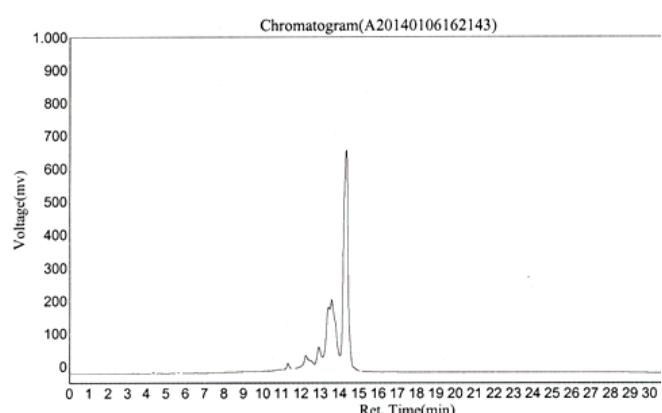
53

58

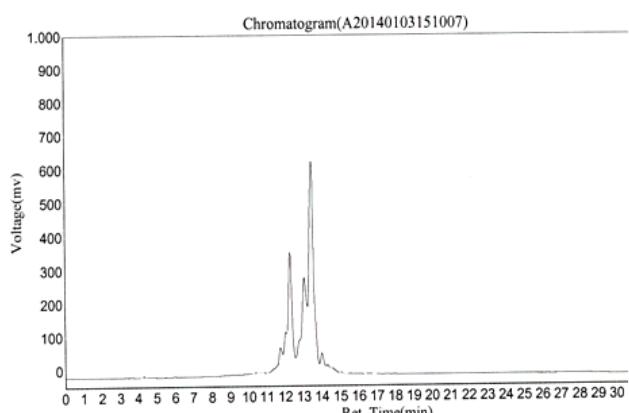
**Figure S4.** HPLC profiles of crude monofunctionalized oligonucleotides **49-58** obtained by stepwise click reaction (first click). Chromatography was performed on a RP-18 HPLC column (4 x 250 nm) at 260 nm with a gradient 0-25 min 0-20% A in B, flow rate 0.7 mL min<sup>-1</sup> where A = MeCN; B = 0.1 M (Et<sub>3</sub>NH)OAc (pH 7.0).



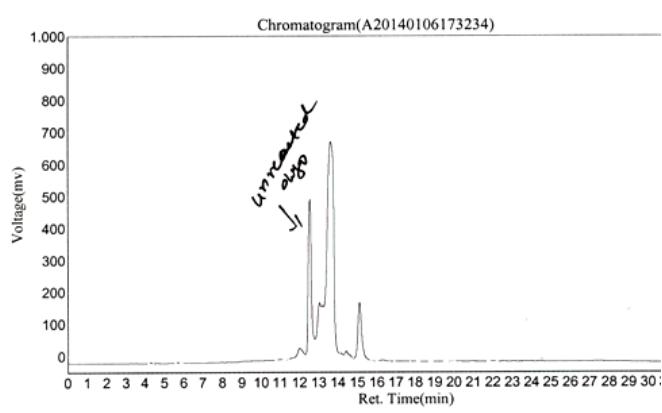
ICL-1



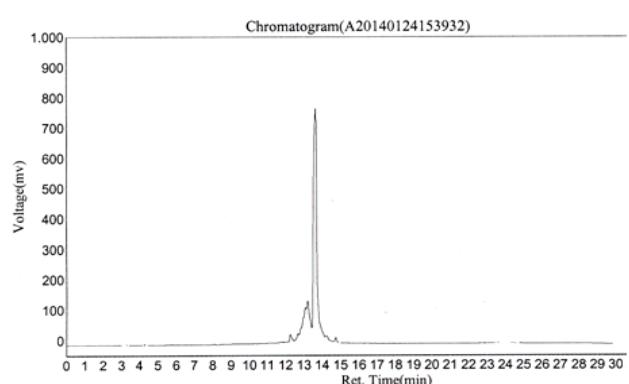
ICL-2



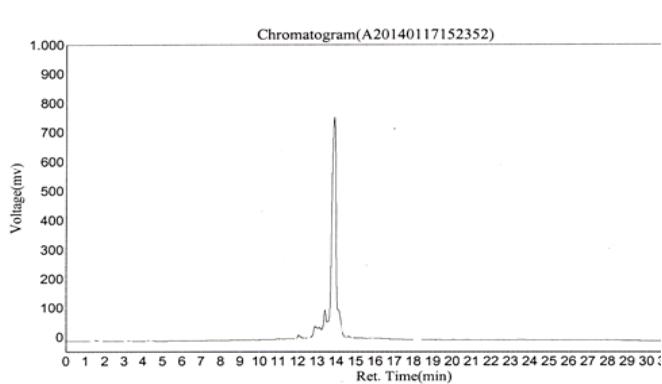
ICL-4



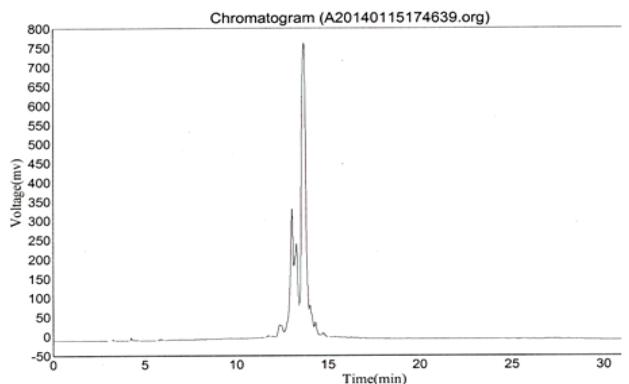
ICL-5



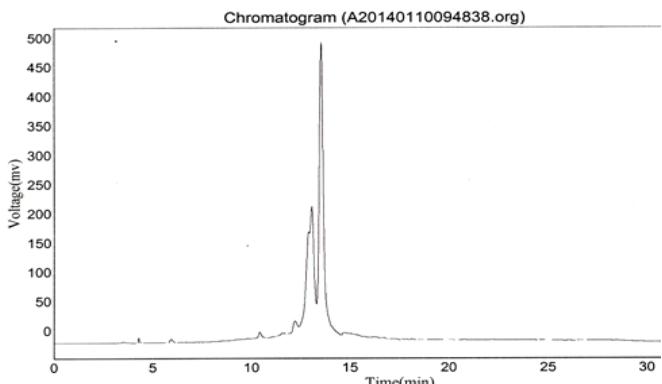
ICL-6



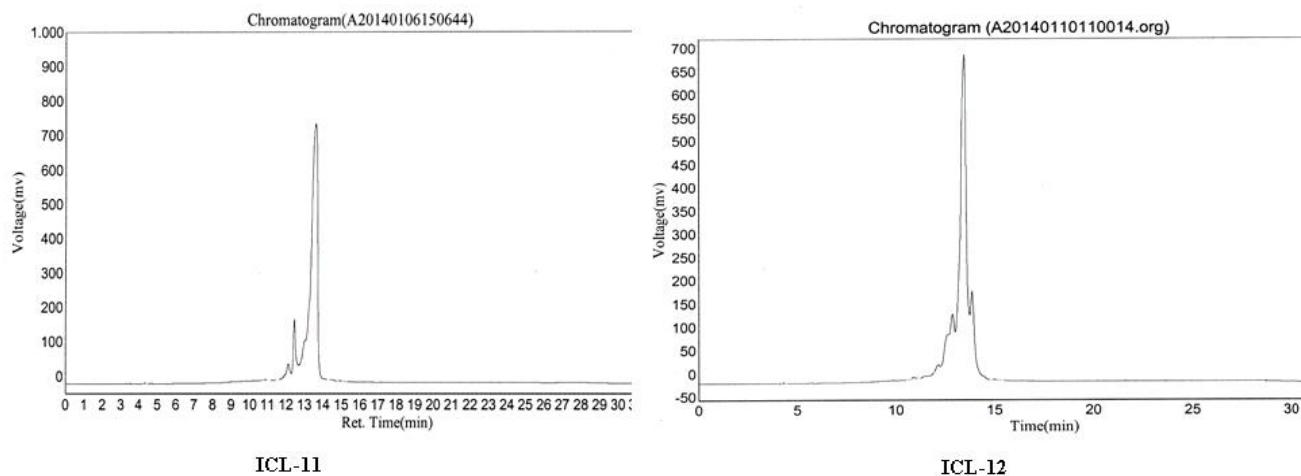
ICL-8



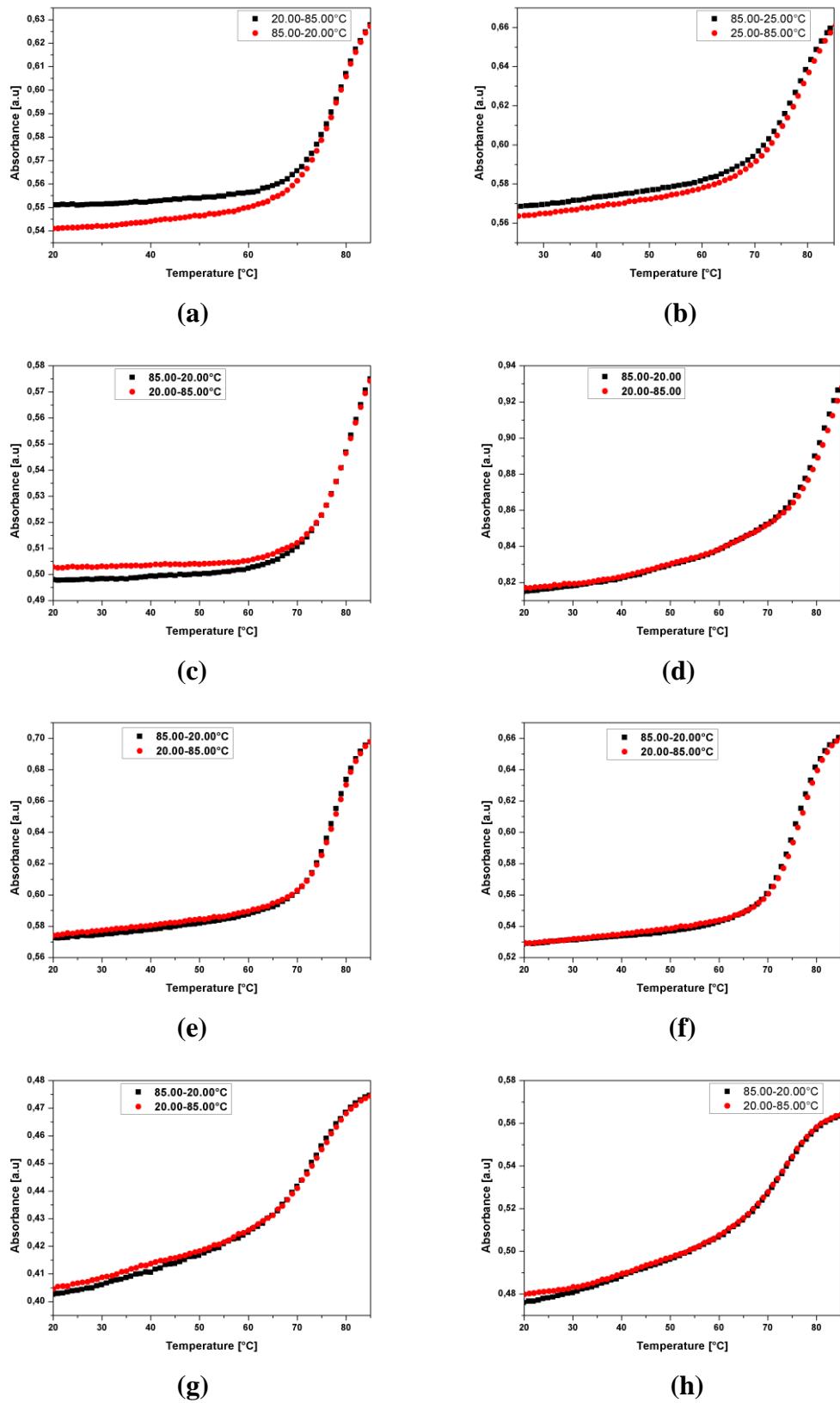
ICL-9

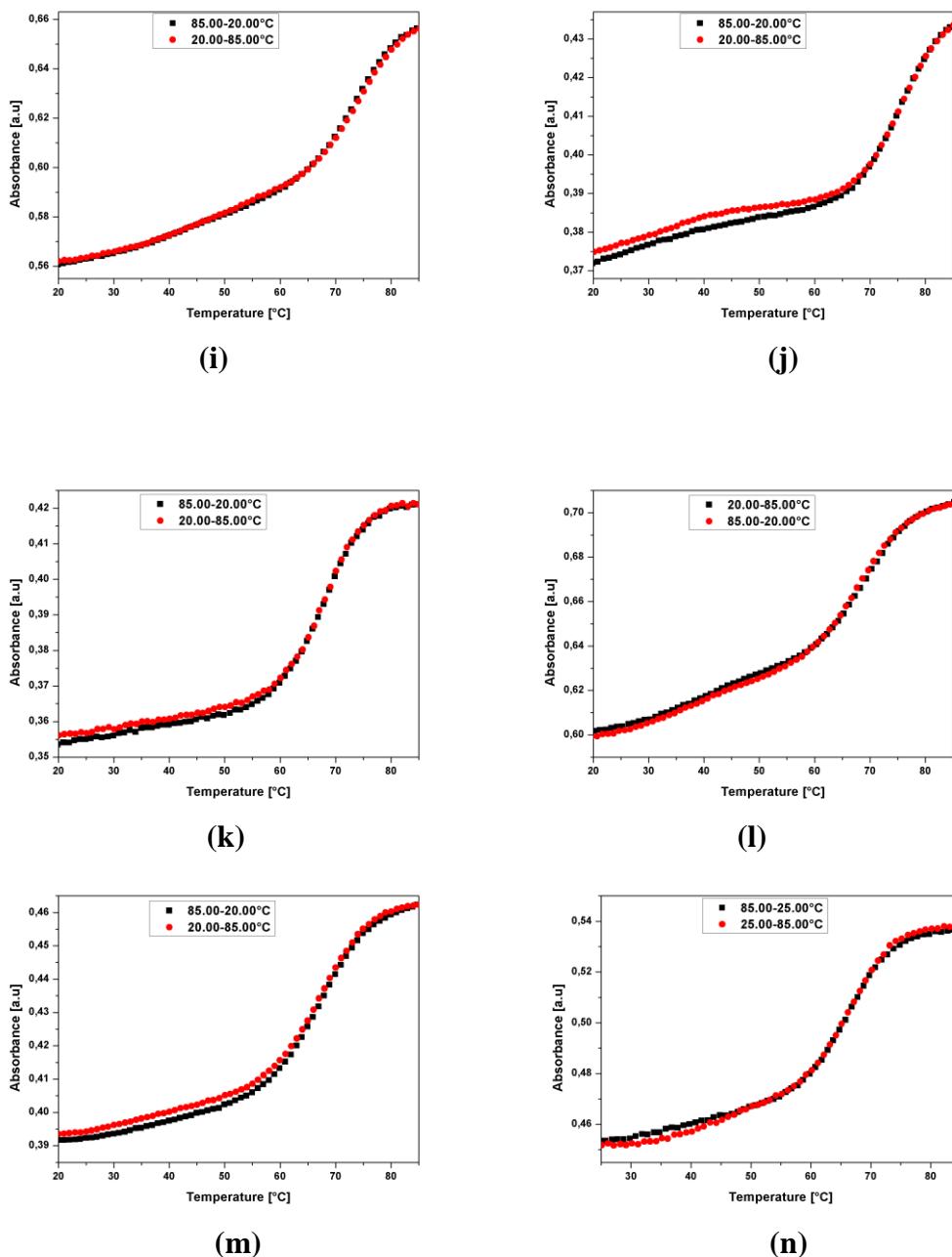


ICL-10

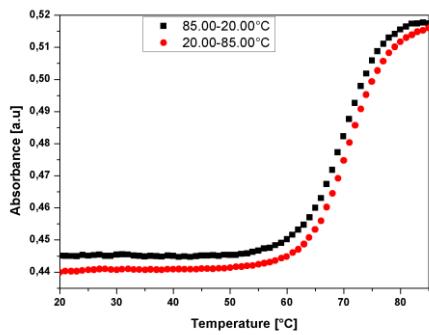


**Figure S5.** HPLC profiles of crude cross-linked oligonucleotides **ICL-1, ICL-2, ICL-4, ICL-5, ICL-6, ICL-8, ICL-9, ICL-10, ICL-11** and **ICL-12** obtained by stepwise click reaction (second click). Chromatography was performed on a RP-18 HPLC column ( $4 \times 250$  nm) at  $260$  nm with a gradient  $0\text{-}25$  min  $0\text{-}20\%$  A in B, flow rate  $0.7$  mL  $\text{min}^{-1}$  where A = MeCN; B =  $0.1$  M  $(\text{Et}_3\text{NH})\text{OAc}$  (pH 7.0).

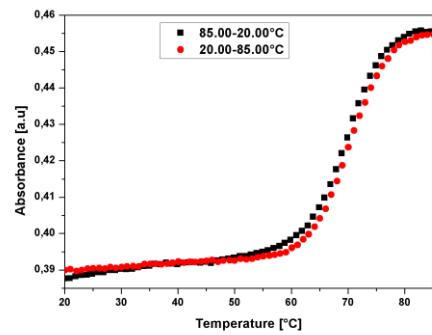




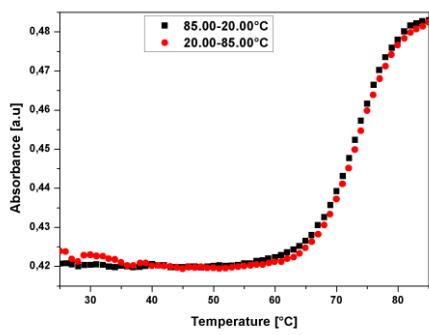
**Figure S6.** The melting profiles of cross-linked oligonucleotides (a) **ICL-1**; (b) **ICL-2**; (c) **ICL-3**; (d) **ICL-4**; (e) hairpin **H1**; (f) hairpin **H2**; (g) **ICL-5**; (h) **ICL-6**; (i) **ICL-7**; (j) **ICL-8**; (k) **ICL-9**; (l) **ICL-10**; (m) **ICL-11**; (n) **ICL-12**. The melting profiles were measured in 1 M NaCl, 100 mM MgCl<sub>2</sub>, 60 mM Na-cacodylate buffer.



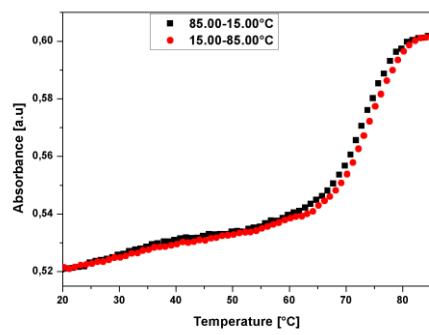
(a)



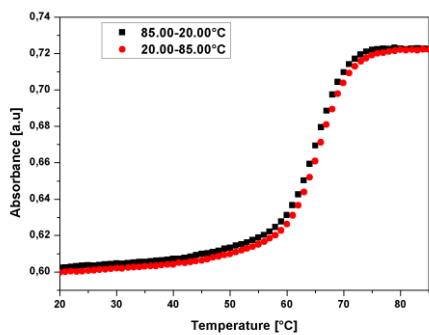
(b)



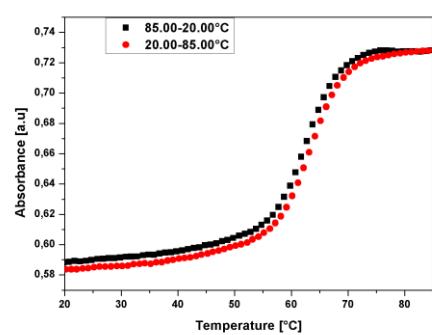
(c)



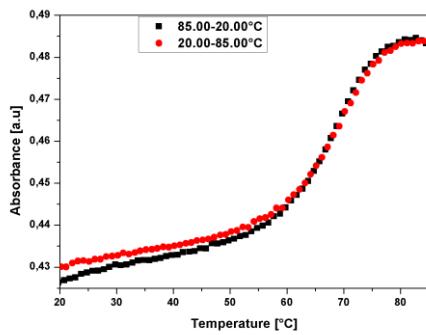
(d)



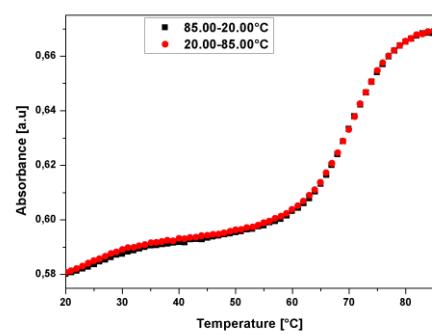
(e)



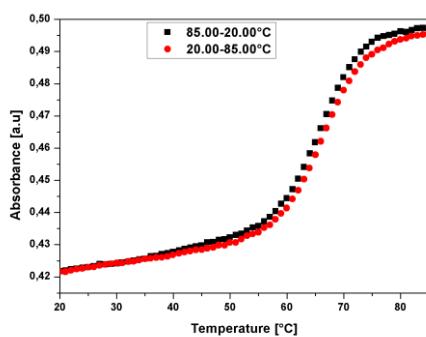
(f)



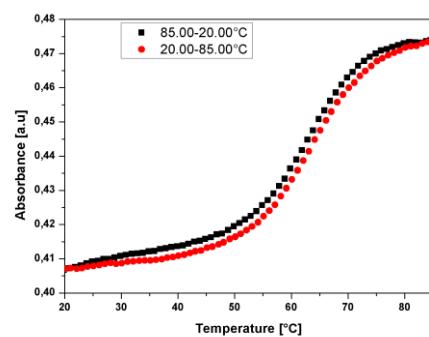
(g)



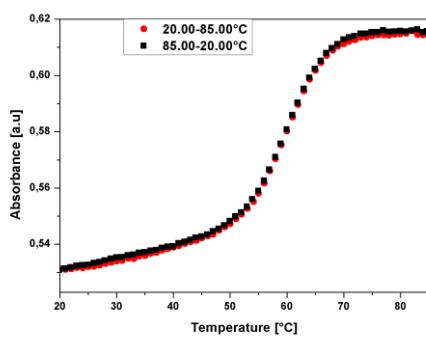
(h)



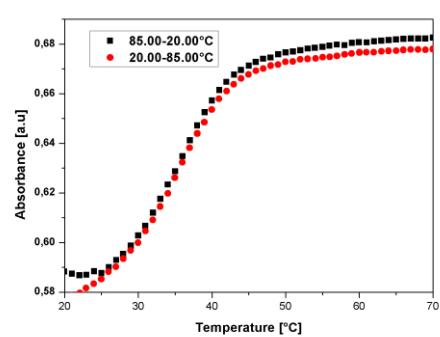
(i)



(j)



(k)



(l)

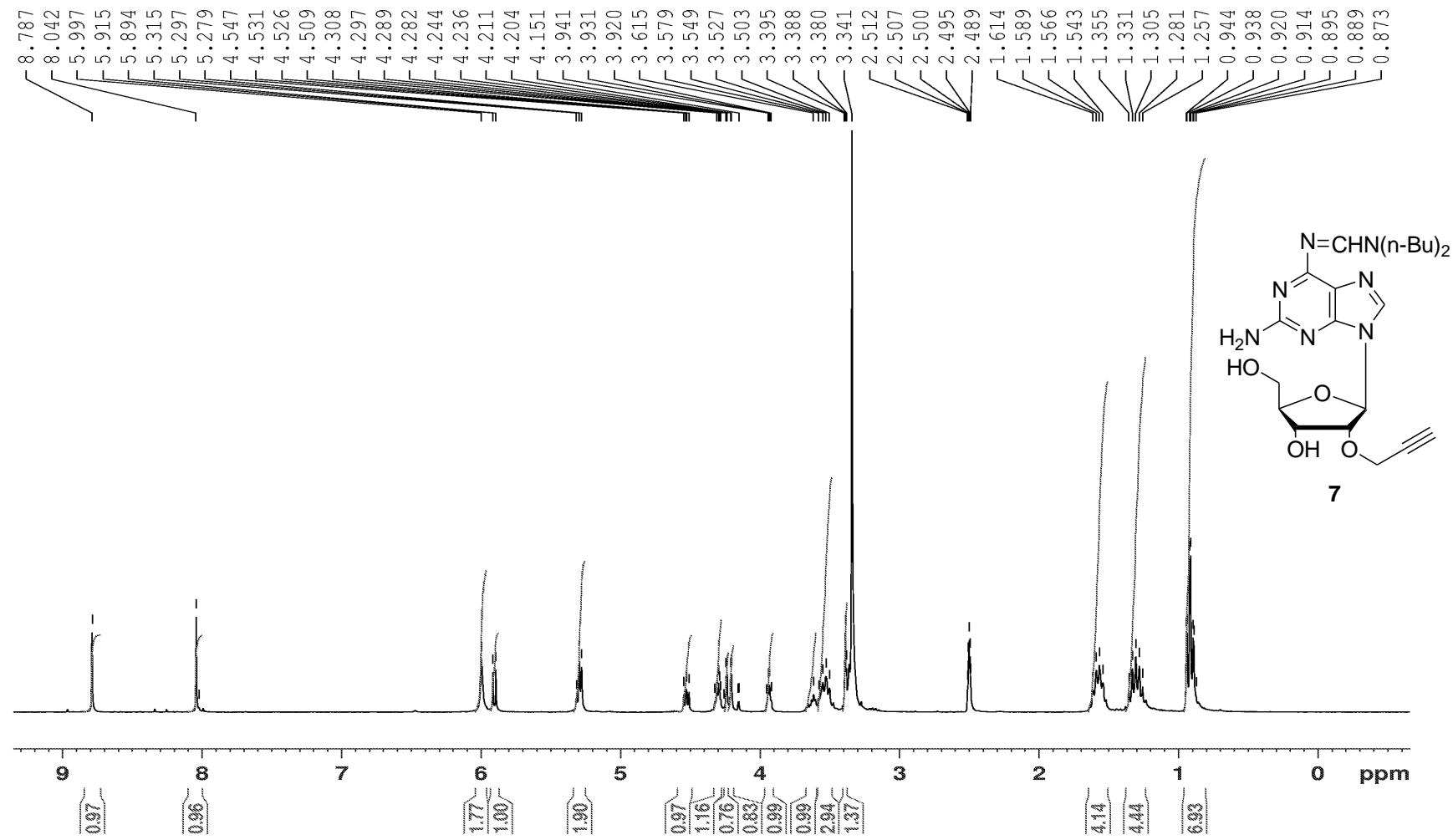
**Figure S7.** The melting profiles of cross-linked oligonucleotides (a) ICL-1; (b) ICL-2; (c) ICL-3; (d) ICL-4; (e) hairpin H1; (f) hairpin H2; (g) ICL-5; (h) ICL-6; (i) ICL-7; (j) ICL-8; (k) ICL-9; (l) duplex 26•27. The melting profiles were measured in 50 mM NaCl, 10 mM Na<sub>3</sub>PO<sub>4</sub>, 0.1 mM EDTA buffer.

**Table S2. UV Maxima<sup>a</sup> and Extinction Coefficients ( $\epsilon$ )<sup>b</sup> of Nucleoside Derivatives.<sup>c</sup>**

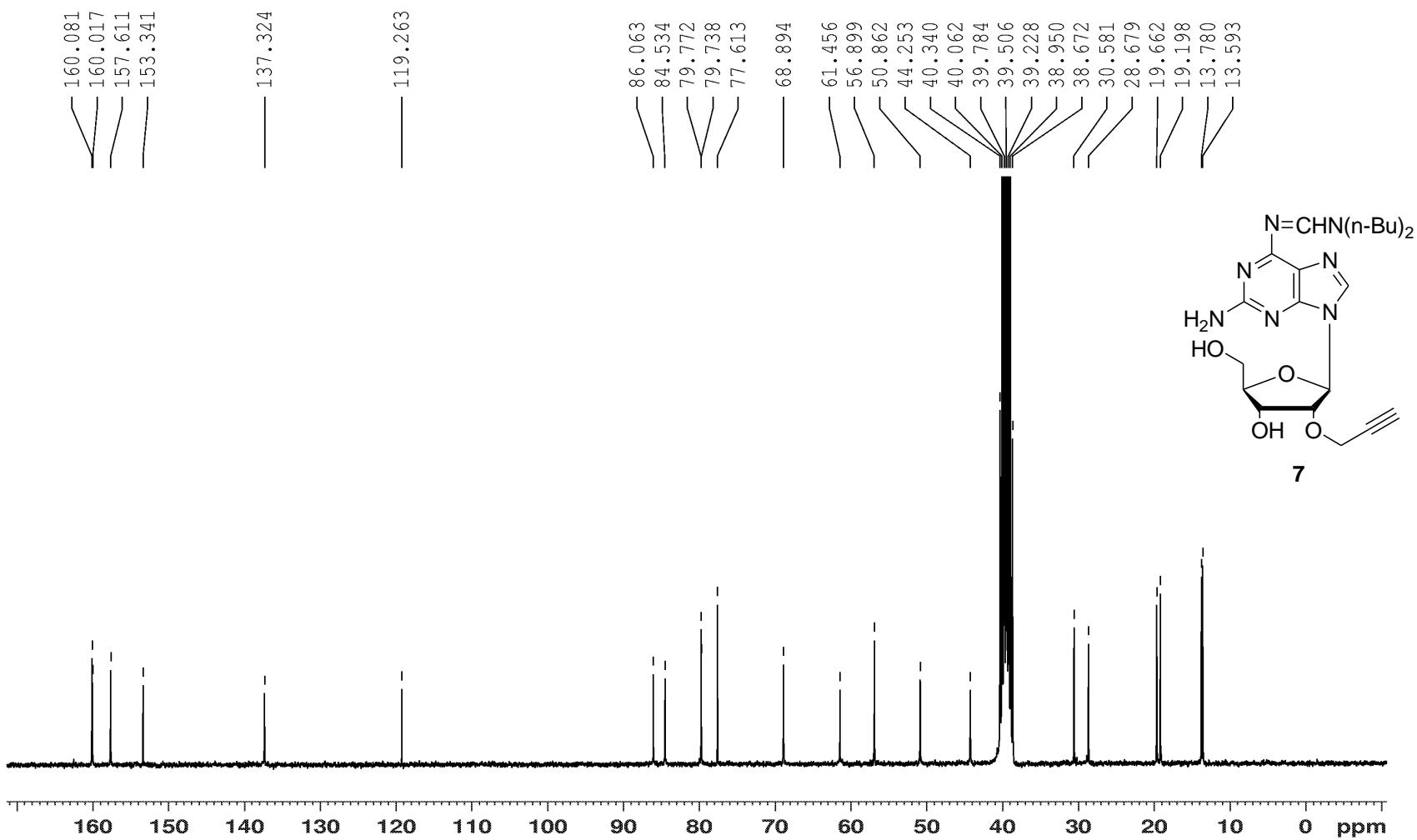
Compound	UV Data	Compound	UV Data
	$\lambda_{\text{max}}$ : 256 ( $\epsilon$ 9800) 281 ( $\epsilon$ 10500) $\lambda_{260}$ : ( $\epsilon$ 9000)		$\lambda_{\text{max}}$ : 256 ( $\epsilon$ 8900) 281 ( $\epsilon$ 9800) $\lambda_{260}$ : ( $\epsilon$ 8200)
	$\lambda_{\text{max}}$ : 256 ( $\epsilon$ 9400) 281 ( $\epsilon$ 10900) $\lambda_{260}$ : ( $\epsilon$ 8800)		
	$\lambda_{\text{max}}$ : 271 ( $\epsilon$ 15800)		$\lambda_{\text{max}}$ : 271 ( $\epsilon$ 14800)
	$\lambda_{\text{max}}$ : 253 ( $\epsilon$ 13900) 279 ( $\epsilon$ 14500) 325 ( $\epsilon$ 20600)		$\lambda_{\text{max}}$ : 260 ( $\epsilon$ 20700) 320 ( $\epsilon$ 29500)
	$\lambda_{\text{max}}$ : 265 ( $\epsilon$ 10000) 286 ( $\epsilon$ 12000)		$\lambda_{\text{max}}$ : 265 ( $\epsilon$ 11500) 287 ( $\epsilon$ 11200)
	$\lambda_{\text{max}}$ : 267 ( $\epsilon$ 14000) 284 ( $\epsilon$ 15900)		$\lambda_{\text{max}}$ : 268 ( $\epsilon$ 16200sh) 276 ( $\epsilon$ 16700)
	$\lambda_{\text{max}}$ : 260 ( $\epsilon$ 27900) 320 ( $\epsilon$ 30700)		

<sup>a</sup>Wavelength is given in nm. <sup>b</sup>Unit for extinction coefficient is  $\text{dm}^3 \text{ mol}^{-1} \text{ cm}^{-1}$ . <sup>c</sup>Measured in MeOH.

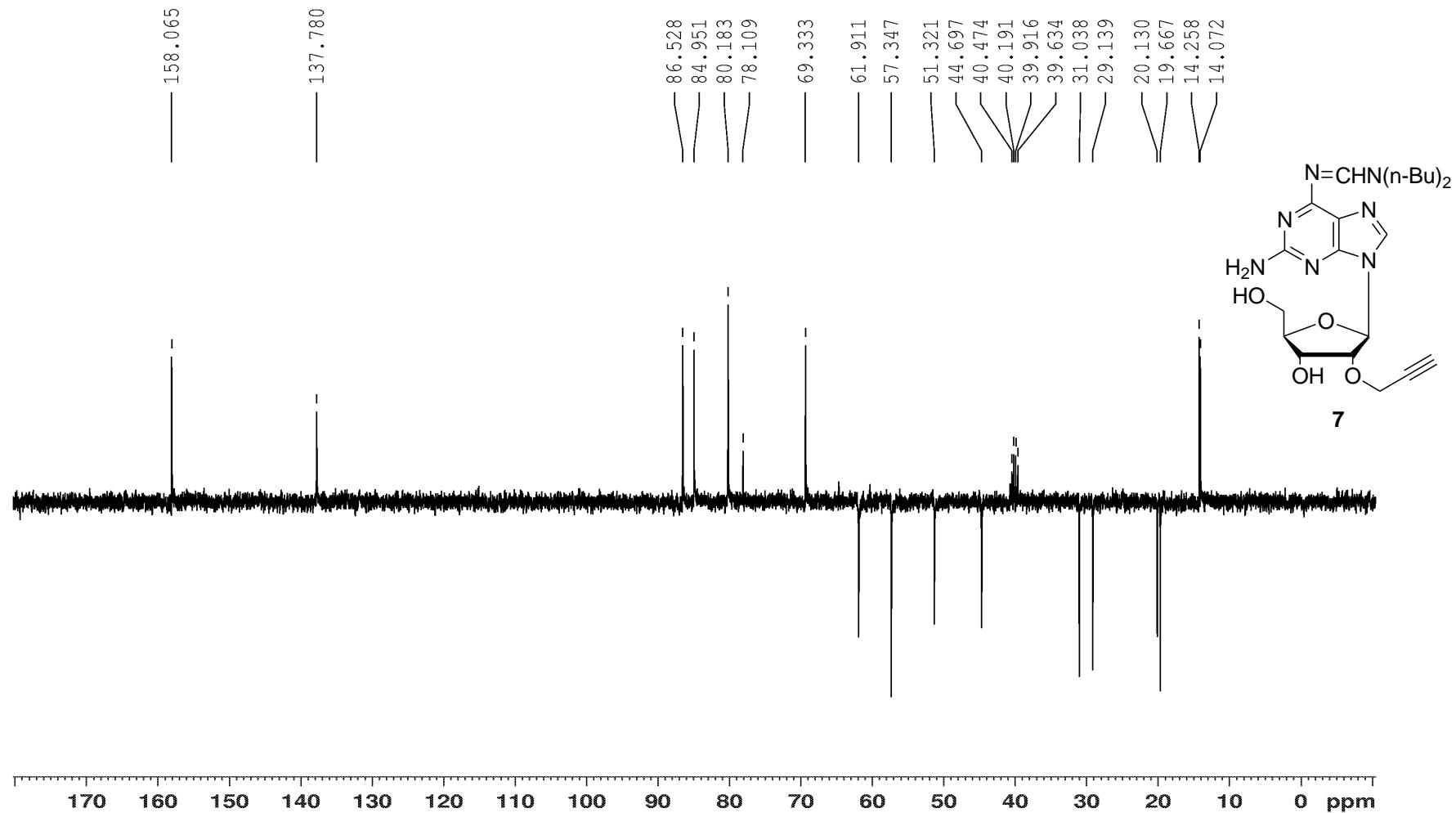
**Figure S8.**  $^1\text{H}$  NMR spectrum of compound 7.



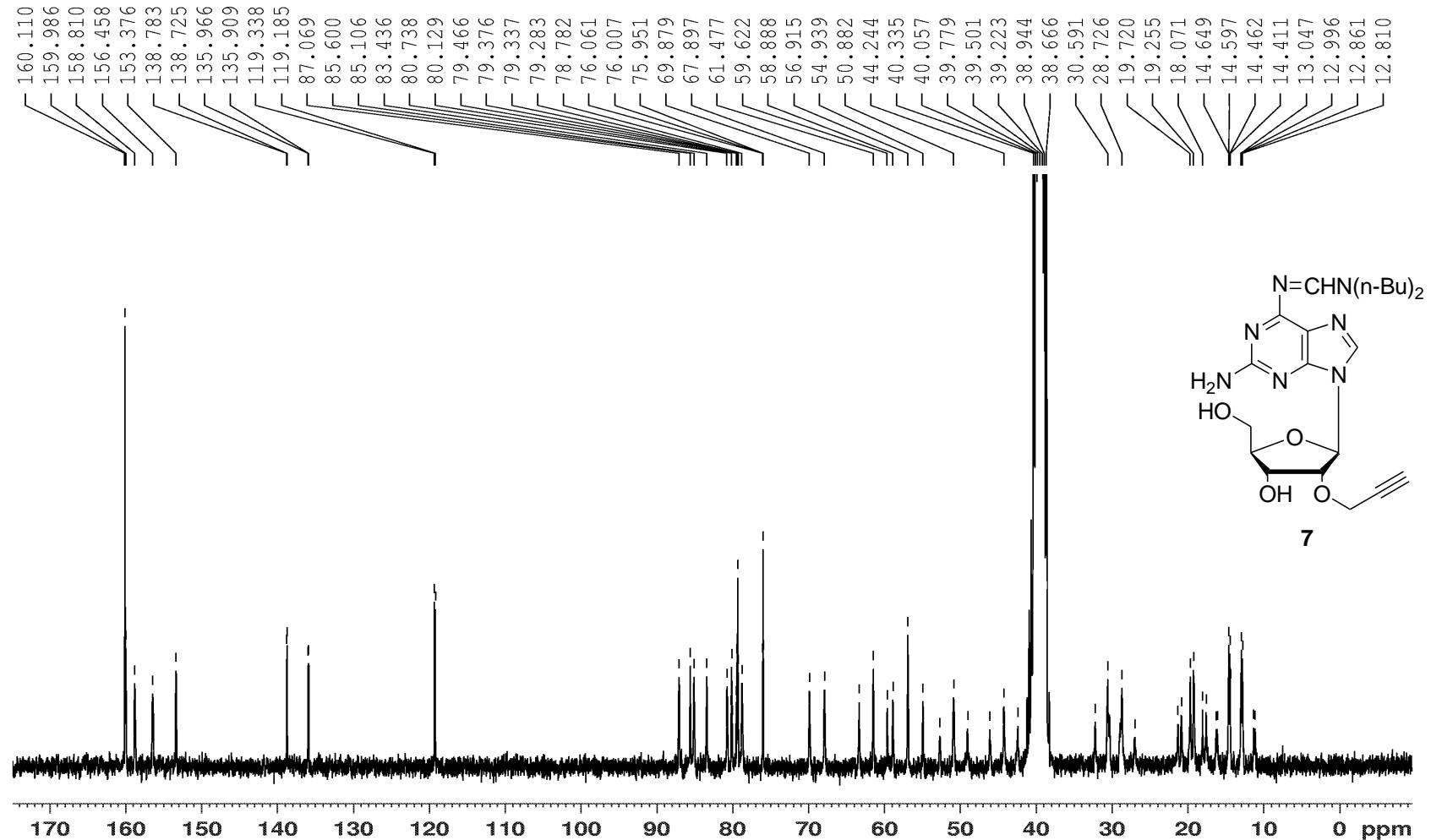
**Figure S9.**  $^{13}\text{C}$  NMR spectrum of compound 7.



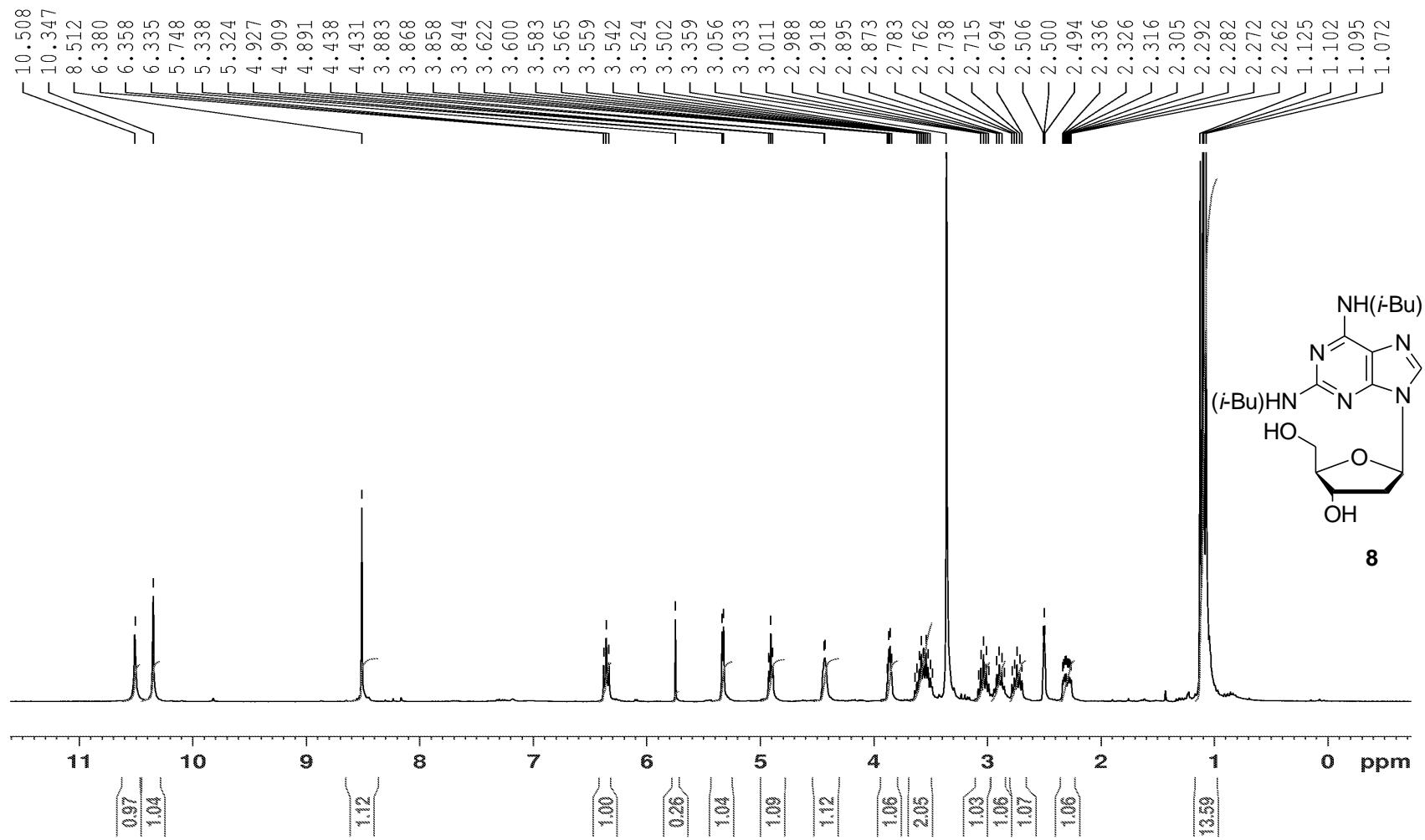
**Figure S10.** DEPT-135 spectrum of compound 7.



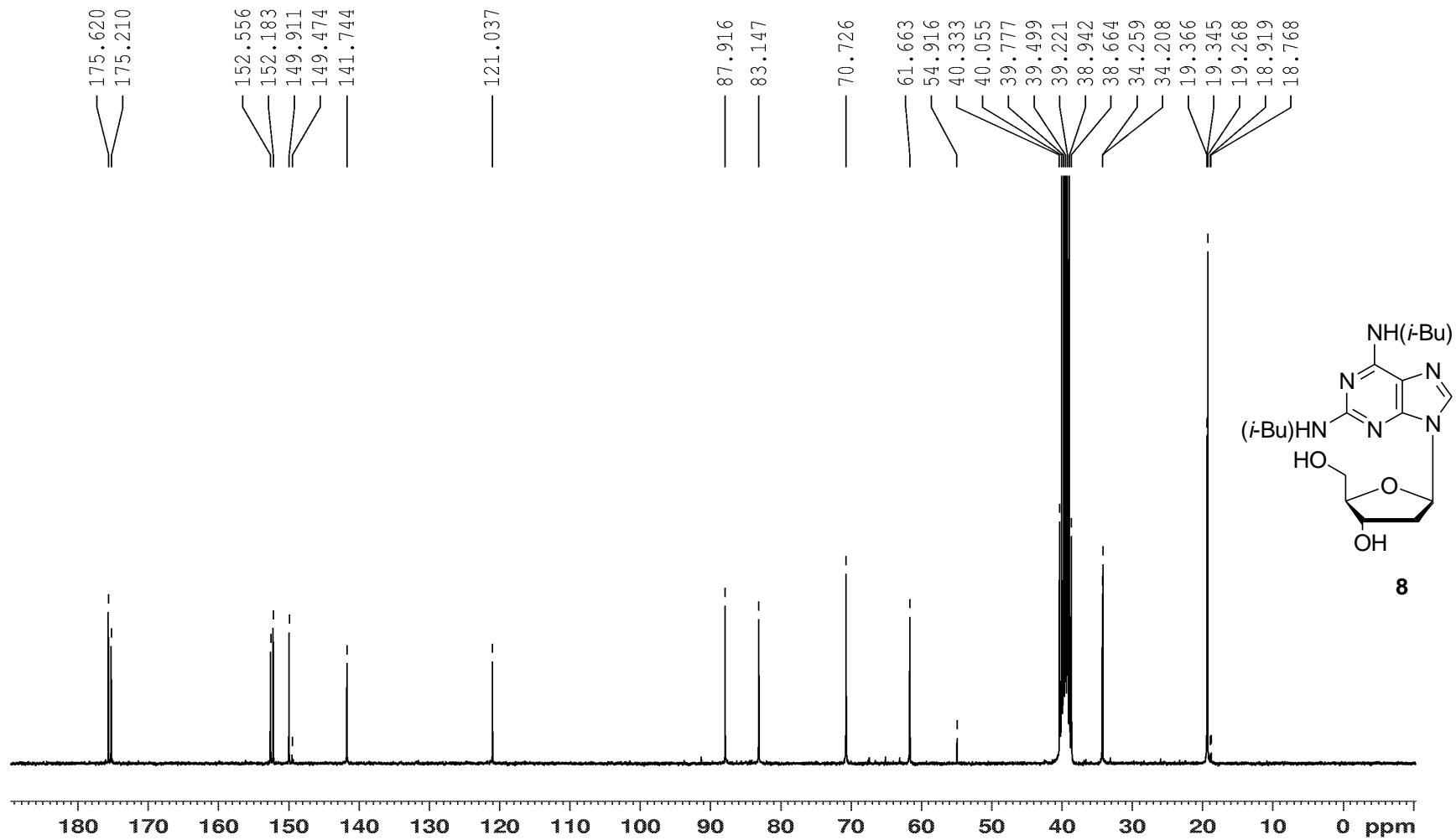
**Figure S11.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 7.



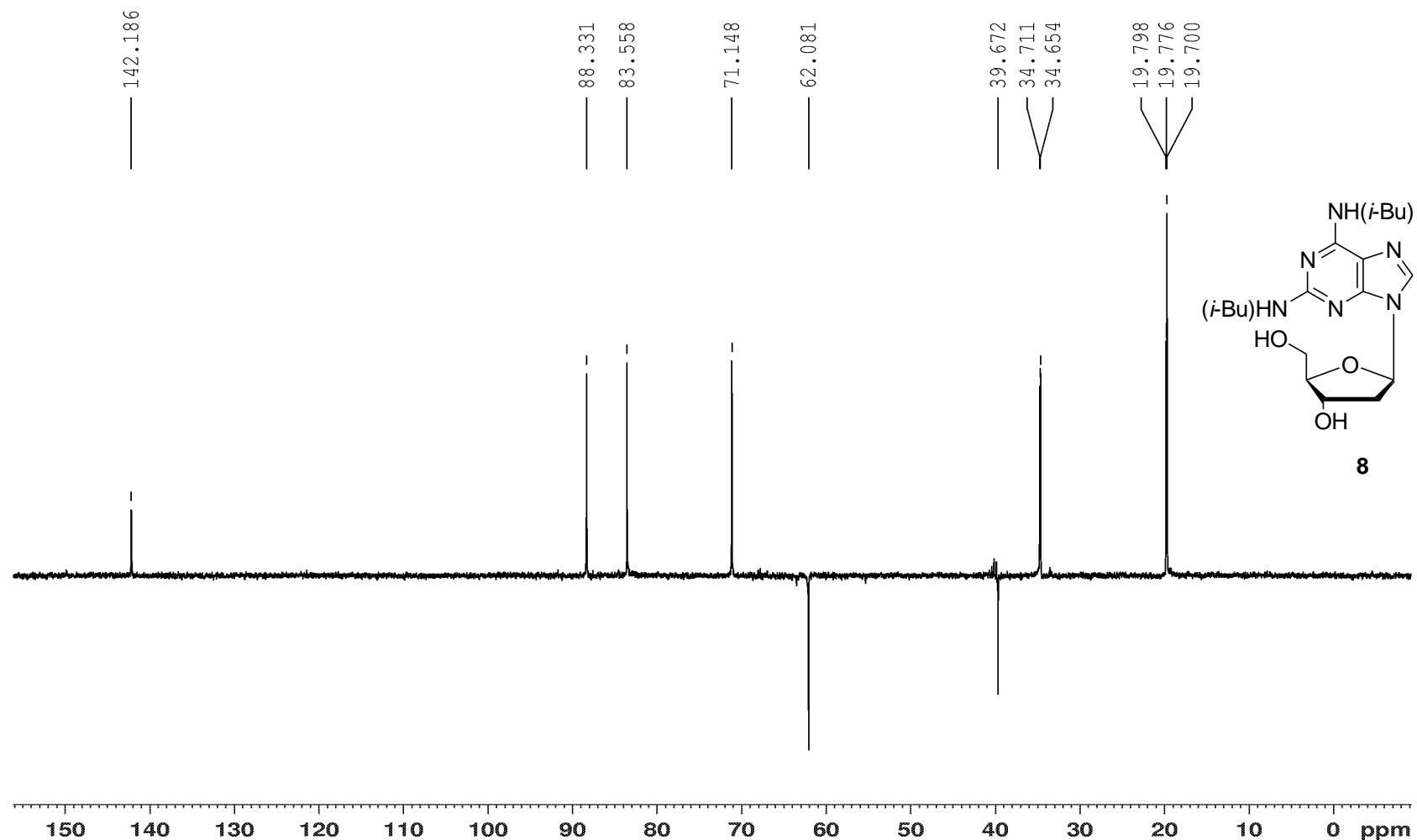
**Figure S12.**  $^1\text{H}$  NMR spectrum of compound 8.



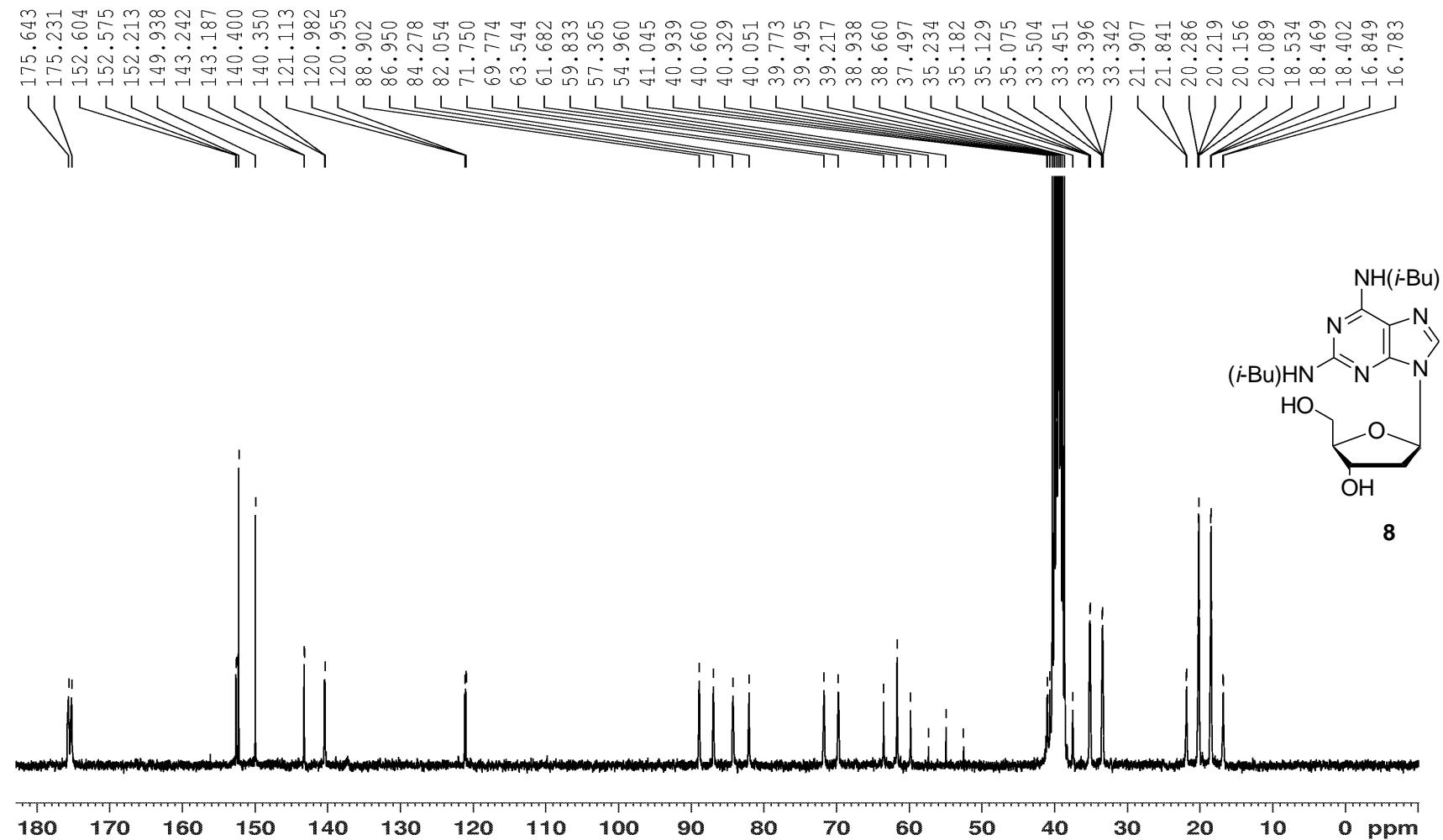
**Figure S13.**  $^{13}\text{C}$  NMR spectrum of compound 8.



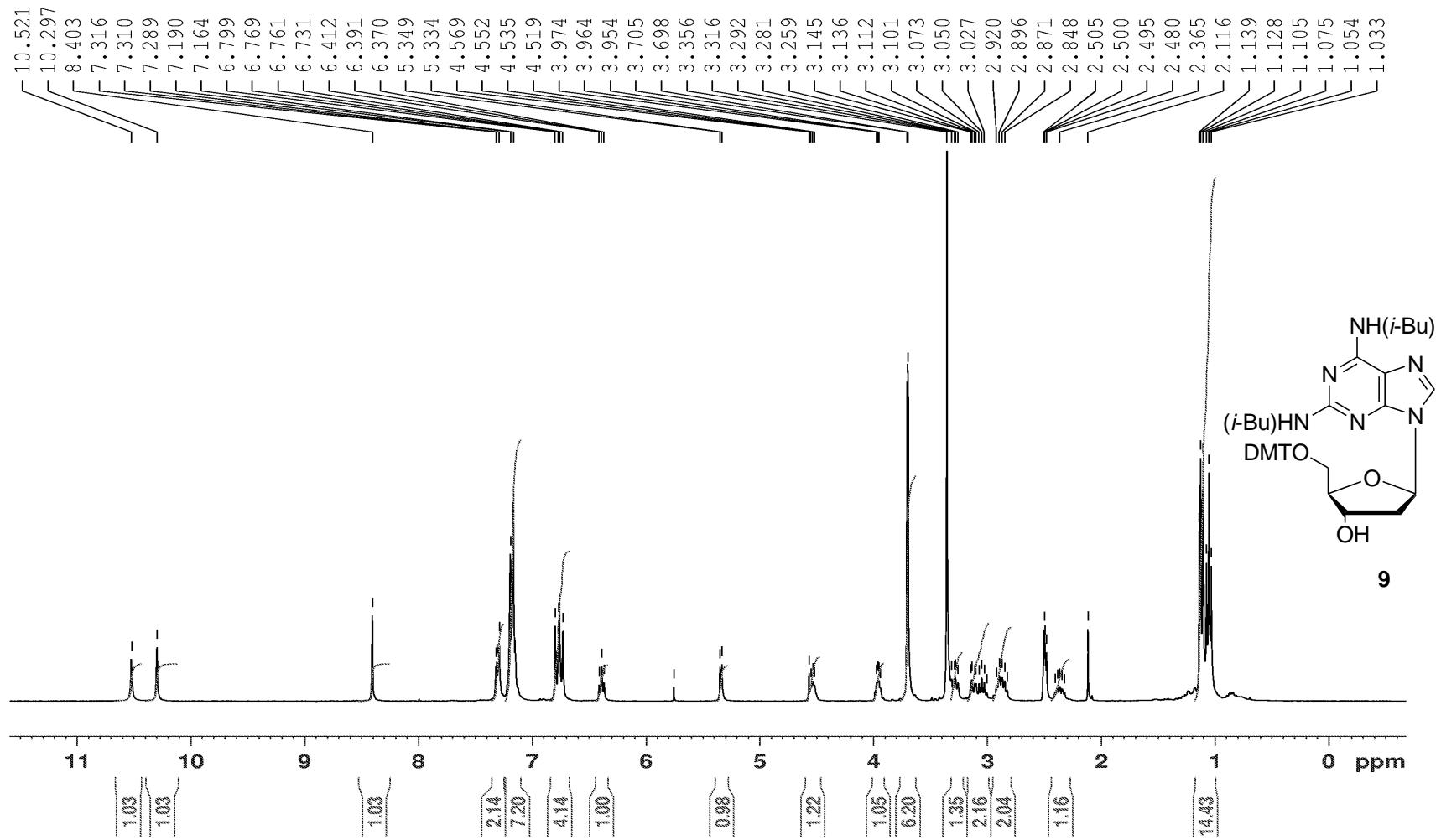
**Figure S14.** DEPT-135 spectrum of compound 8.



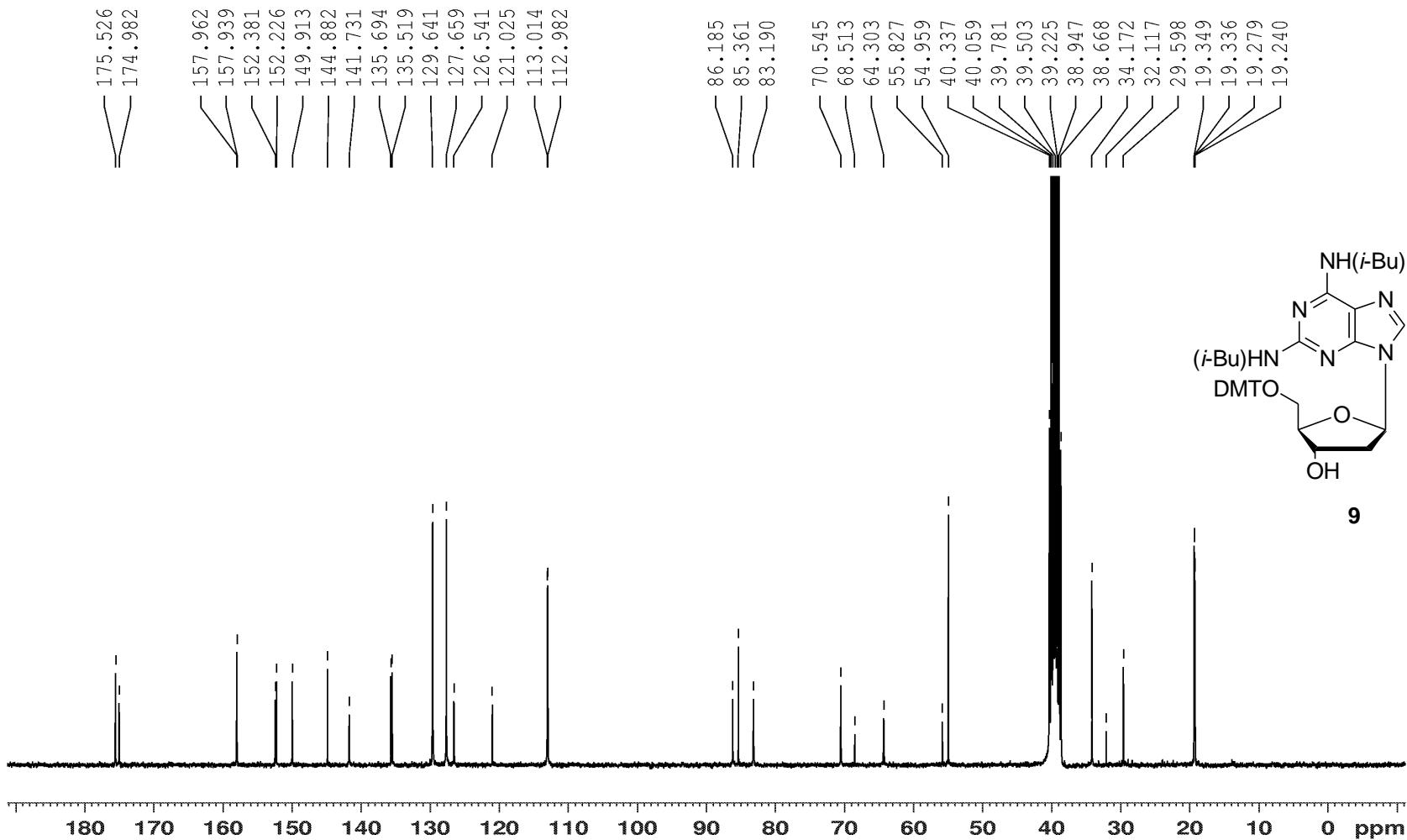
**Figure S15.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 8.



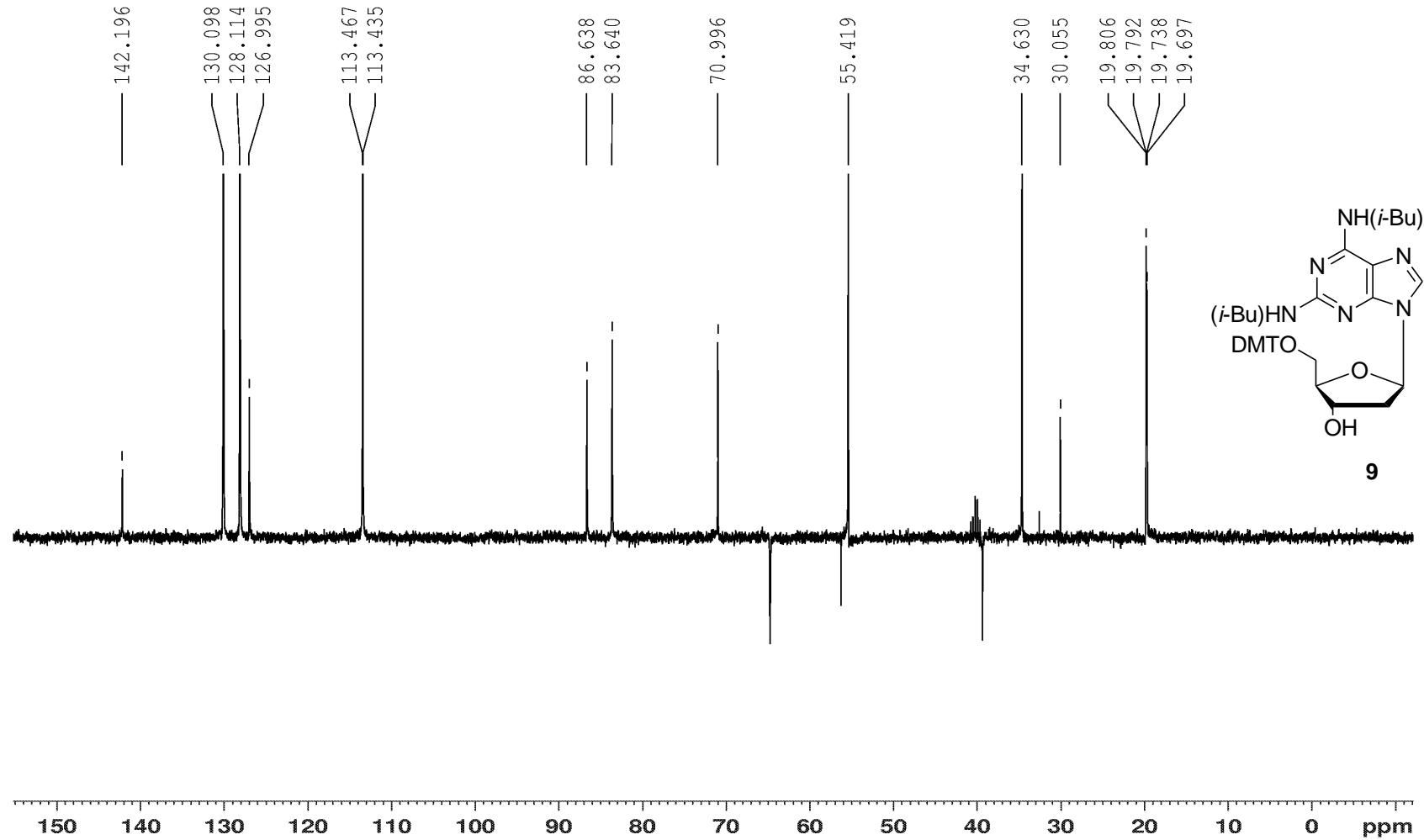
**Figure S16.**  $^1\text{H}$  NMR spectrum of compound 9.



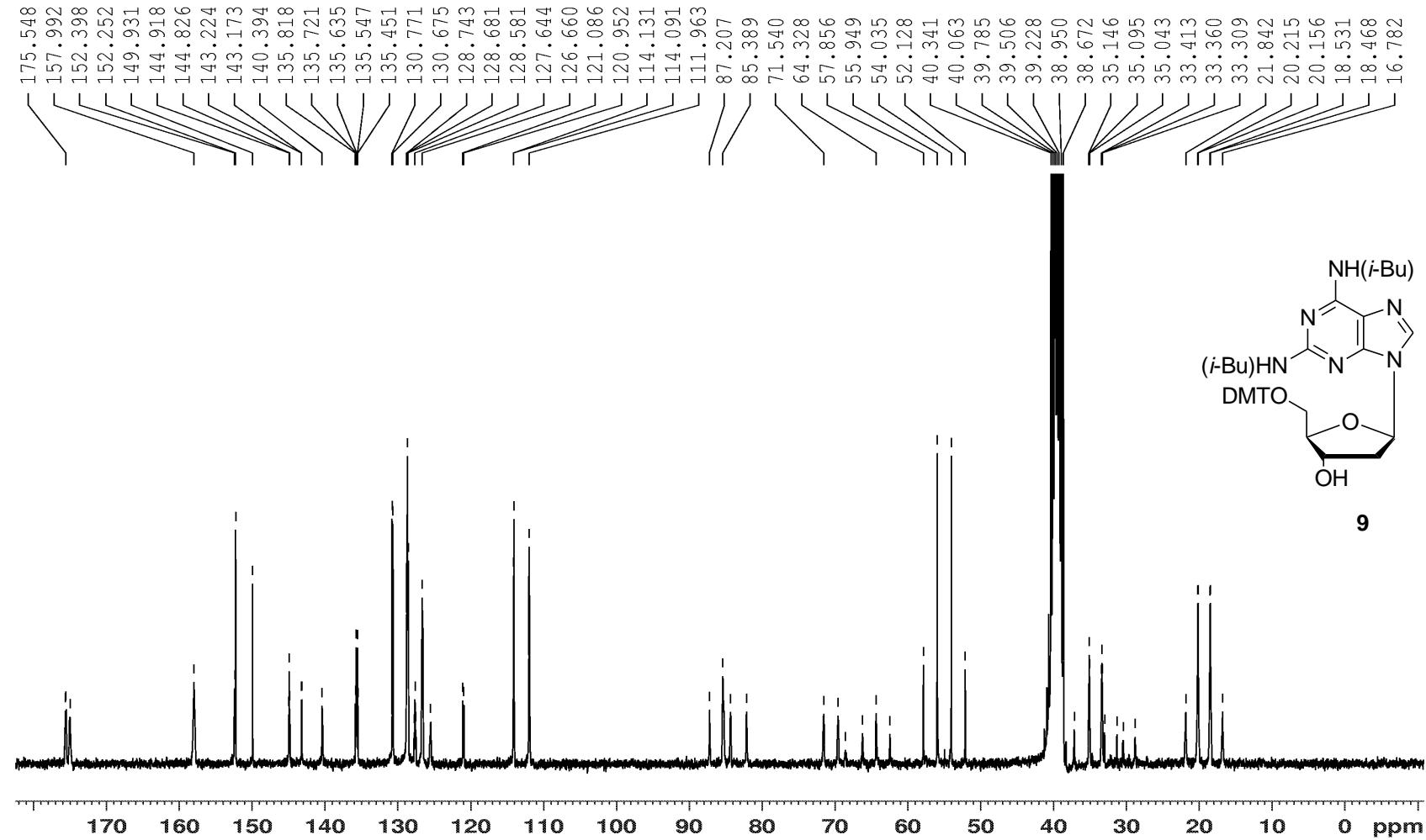
**Figure S17.**  $^{13}\text{C}$  NMR spectrum of compound 9.



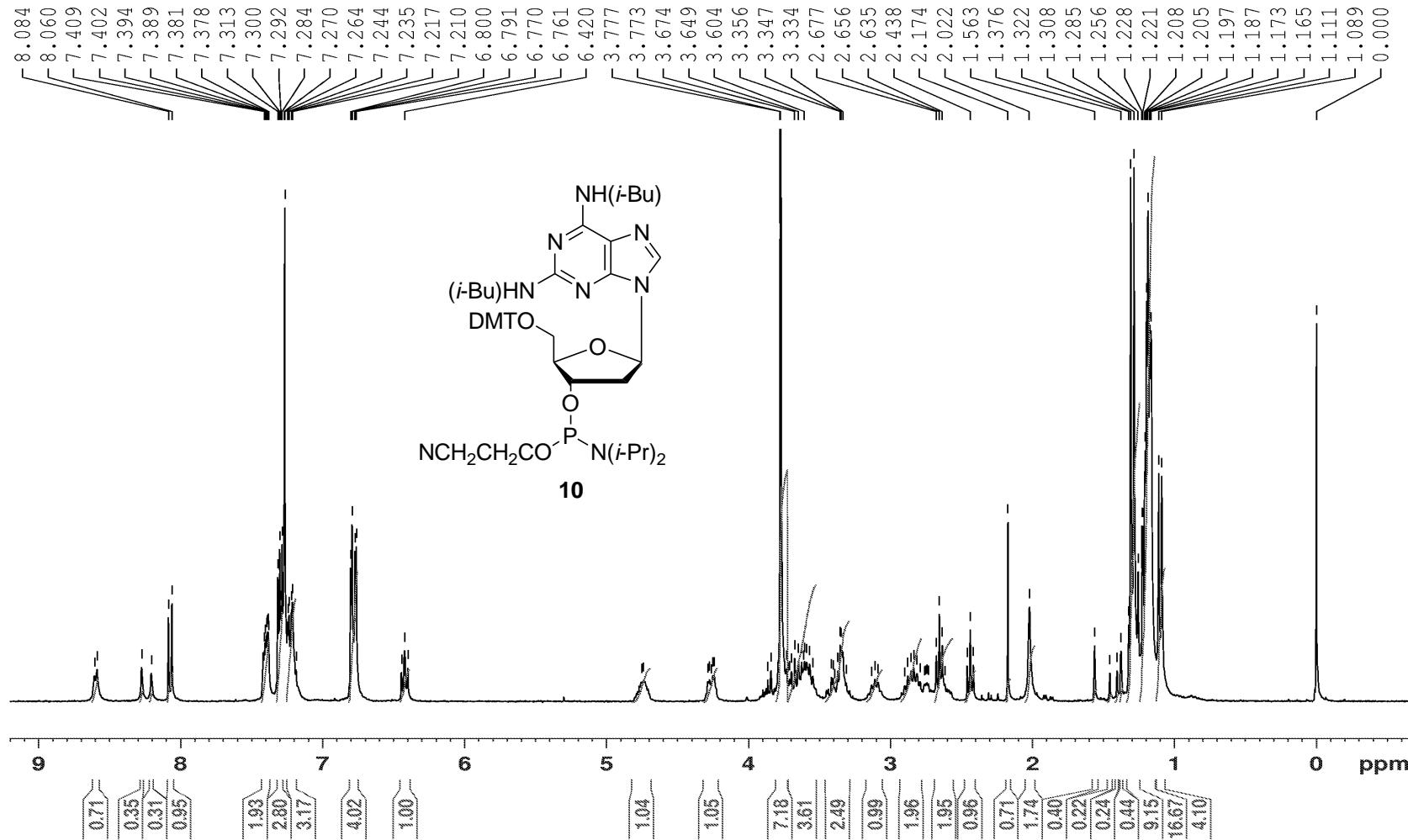
**Figure S18. DEPT-135 spectrum of compound 9.**



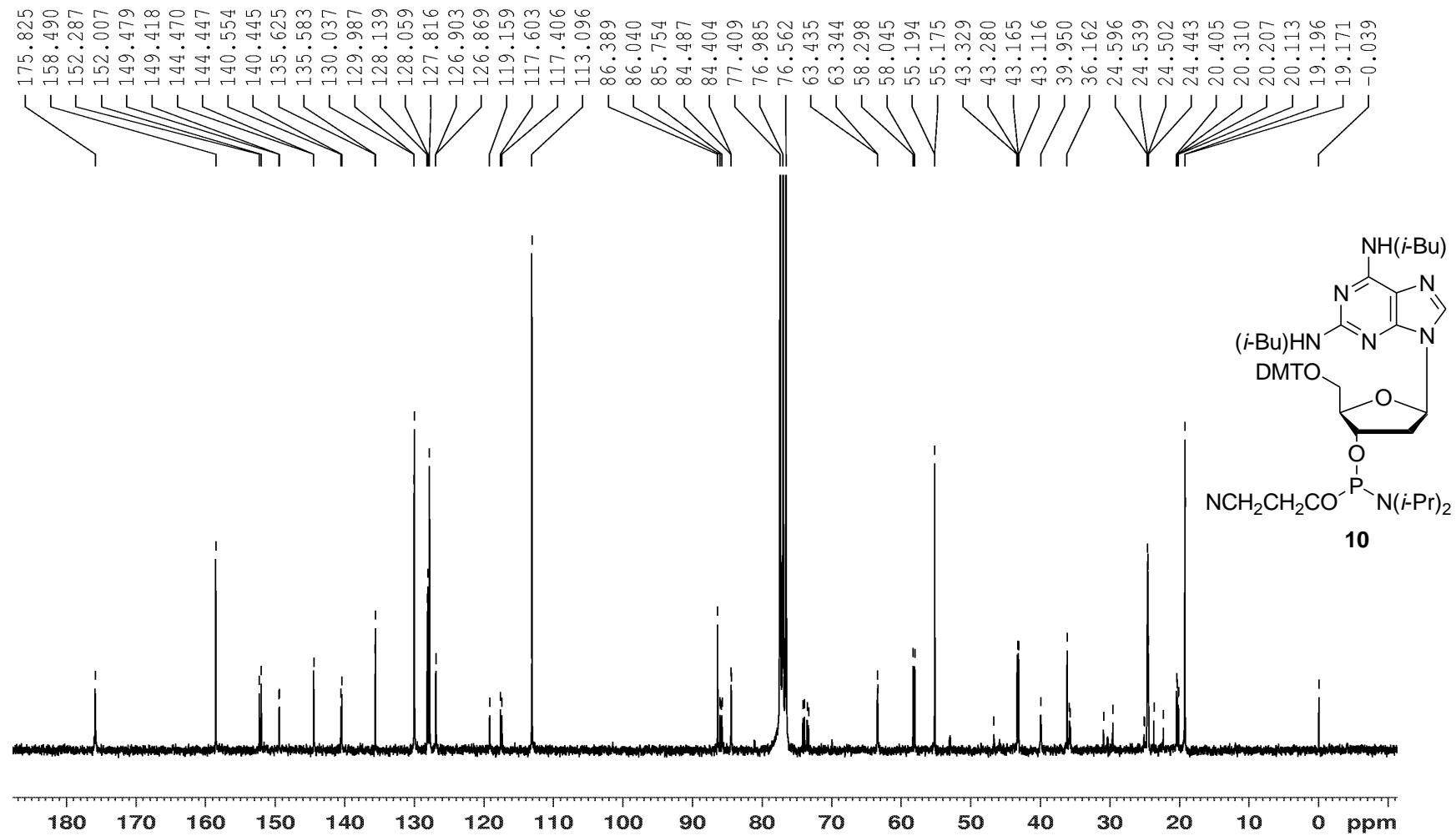
**Figure S19.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 9.



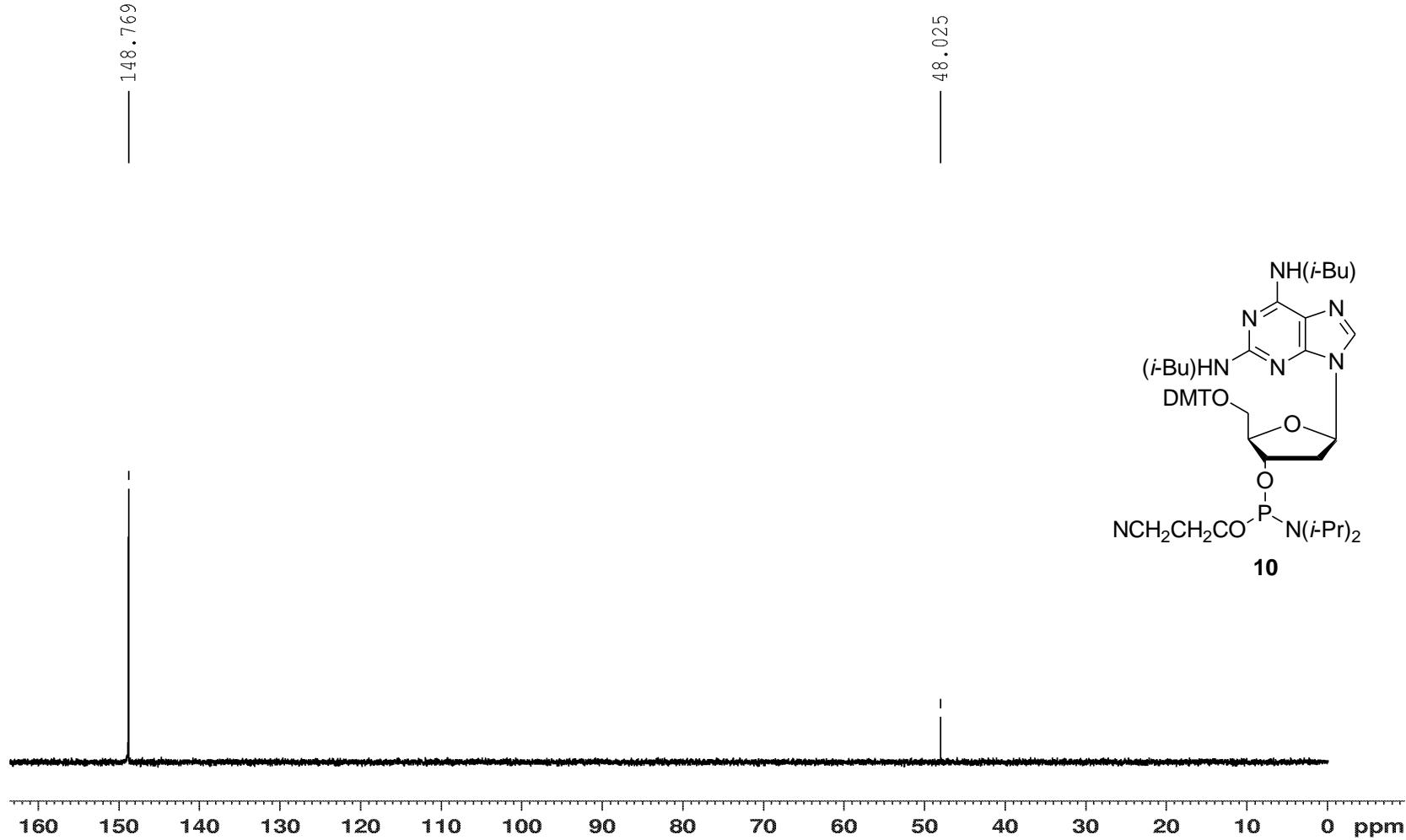
**Figure S20.**  $^1\text{H}$  NMR spectrum of compound 10.



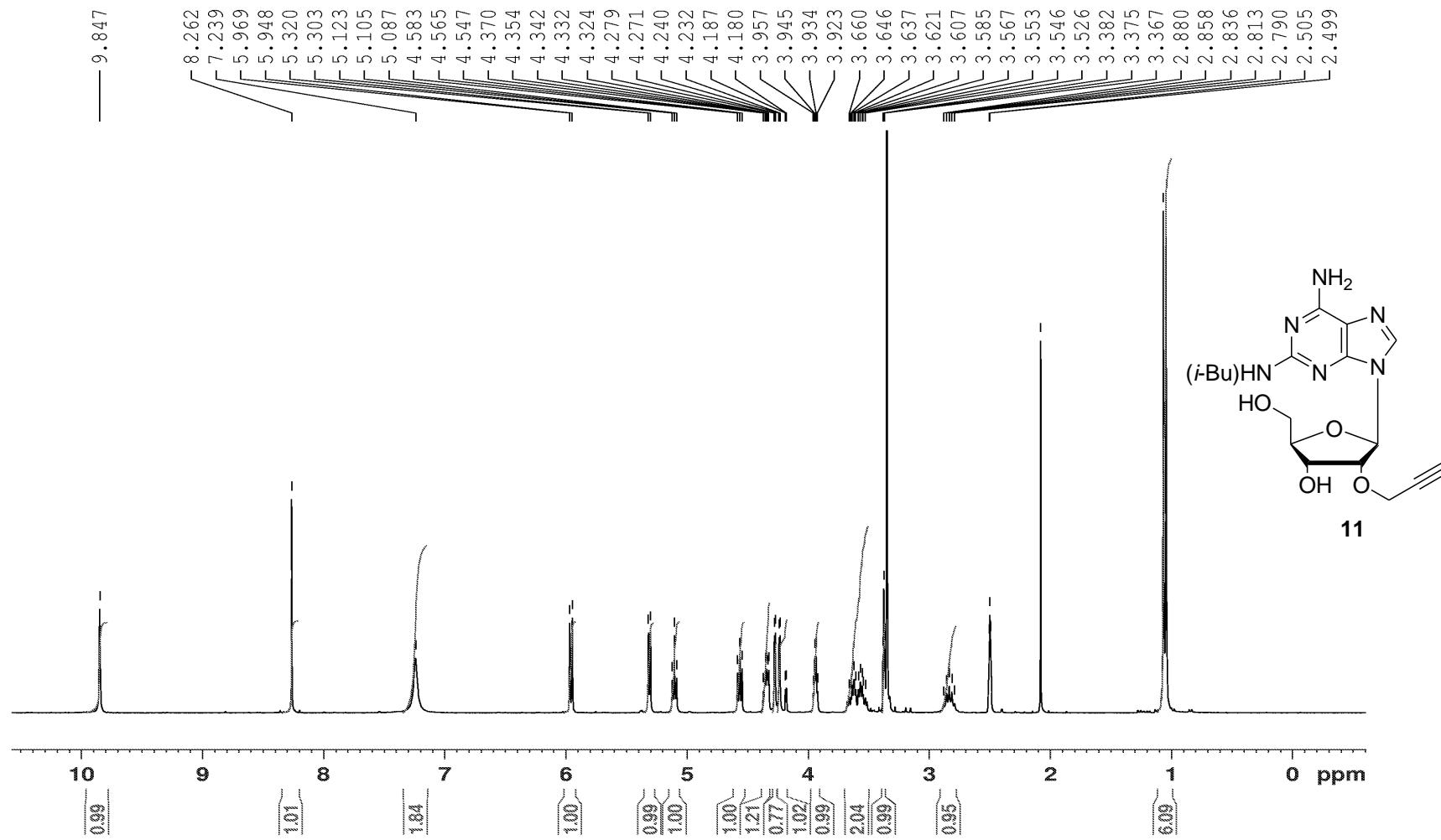
**Figure S21.**  $^{13}\text{C}$  NMR spectrum of compound **10**.



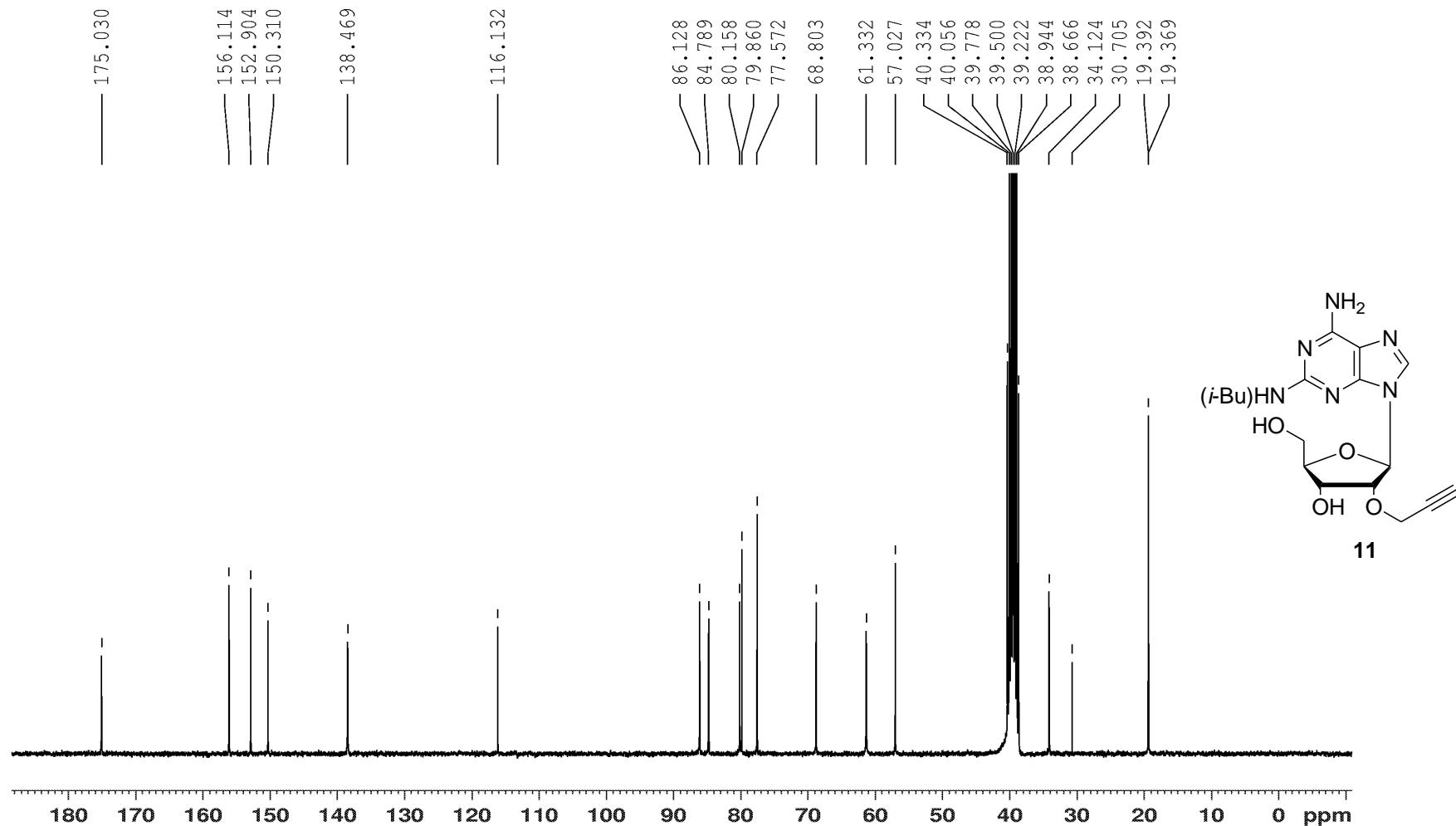
**Figure S22.**  $^{31}\text{P}$  NMR spectrum of compound **10**.



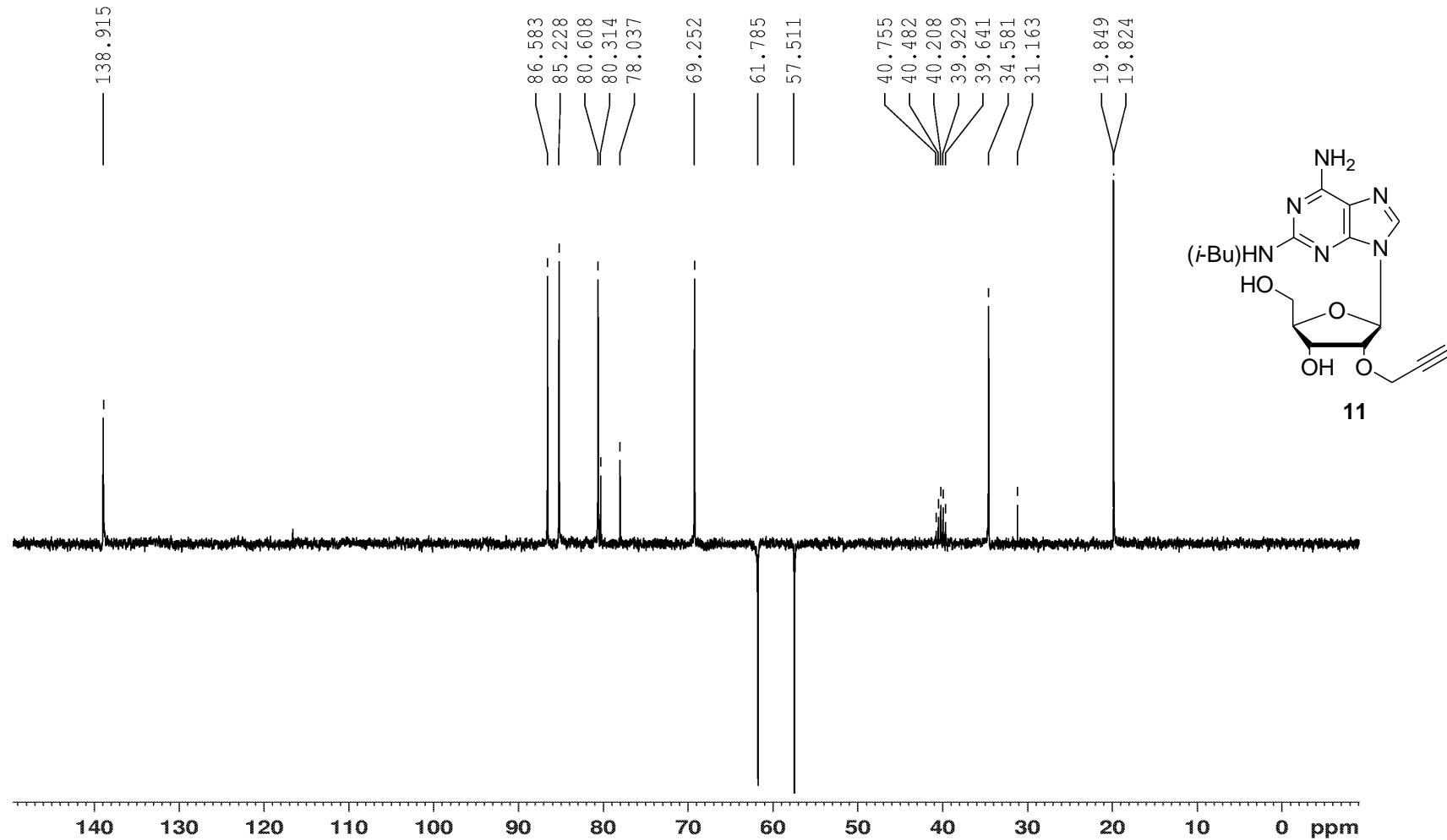
**Figure S23.**  $^1\text{H}$  NMR spectrum of compound 11.



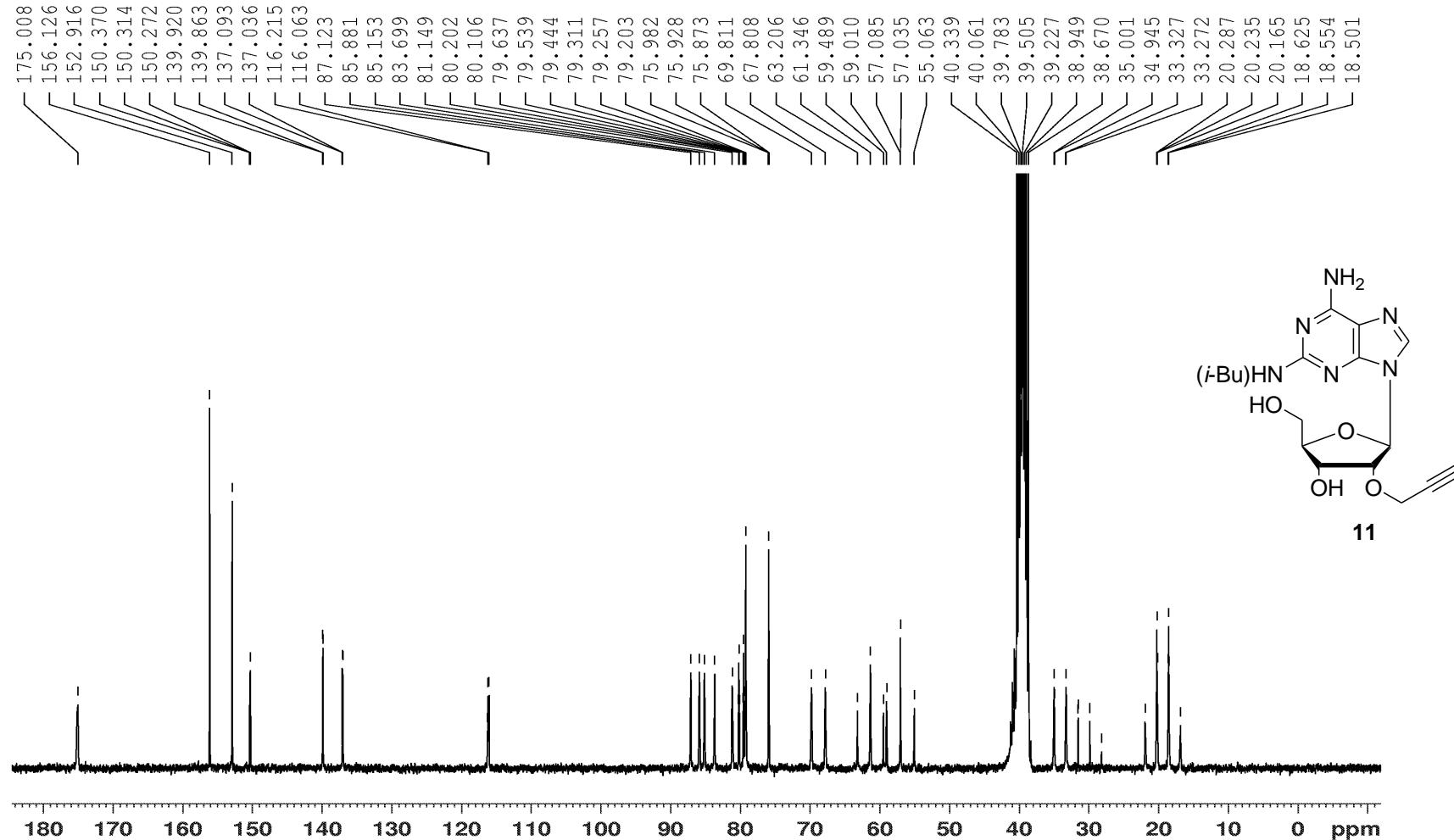
**Figure S24.**  $^{13}\text{C}$  NMR spectrum of compound 11.



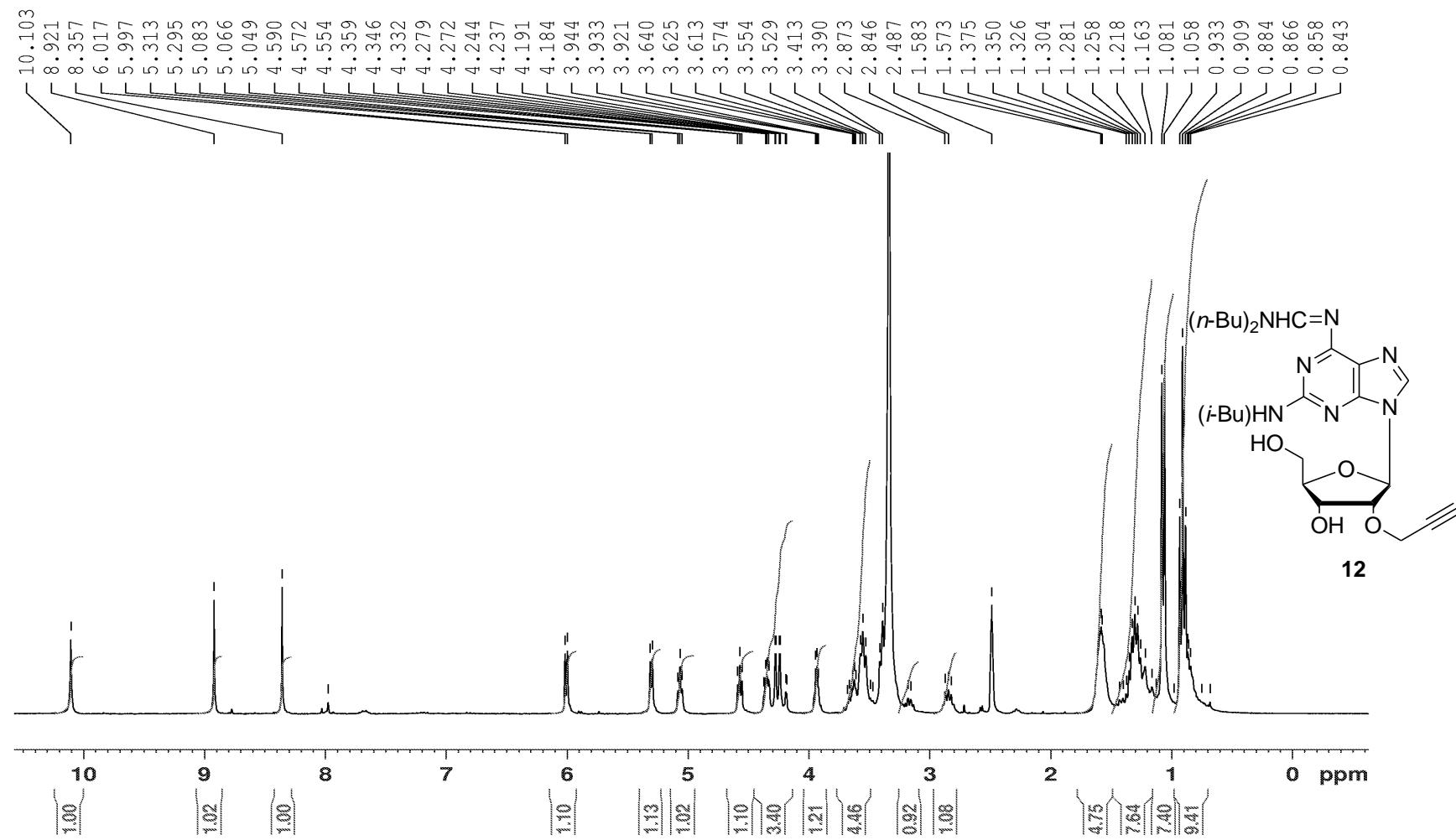
**Figure S25.** DEPT-135 spectrum of compound 11.



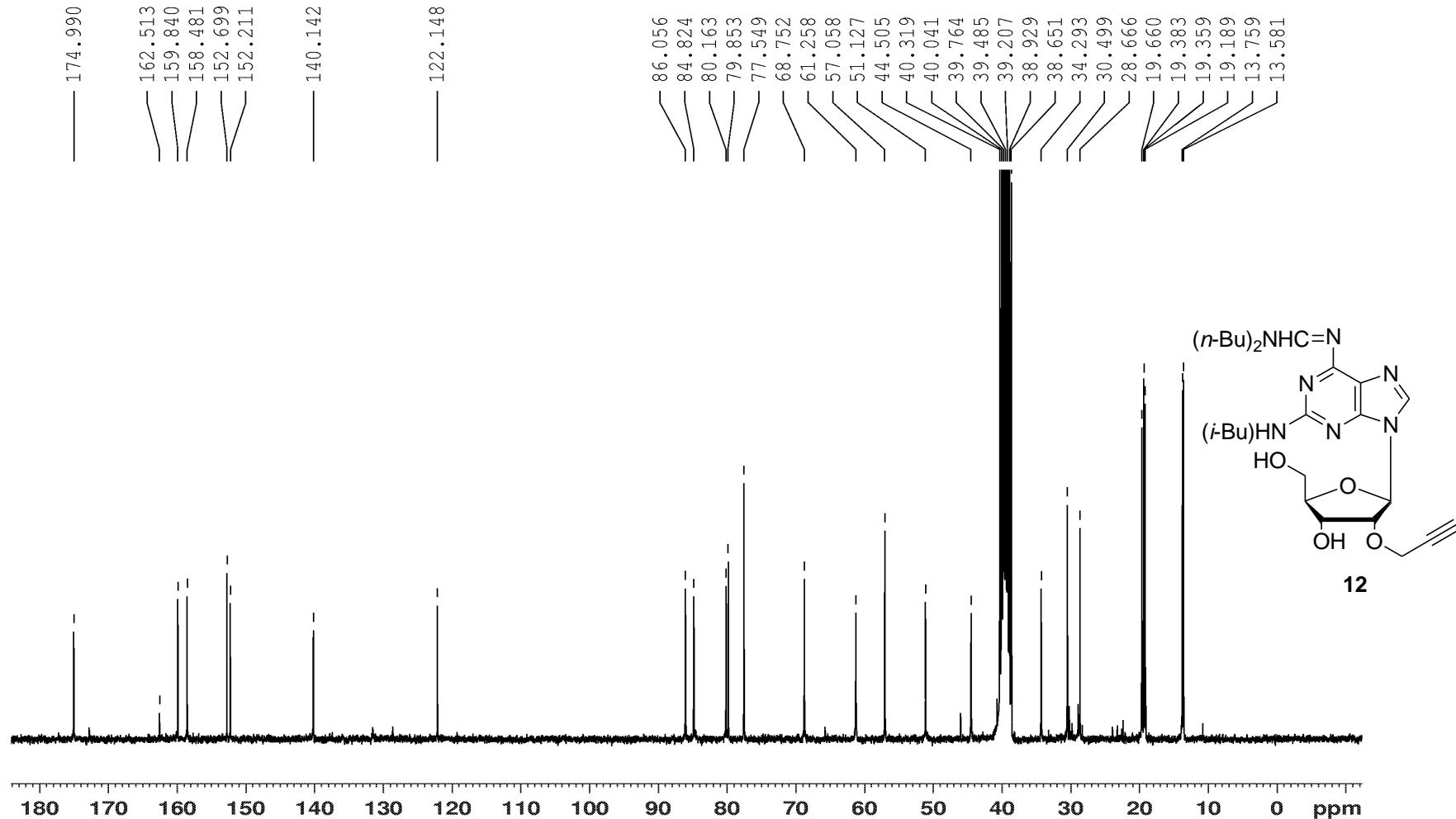
**Figure S26.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 11.



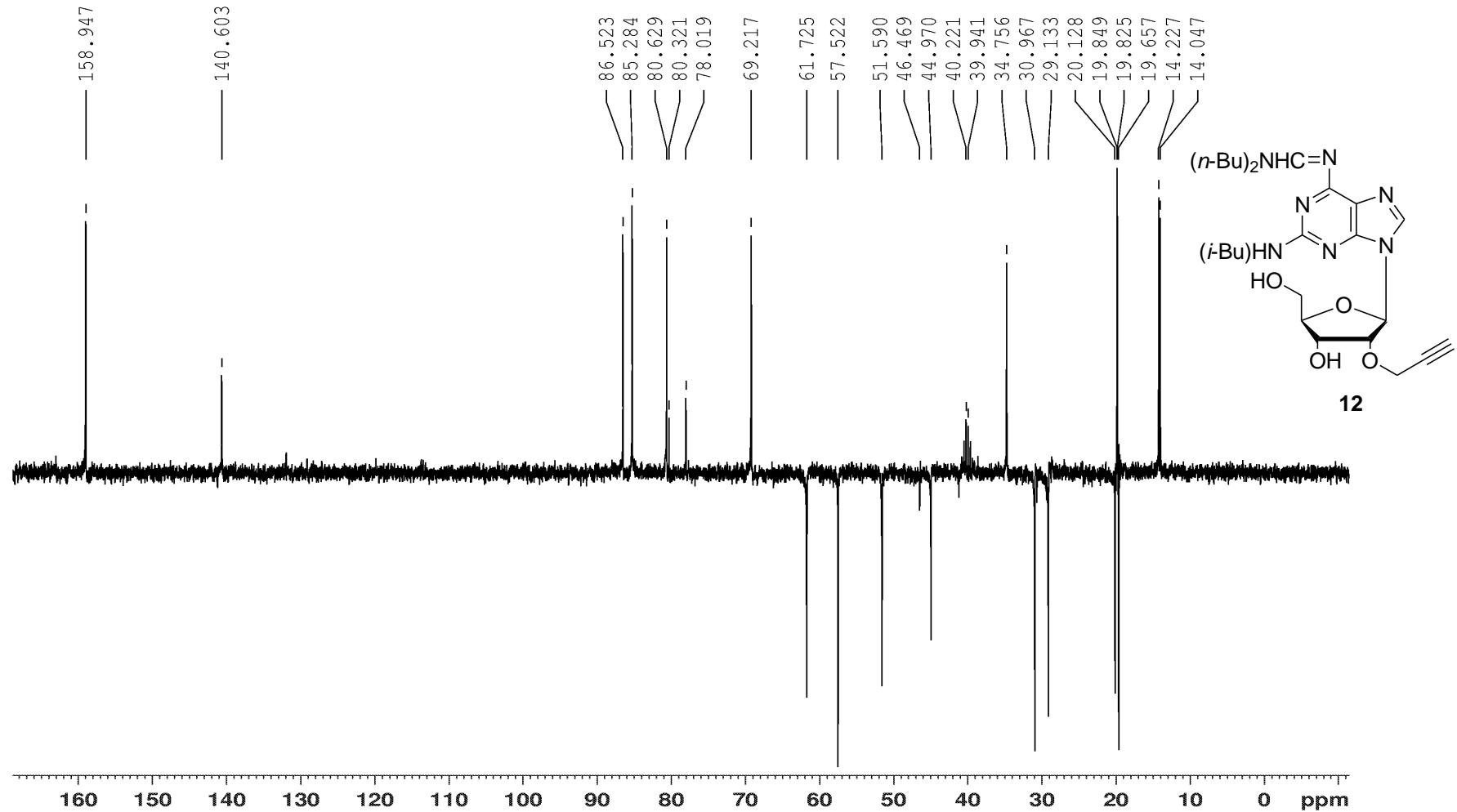
**Figure S27.**  $^1\text{H}$  NMR spectrum of compound 12.



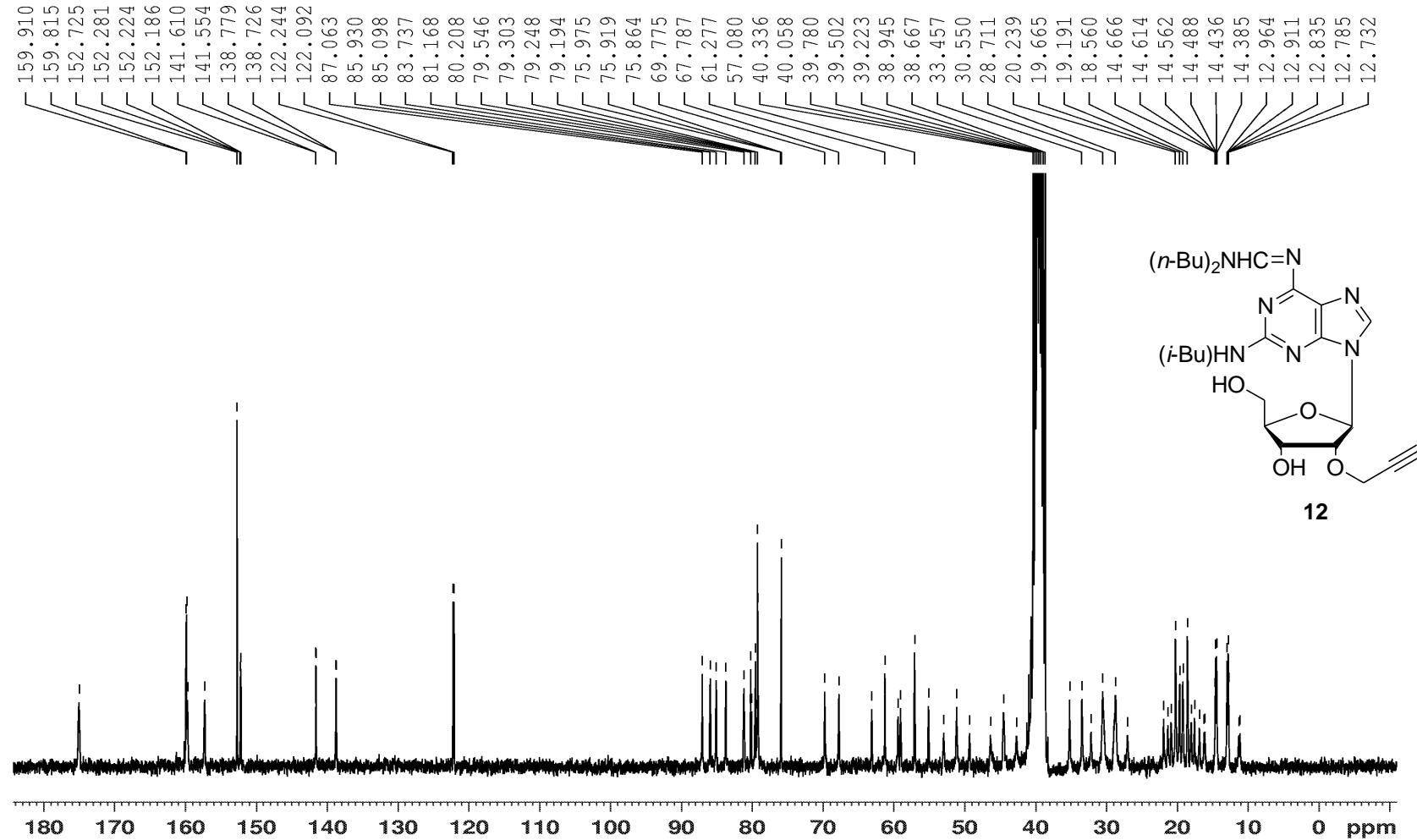
**Figure S28.**  $^{13}\text{C}$  NMR spectrum of compound 12.



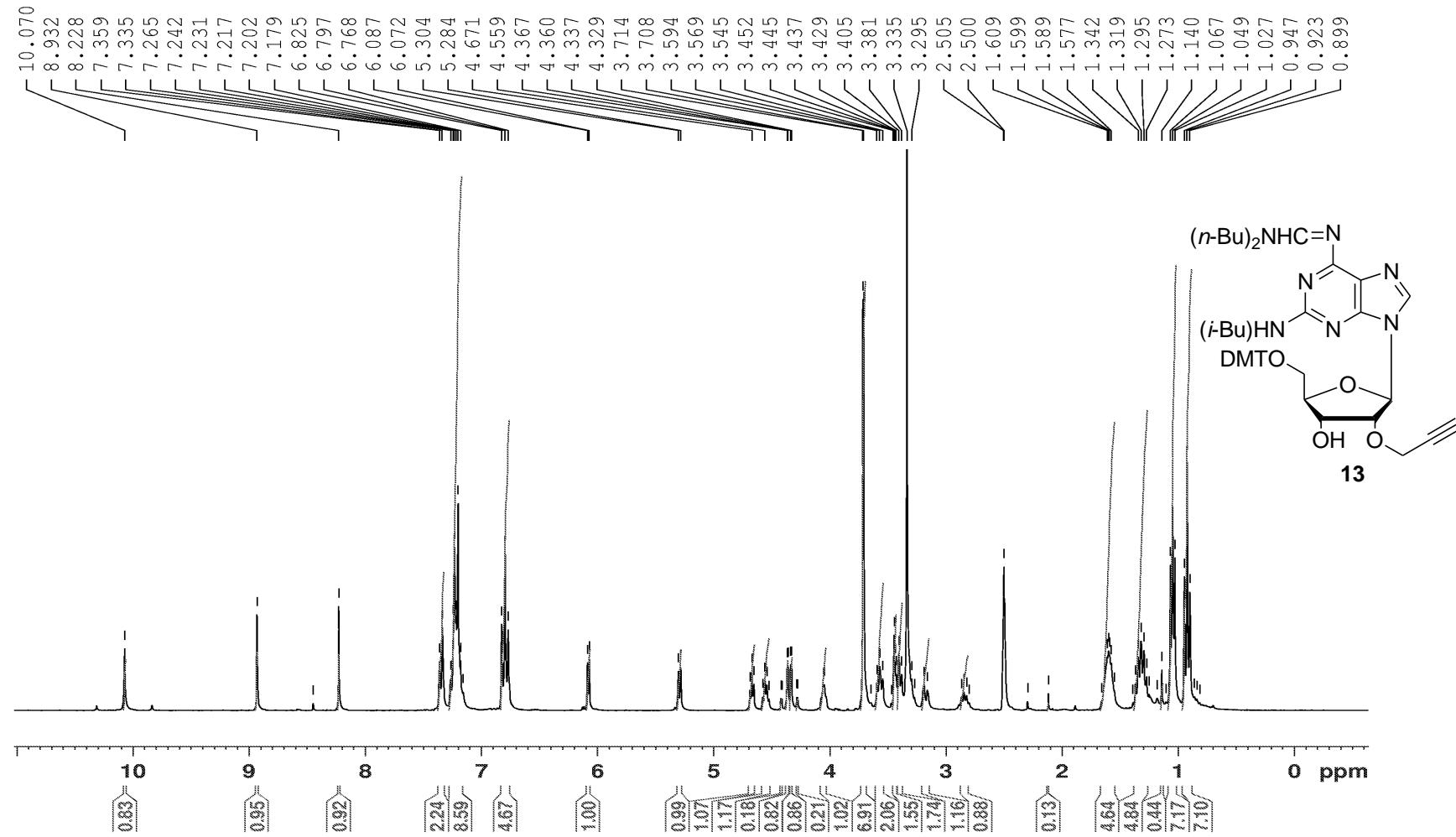
**Figure S29. DEPT-135 spectrum of compound 12.**



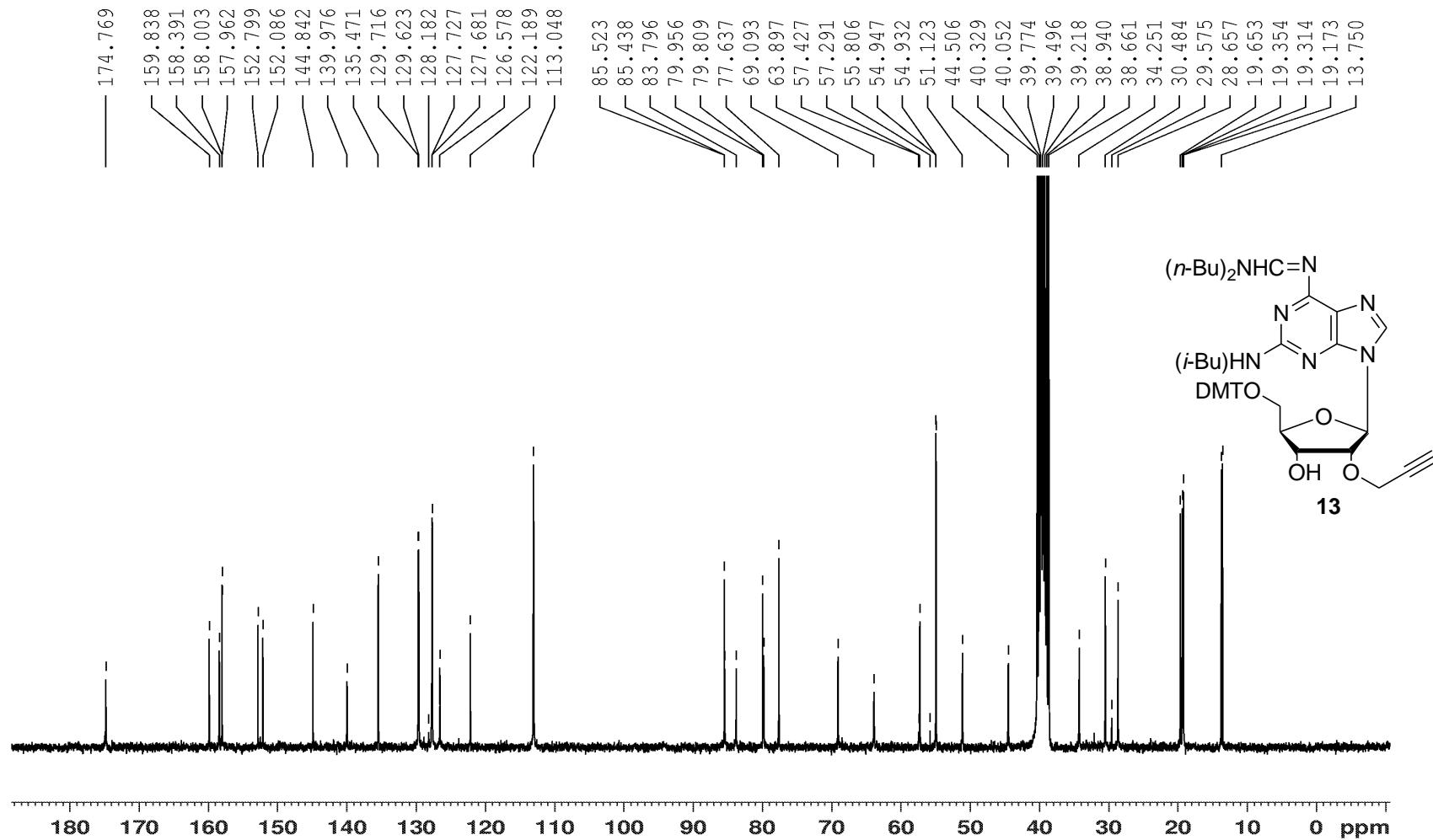
**Figure S30.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 12.



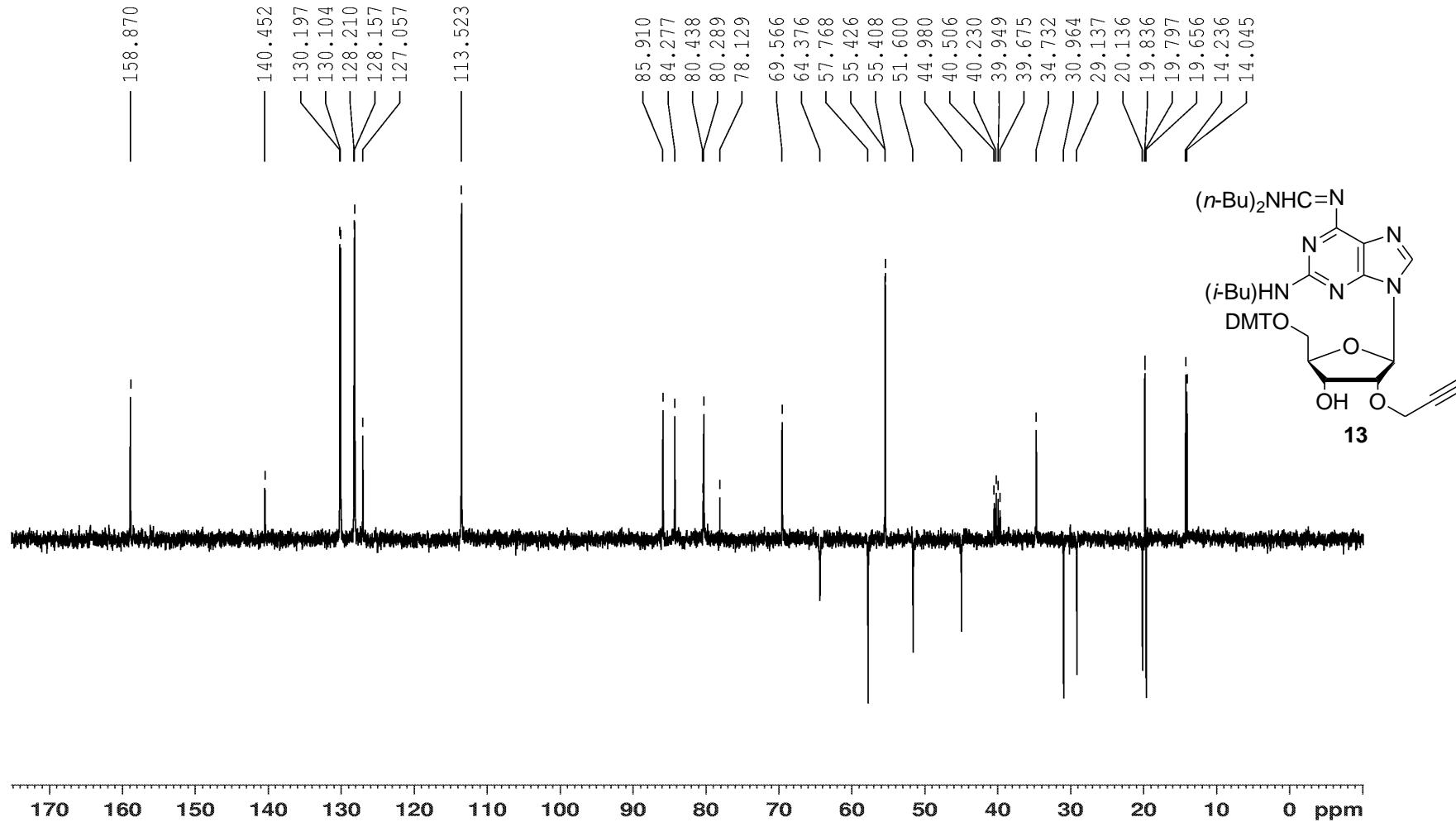
**Figure S31.**  $^1\text{H}$  NMR spectrum of compound 13.



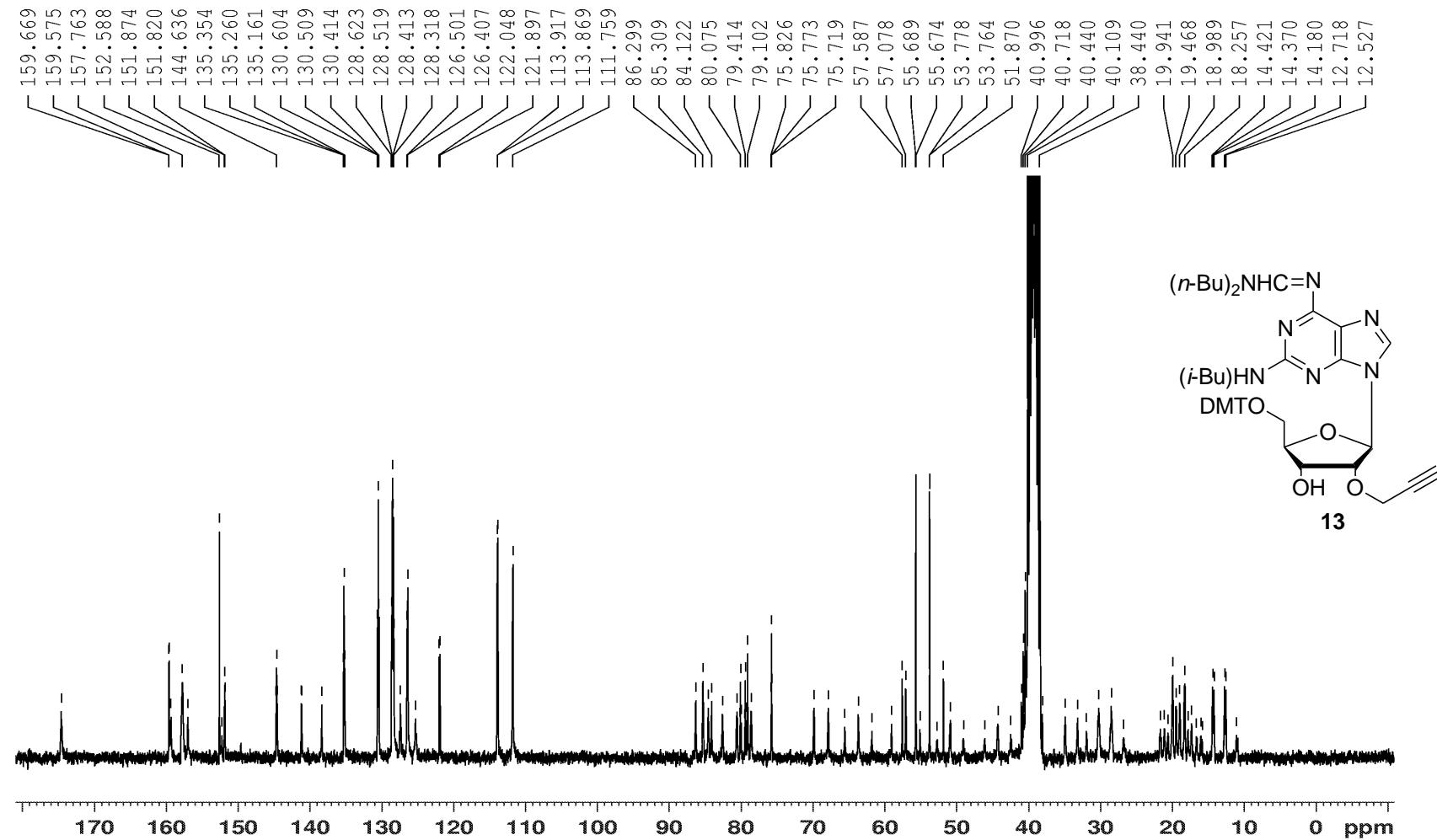
**Figure S32.**  $^{13}\text{C}$  NMR spectrum of compound 13.



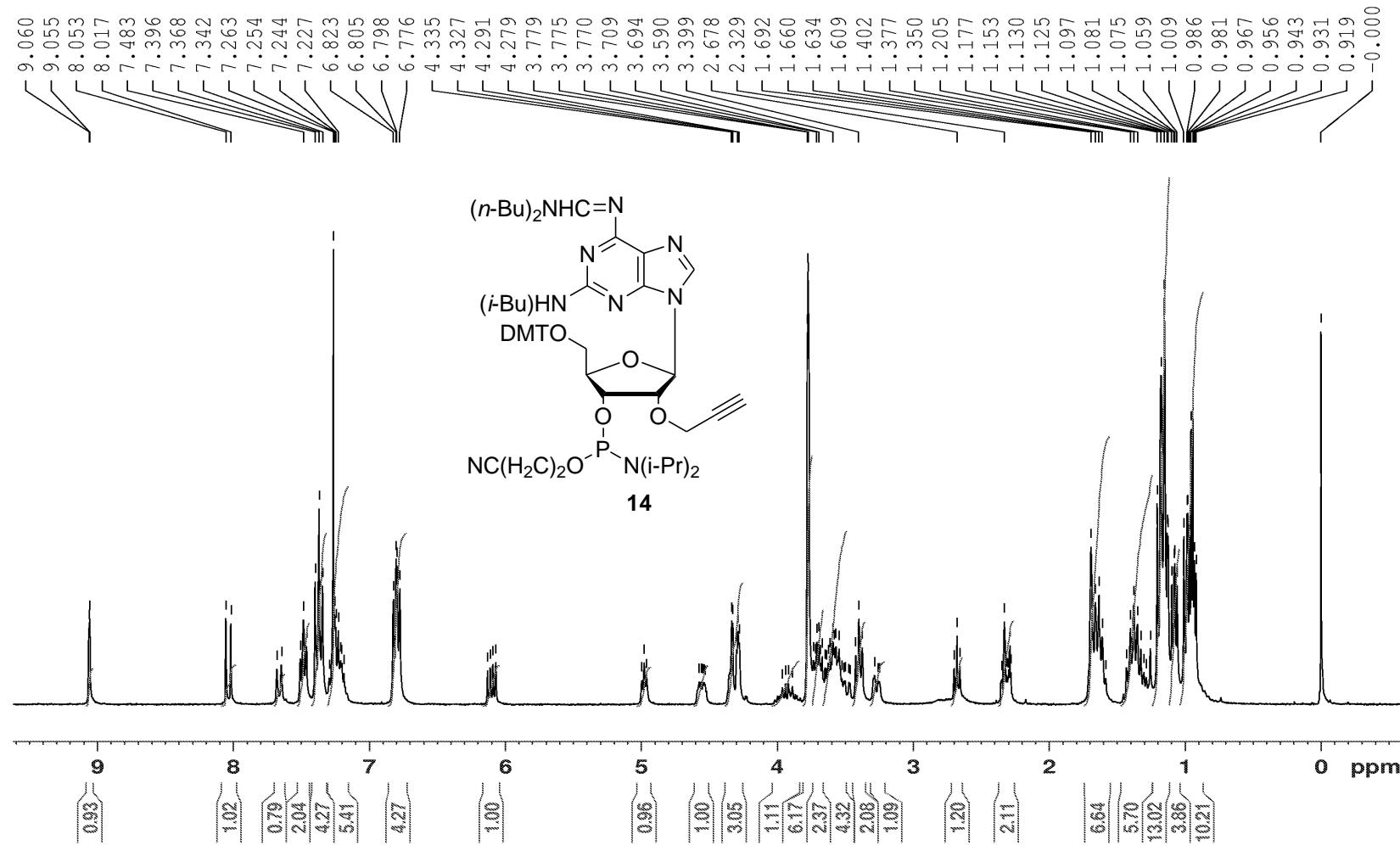
**Figure S33. DEPT-135 spectrum of compound 13.**



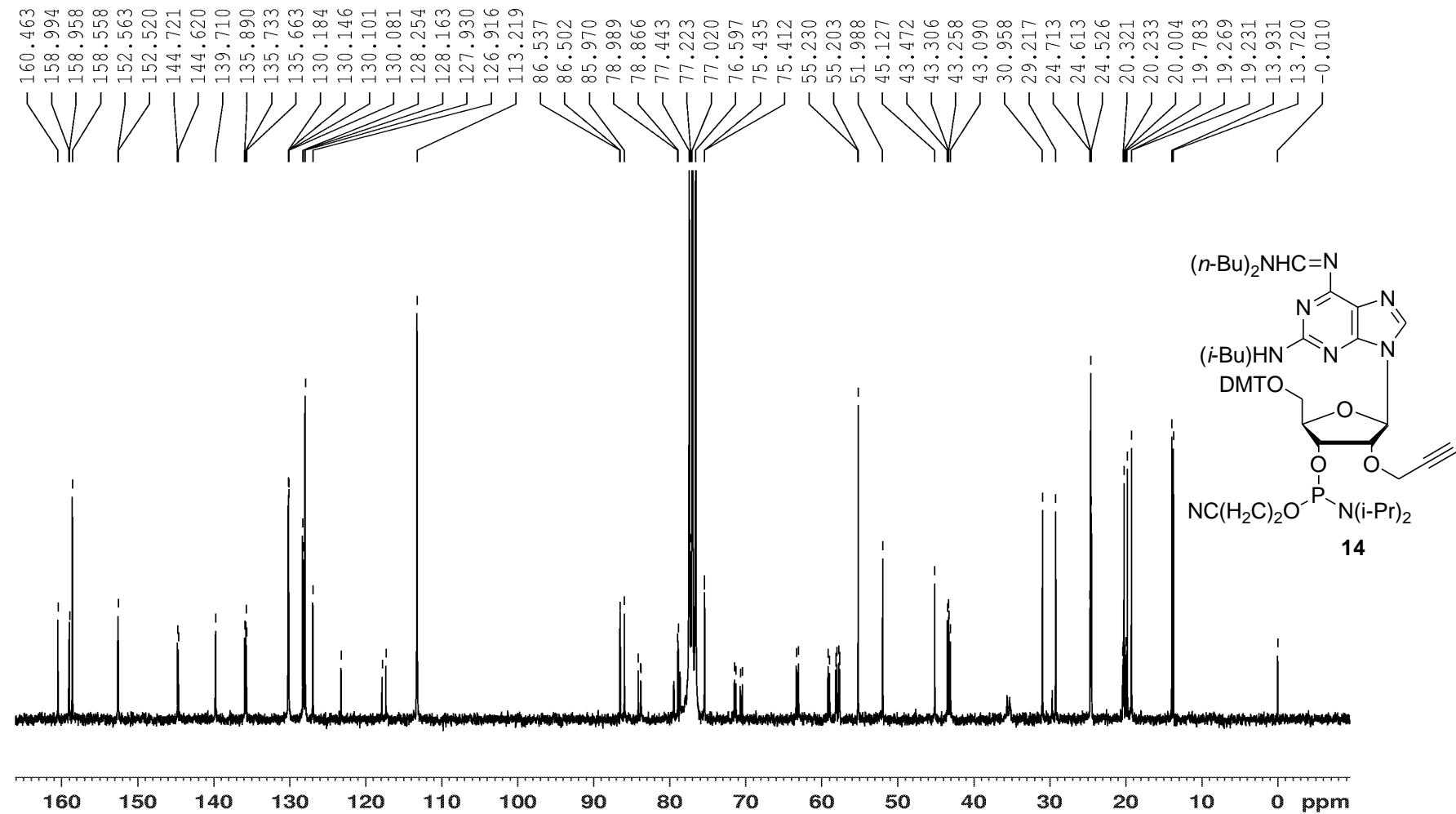
**Figure S34.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 13.



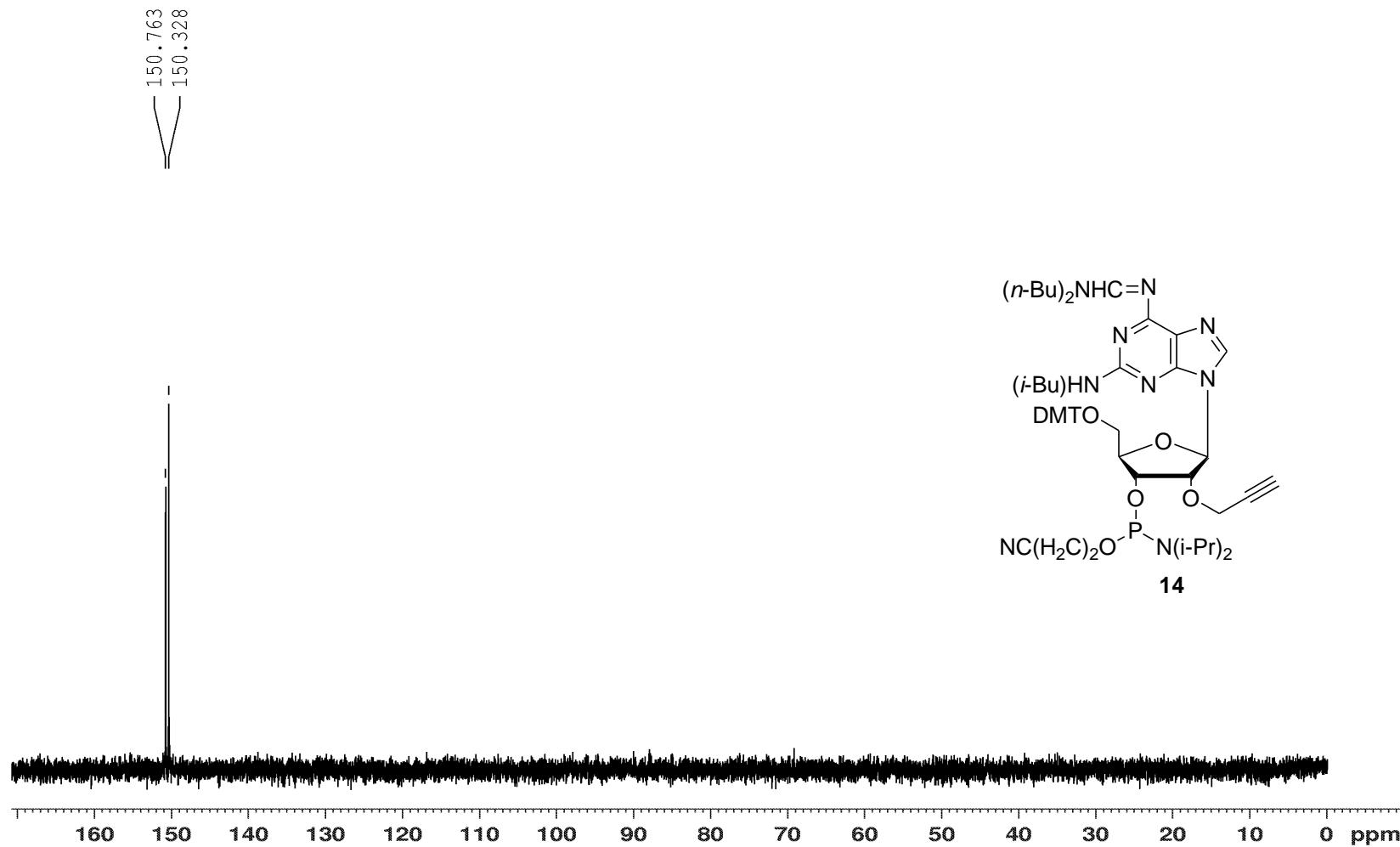
**Figure S35.**  $^1\text{H}$  NMR spectrum of compound 14.



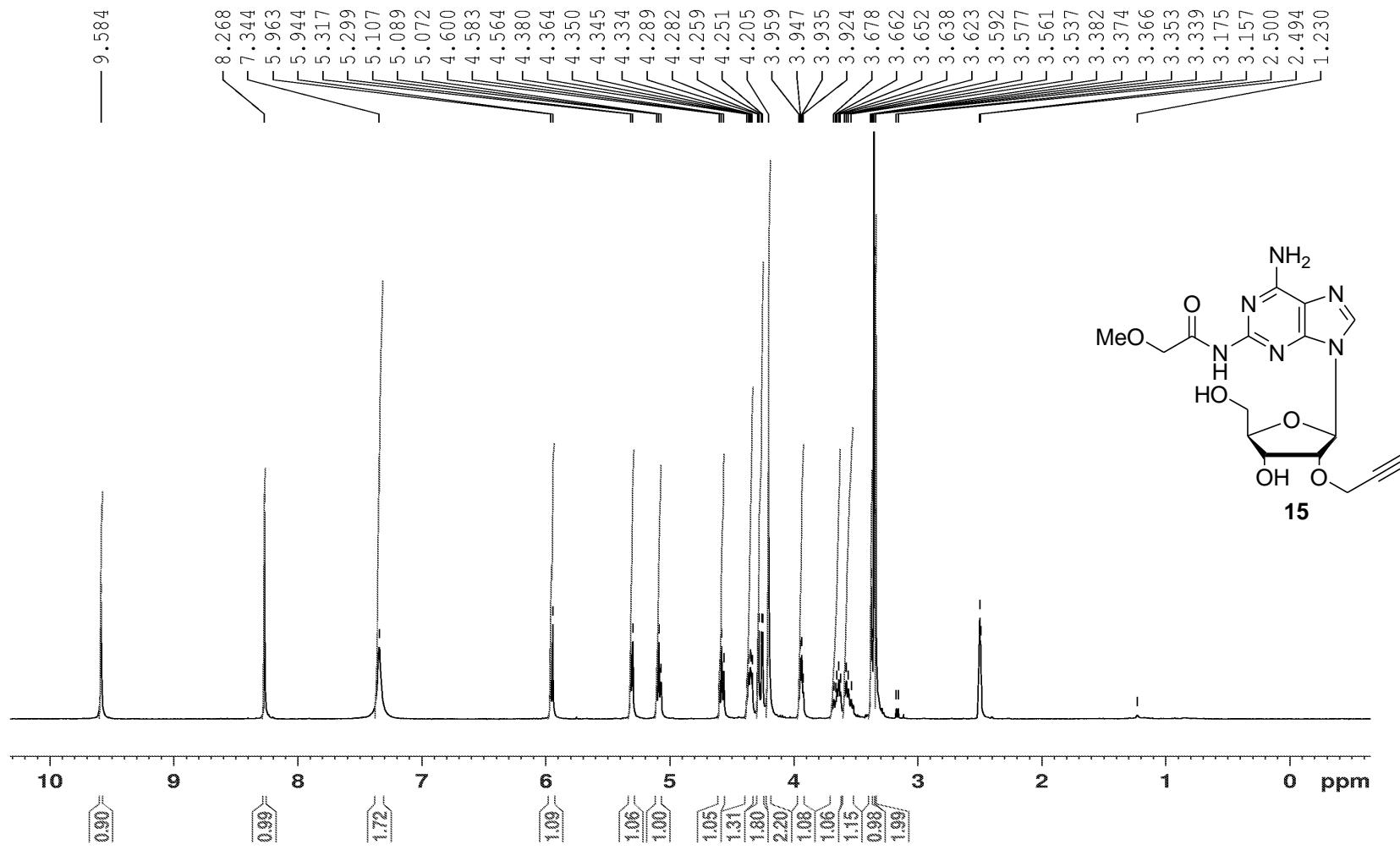
**Figure S36.**  $^{13}\text{C}$  NMR spectrum of compound **14**.



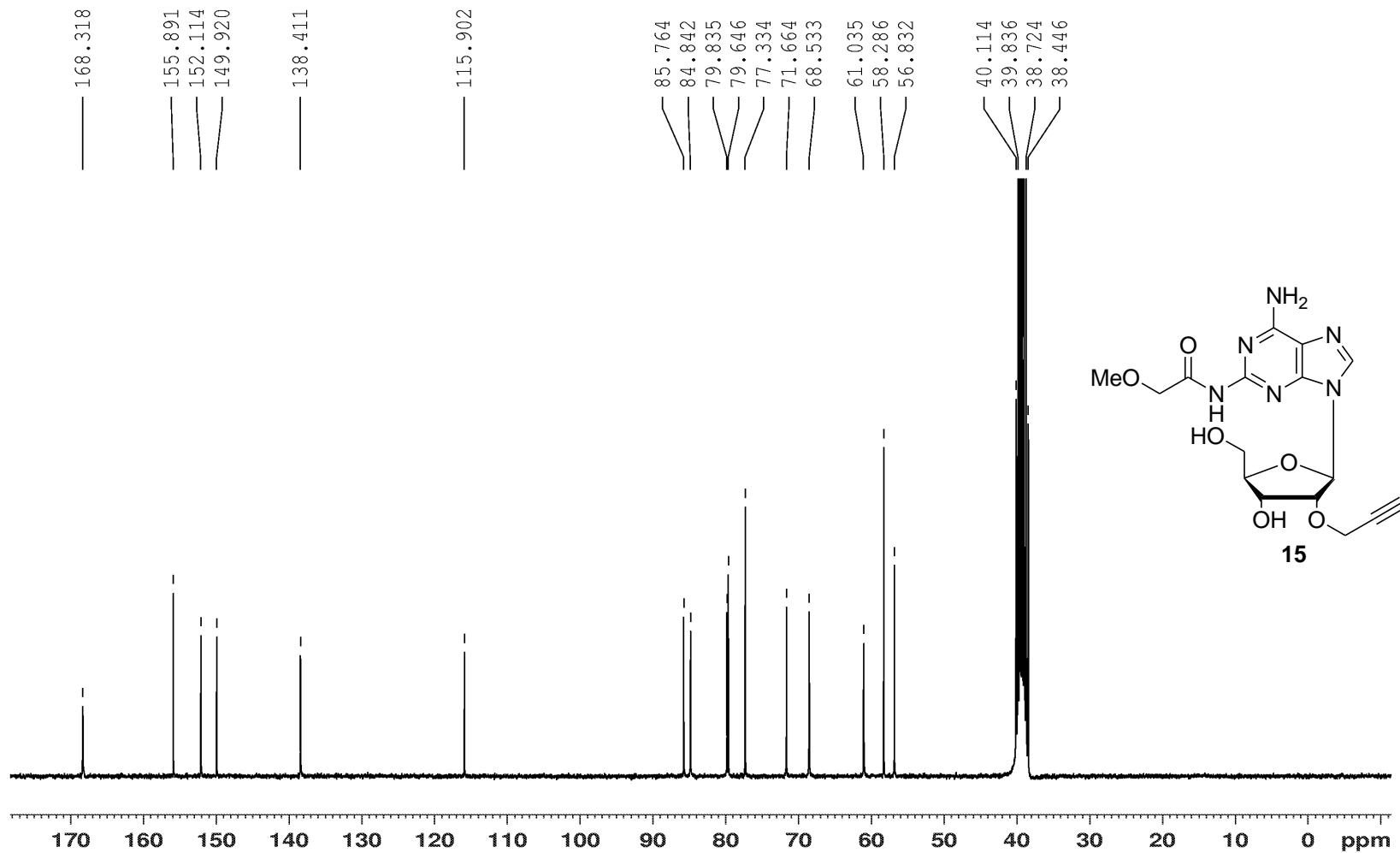
**Figure S37.**  $^{31}\text{P}$  NMR spectrum of compound 14.



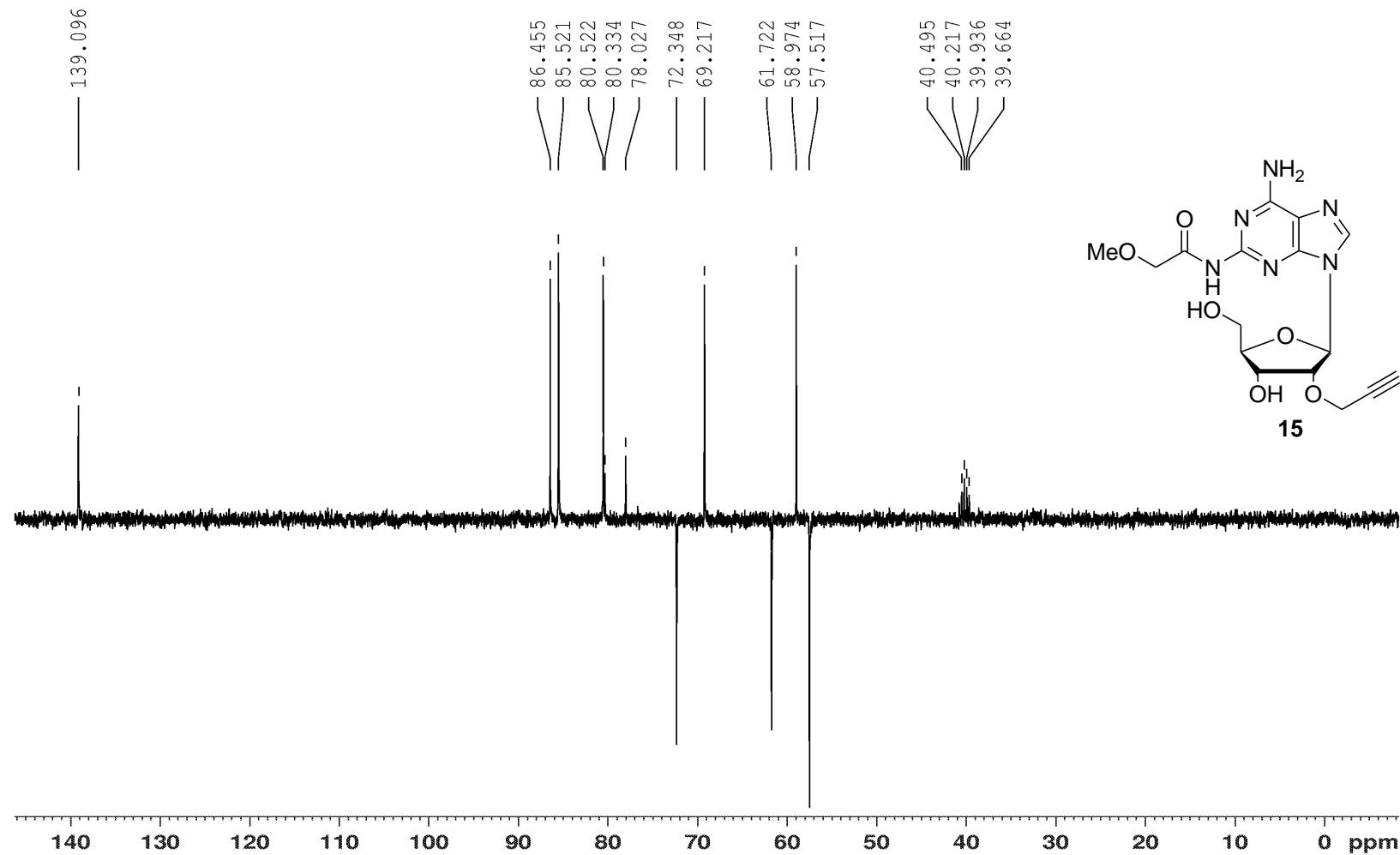
**Figure S38.**  $^1\text{H}$  NMR spectrum of compound 15.



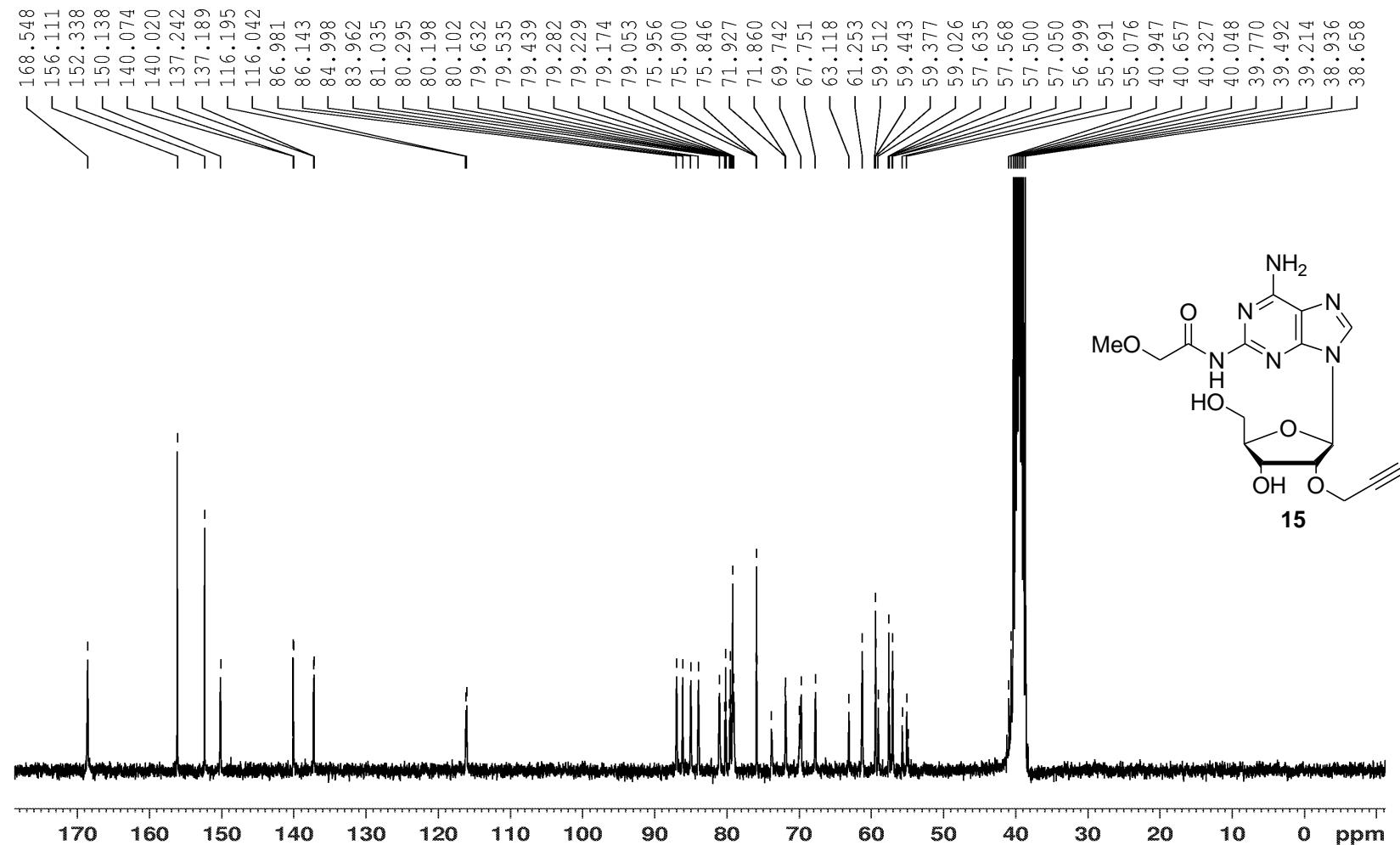
**Figure S39.**  $^{13}\text{C}$  NMR spectrum of compound 15.



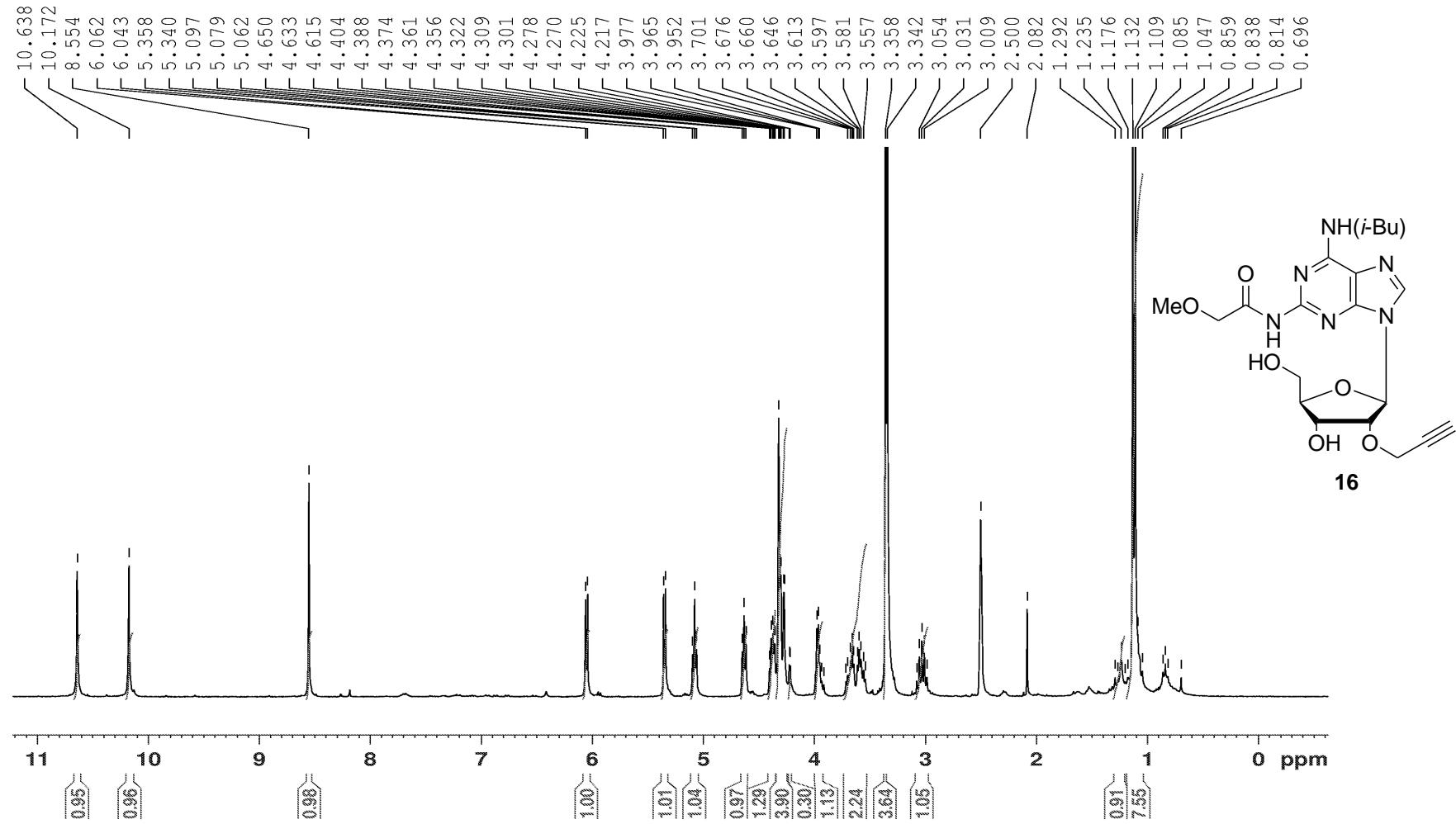
**Figure S40.** DEPT-135 spectrum of compound 15.



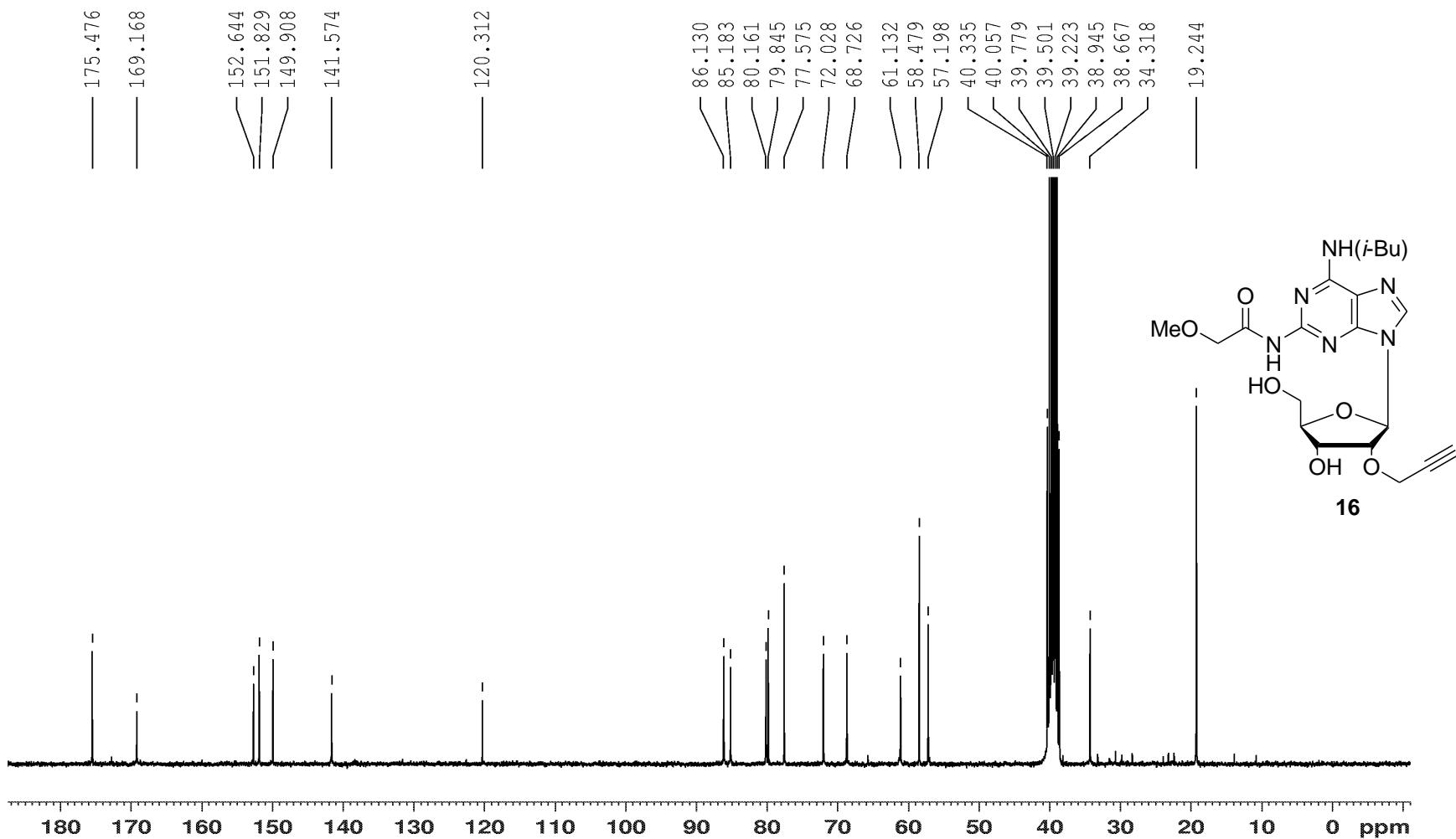
**Figure S41.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 15.



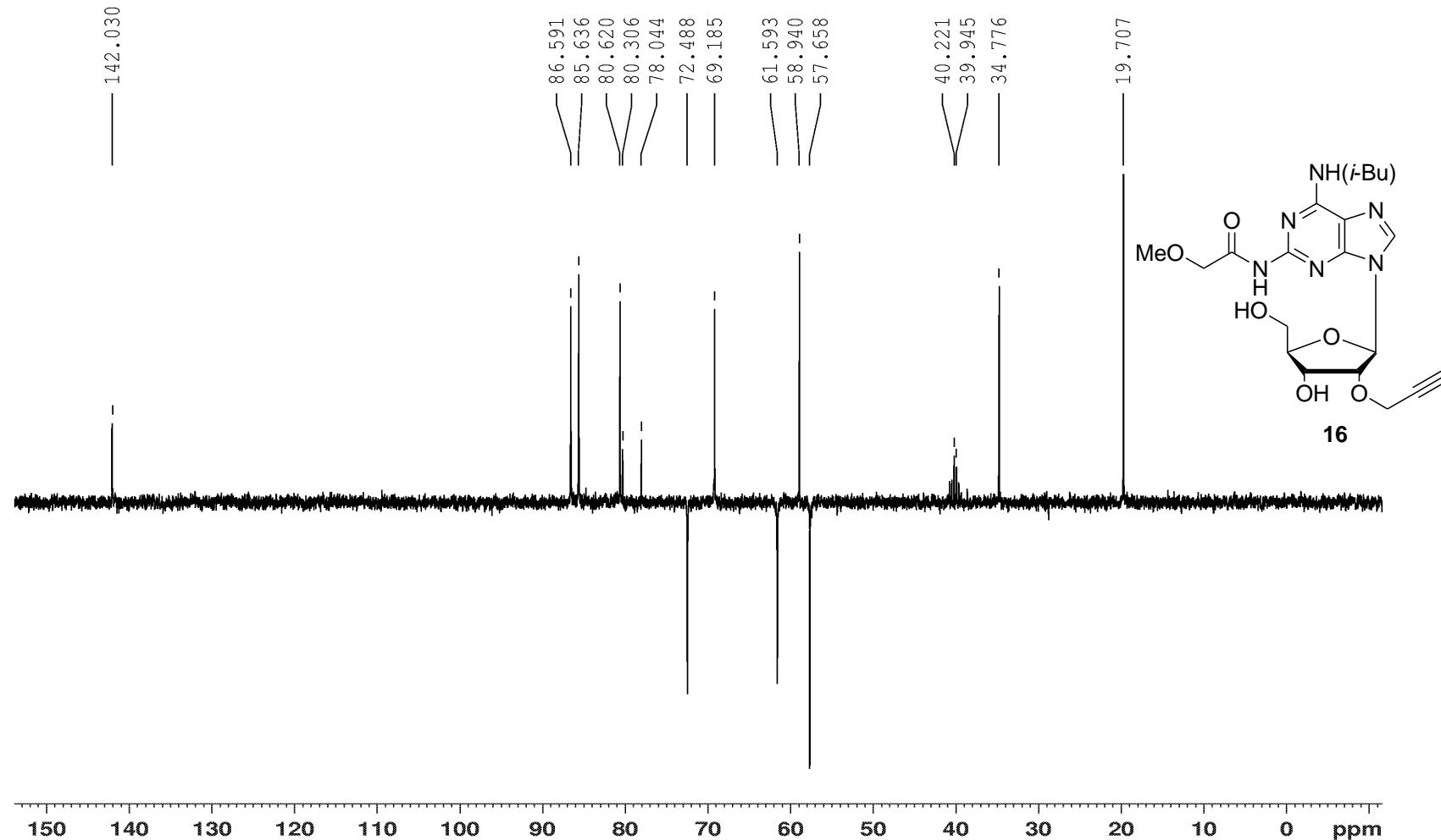
**Figure S42.**  $^1\text{H}$  NMR spectrum of compound 16.



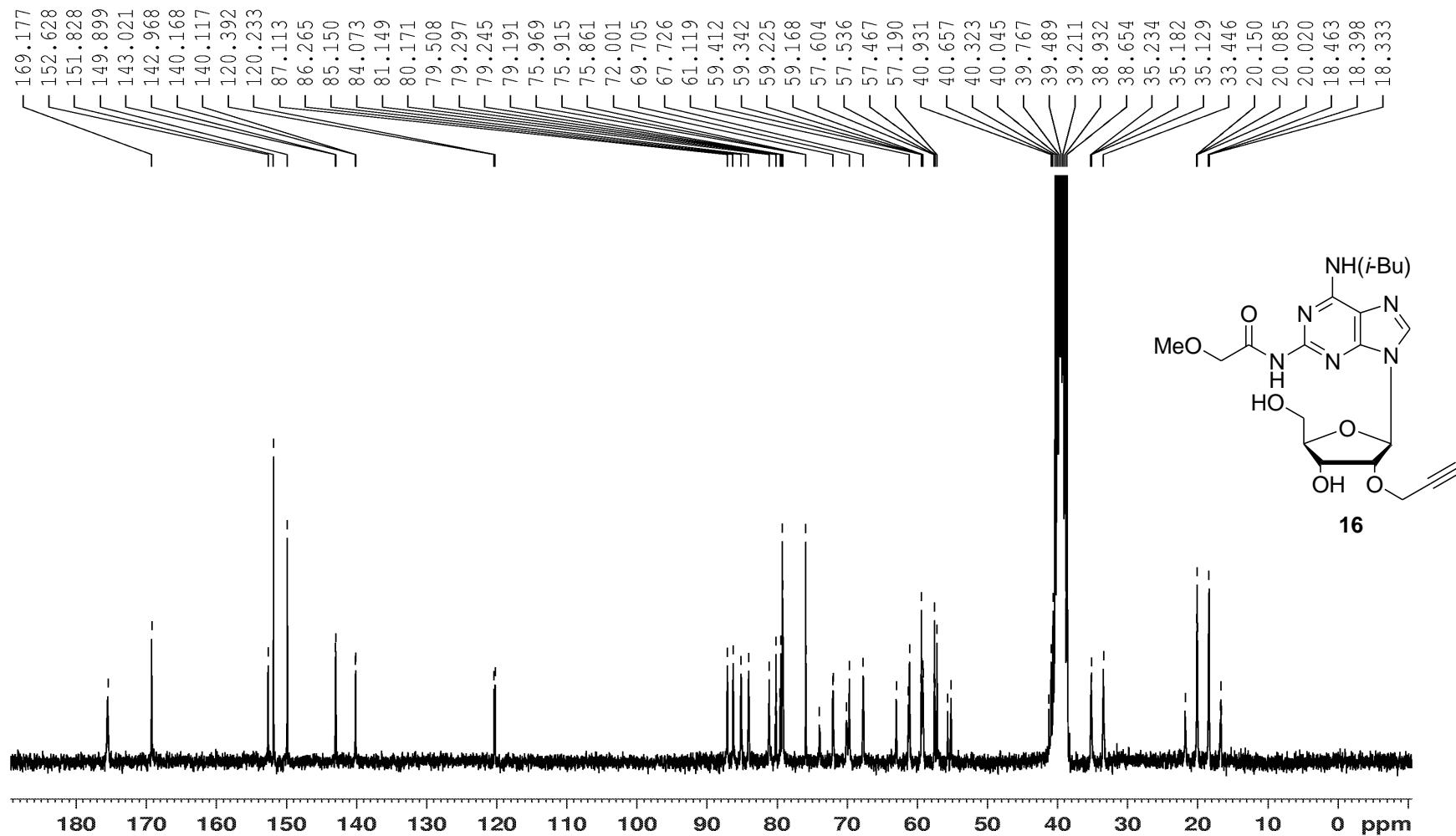
**Figure S43.**  $^{13}\text{C}$  NMR spectrum of compound **16**.



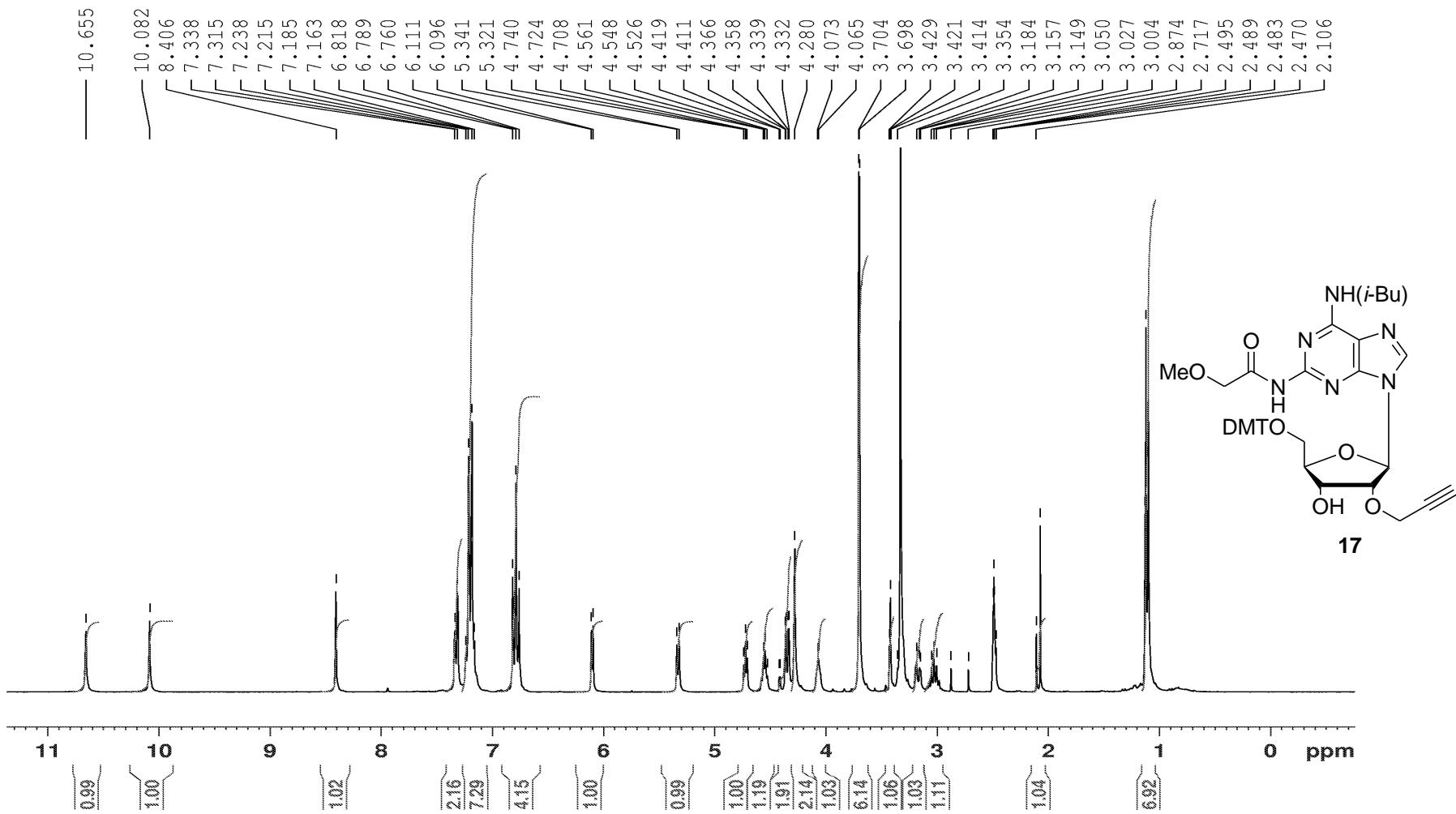
**Figure S44.** DEPT-135 spectrum of compound **16**.



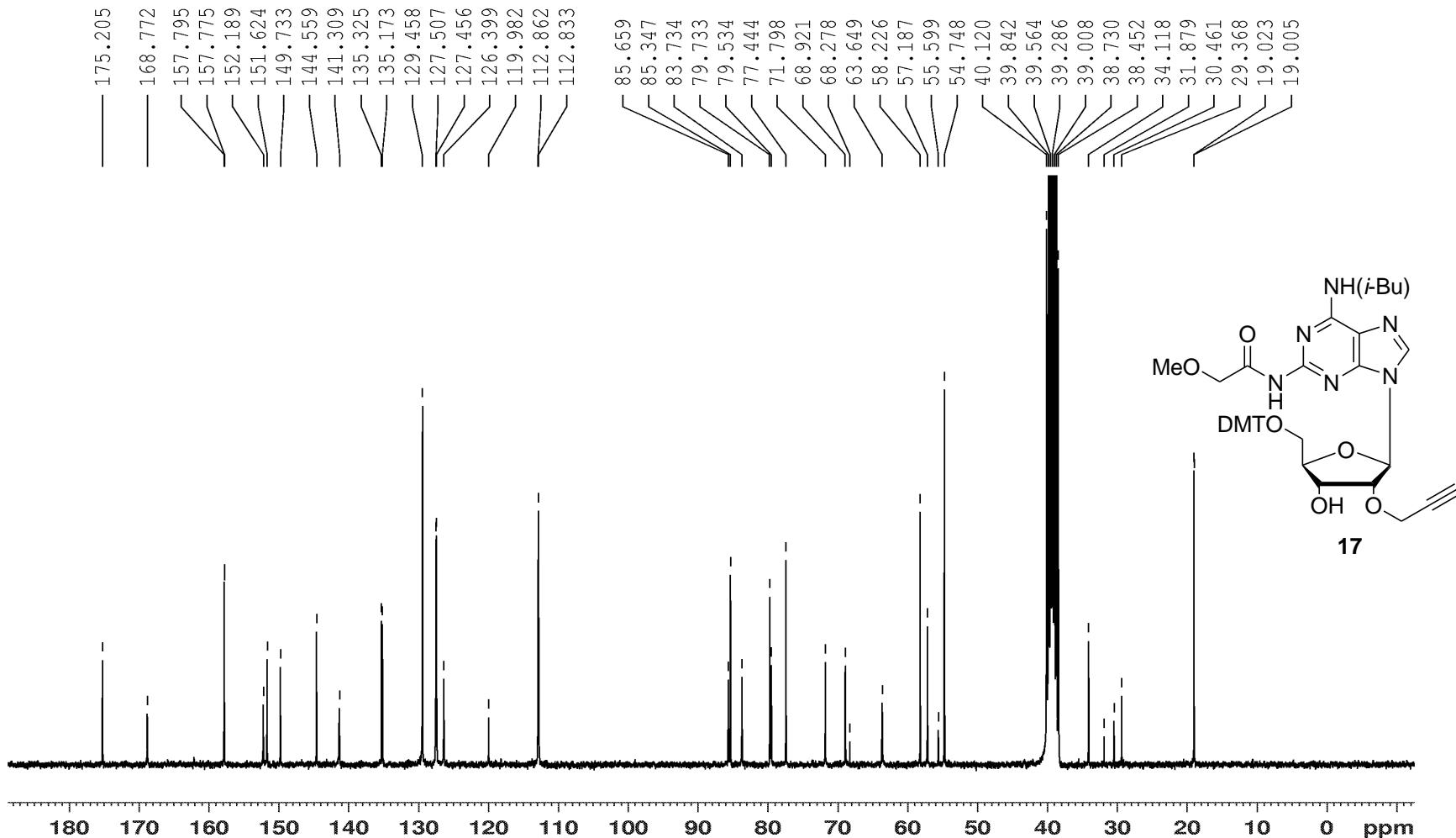
**Figure S45.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 16.



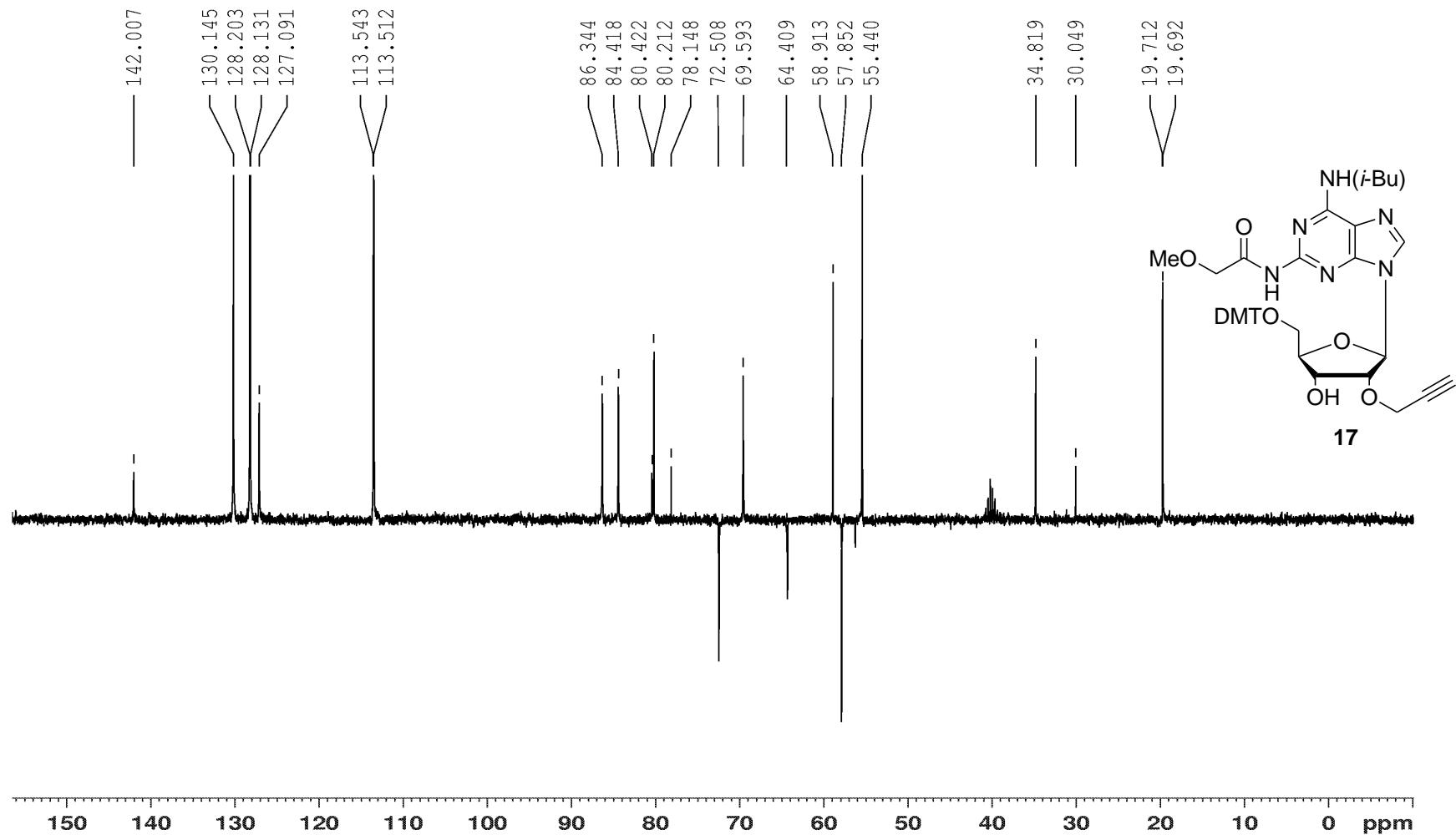
**Figure S46.**  $^1\text{H}$  NMR spectrum of compound 17.



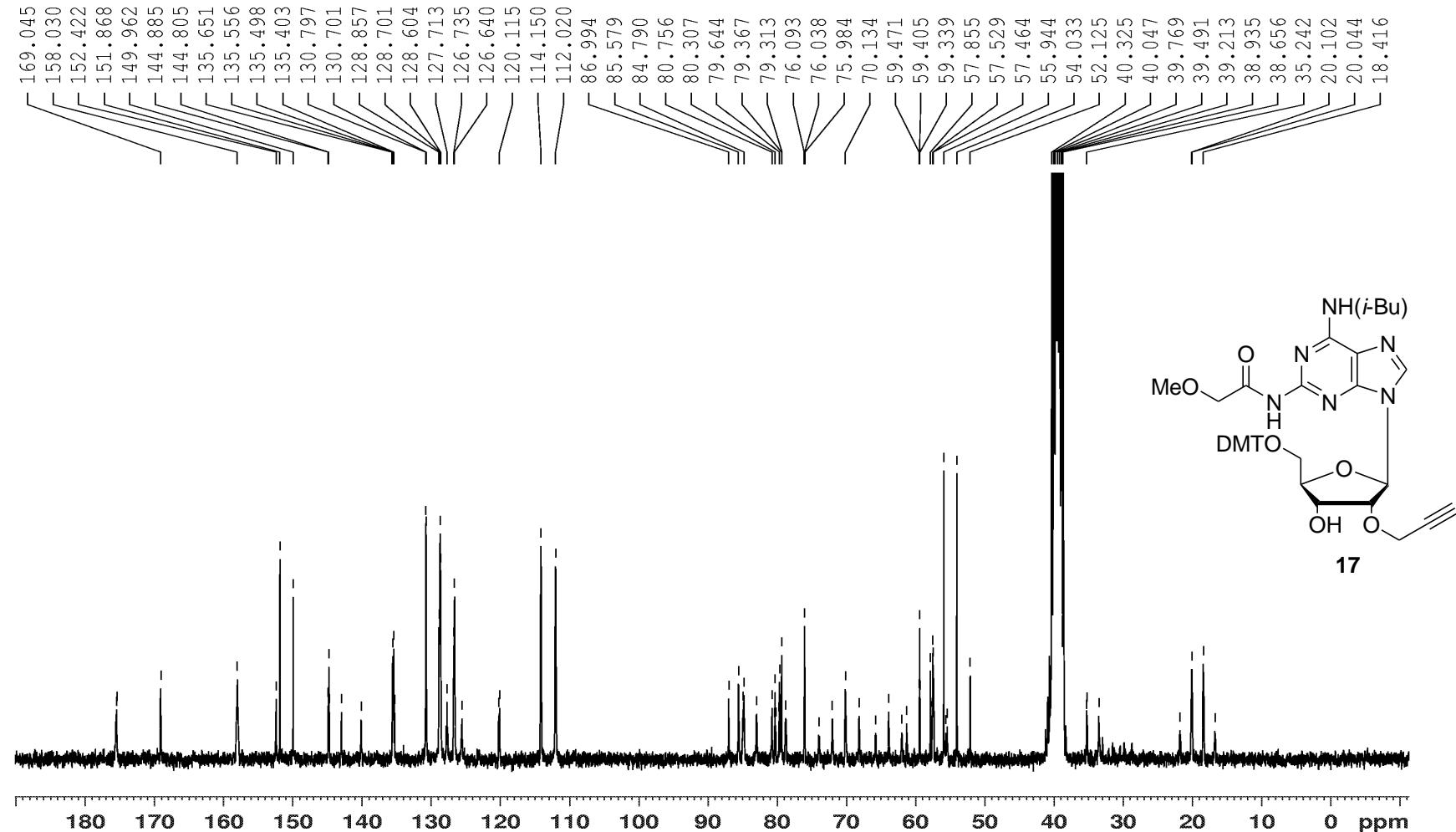
**Figure S47.**  $^{13}\text{C}$  NMR spectrum of compound 17.



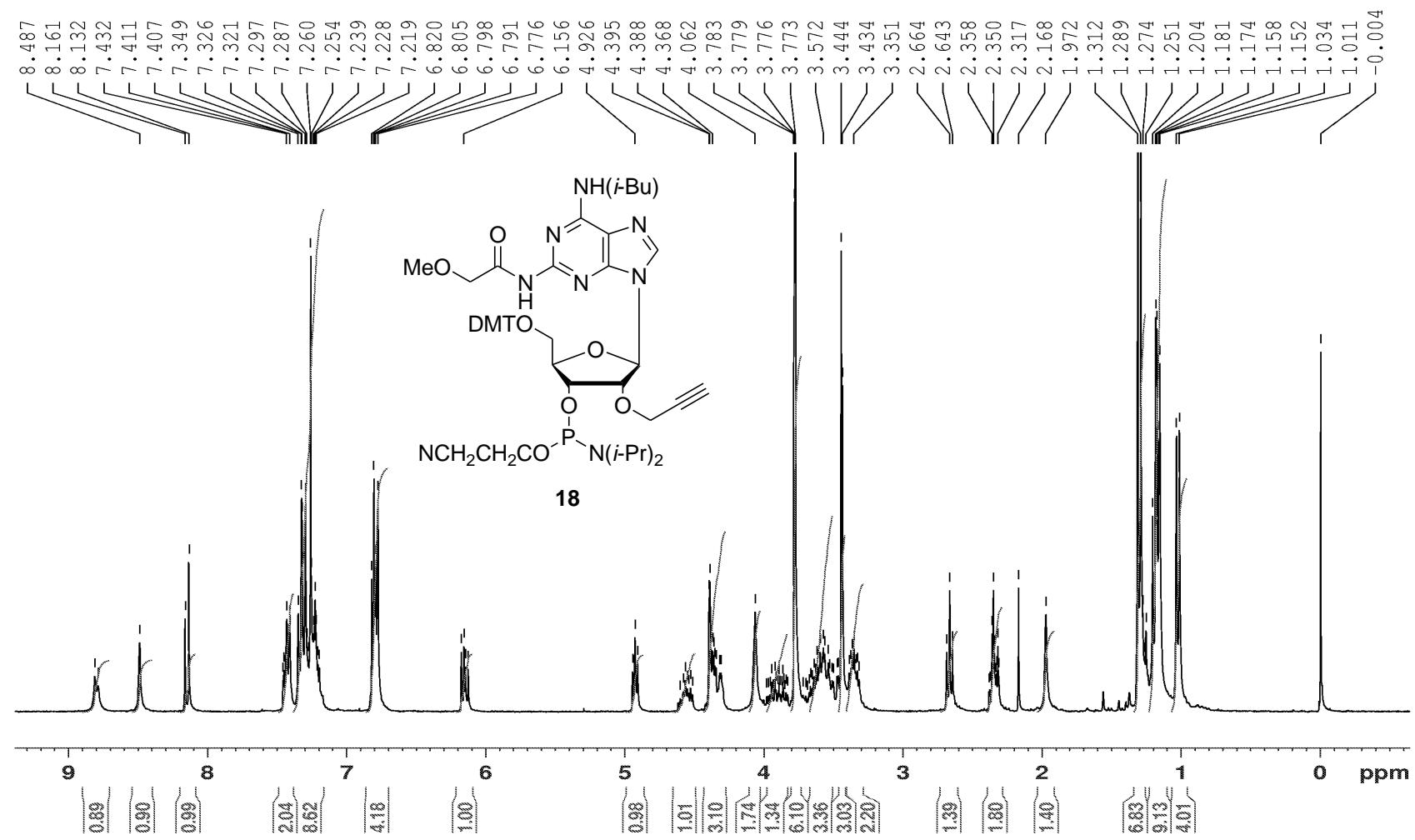
**Figure S48.** DEPT-135 spectrum of compound 17.



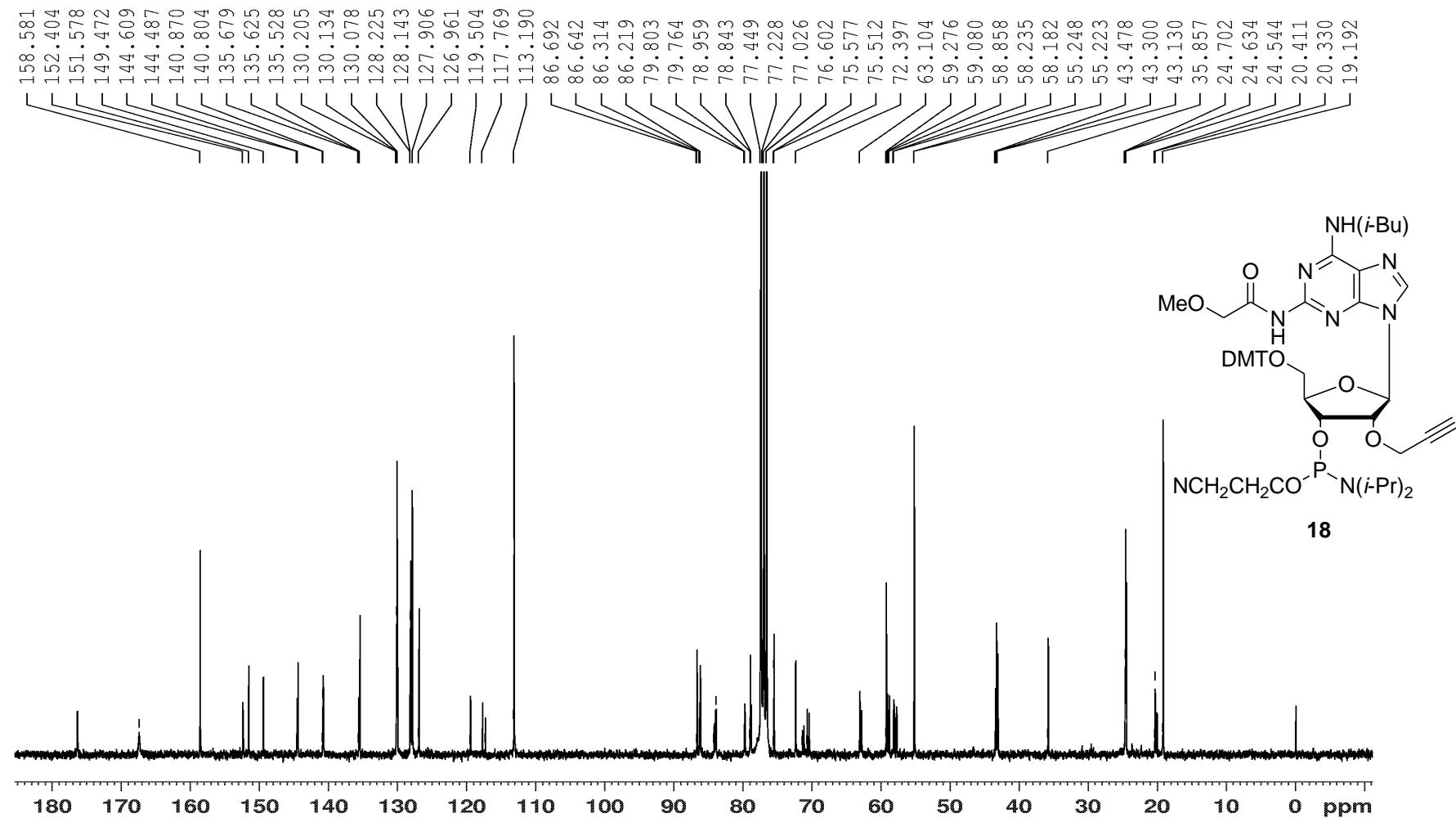
**Figure S49.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 17.



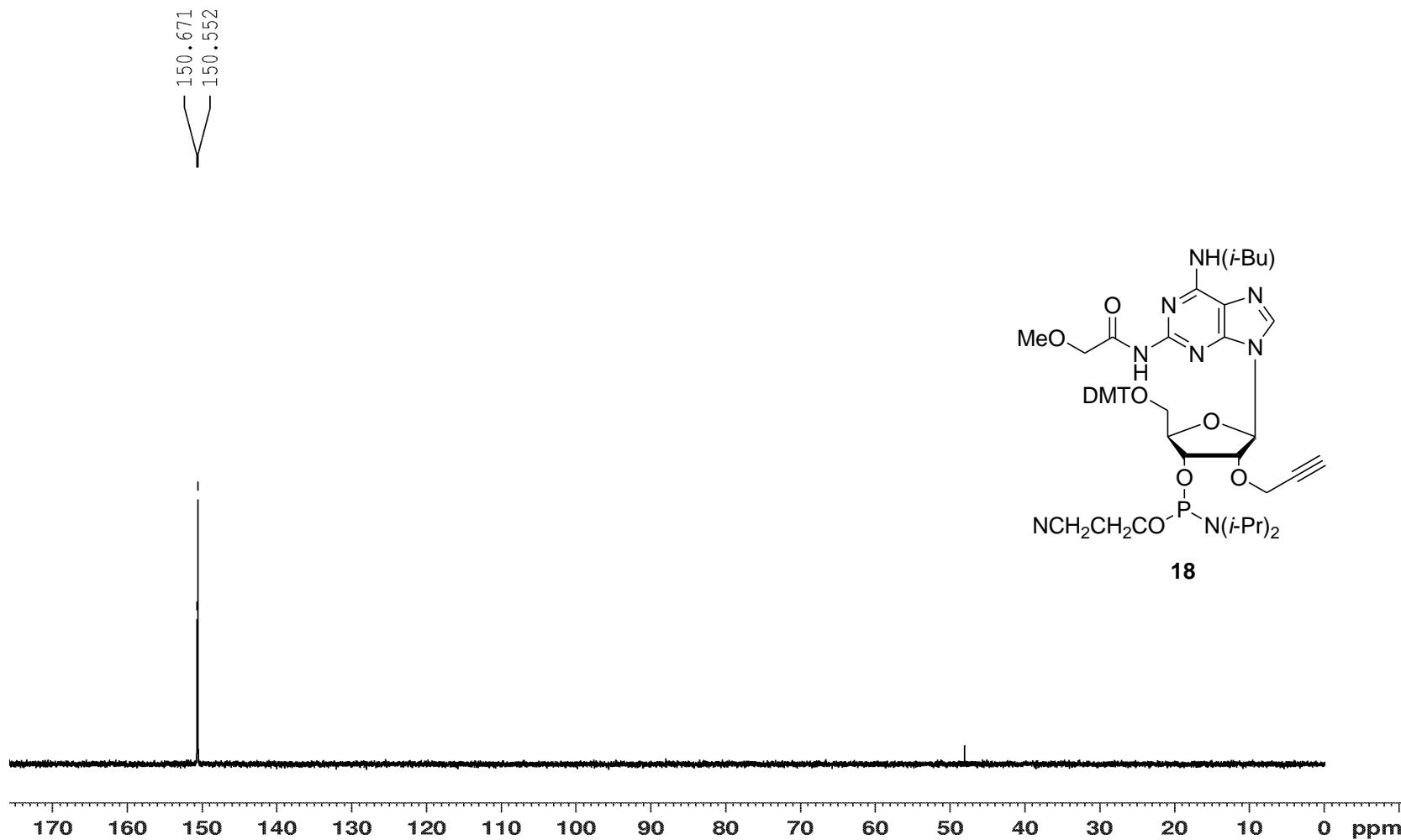
**Figure S50.**  $^1\text{H}$  NMR spectrum of compound 18.



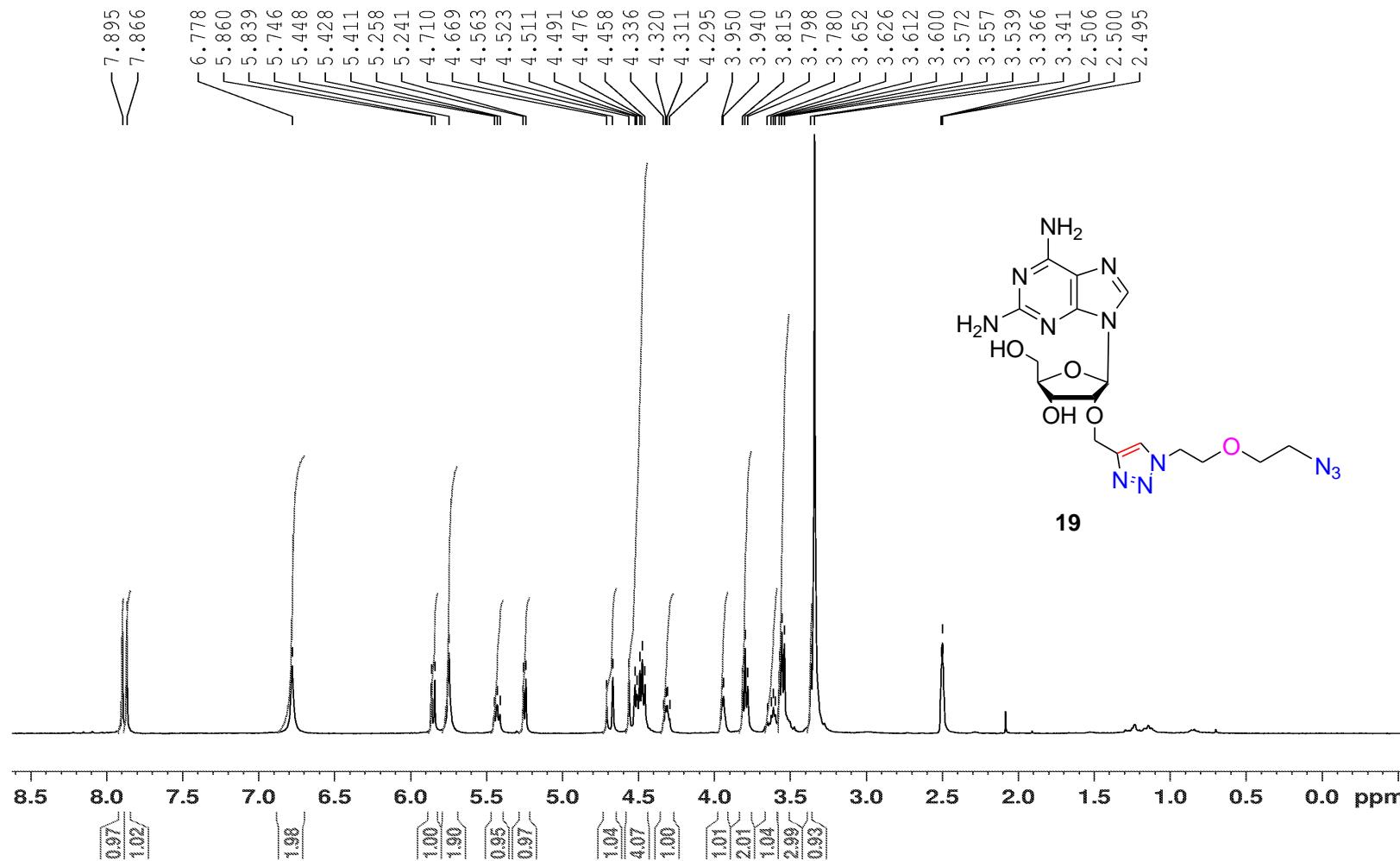
**Figure S51.**  $^{13}\text{C}$  NMR spectrum of compound **18**.



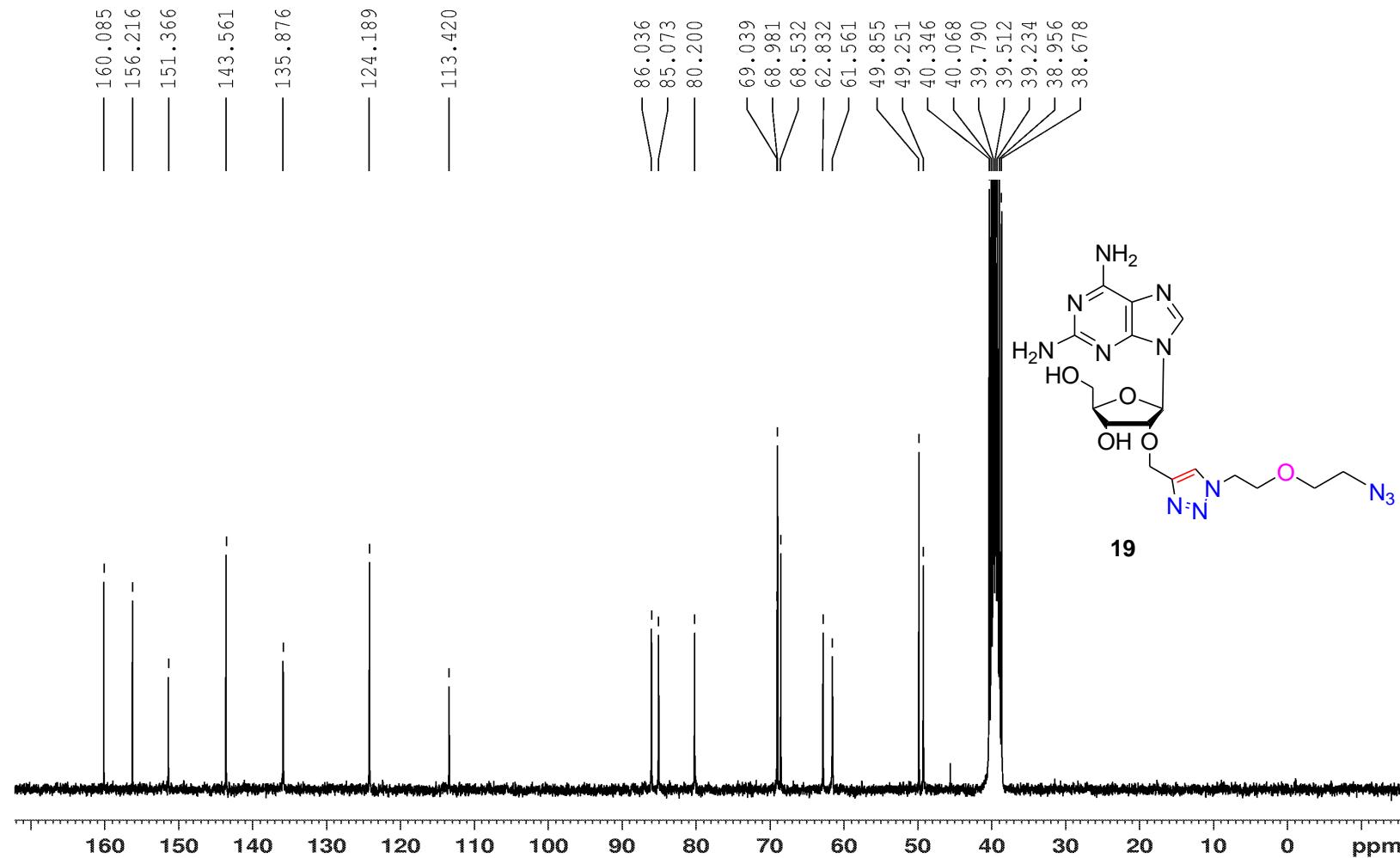
**Figure S52.**  $^{31}\text{P}$  NMR spectrum of compound 18.



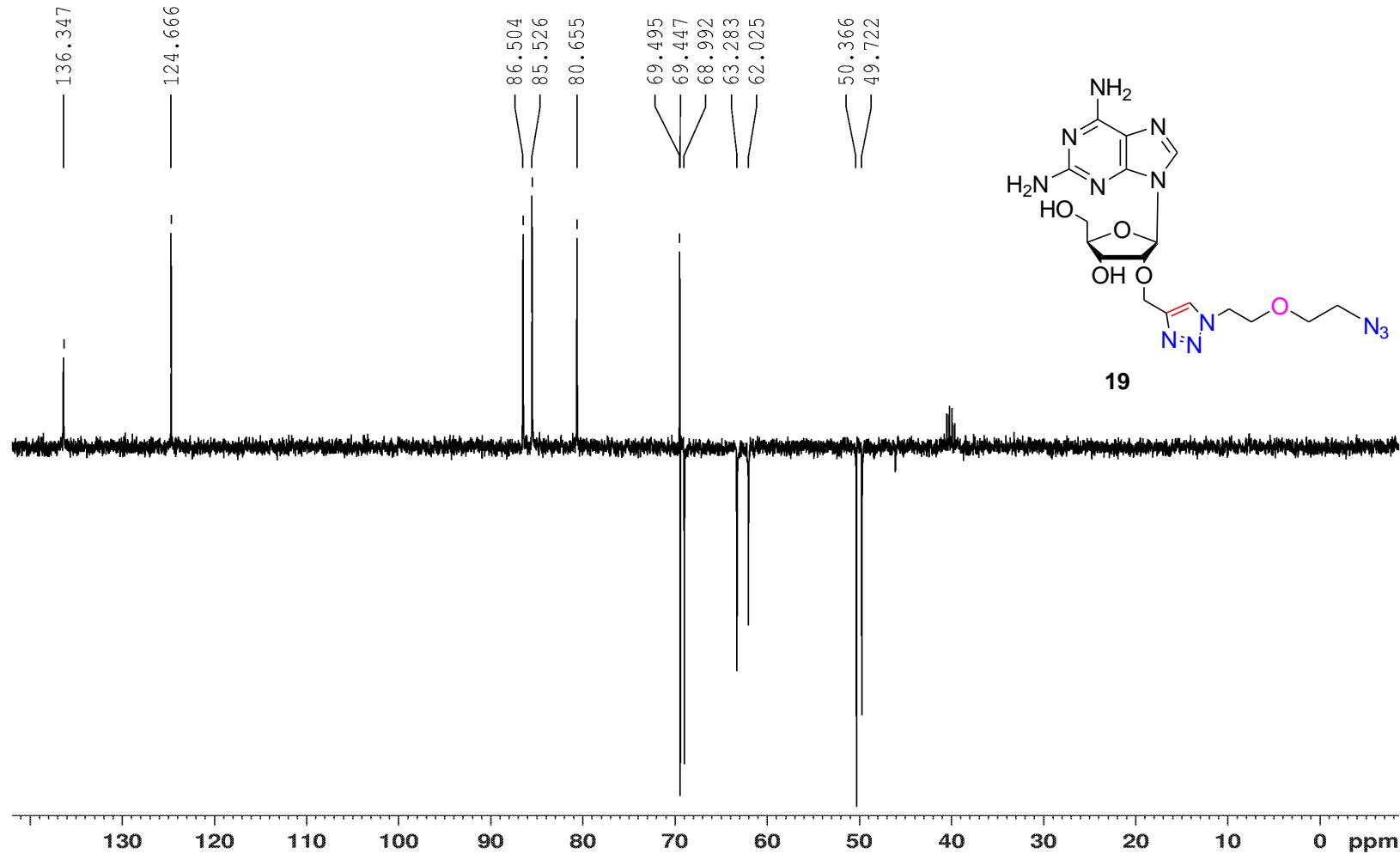
**Figure S53.**  $^1\text{H}$  NMR spectrum of compound 19.



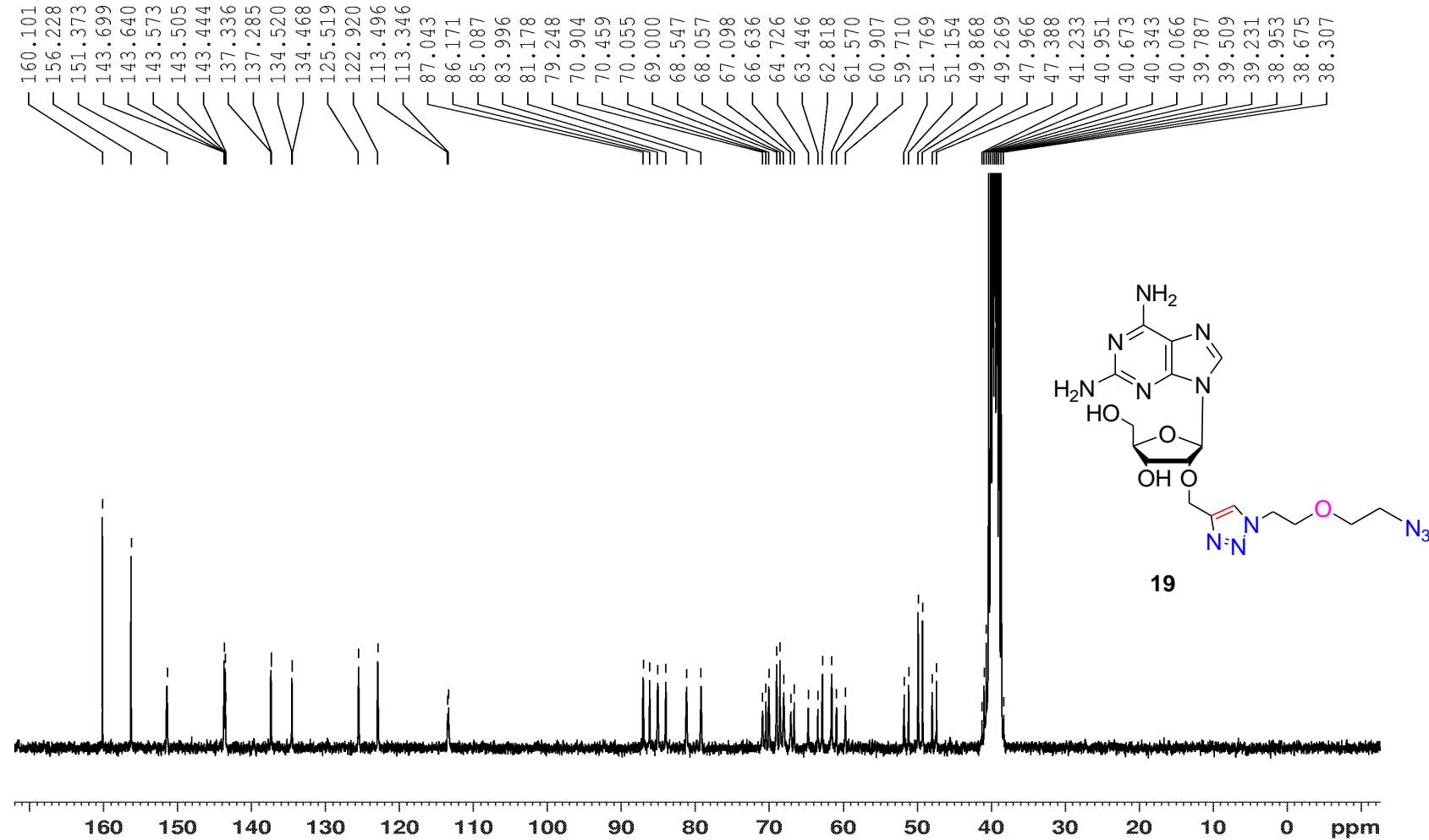
**Figure S54.**  $^{13}\text{C}$  NMR spectrum of compound **19**.



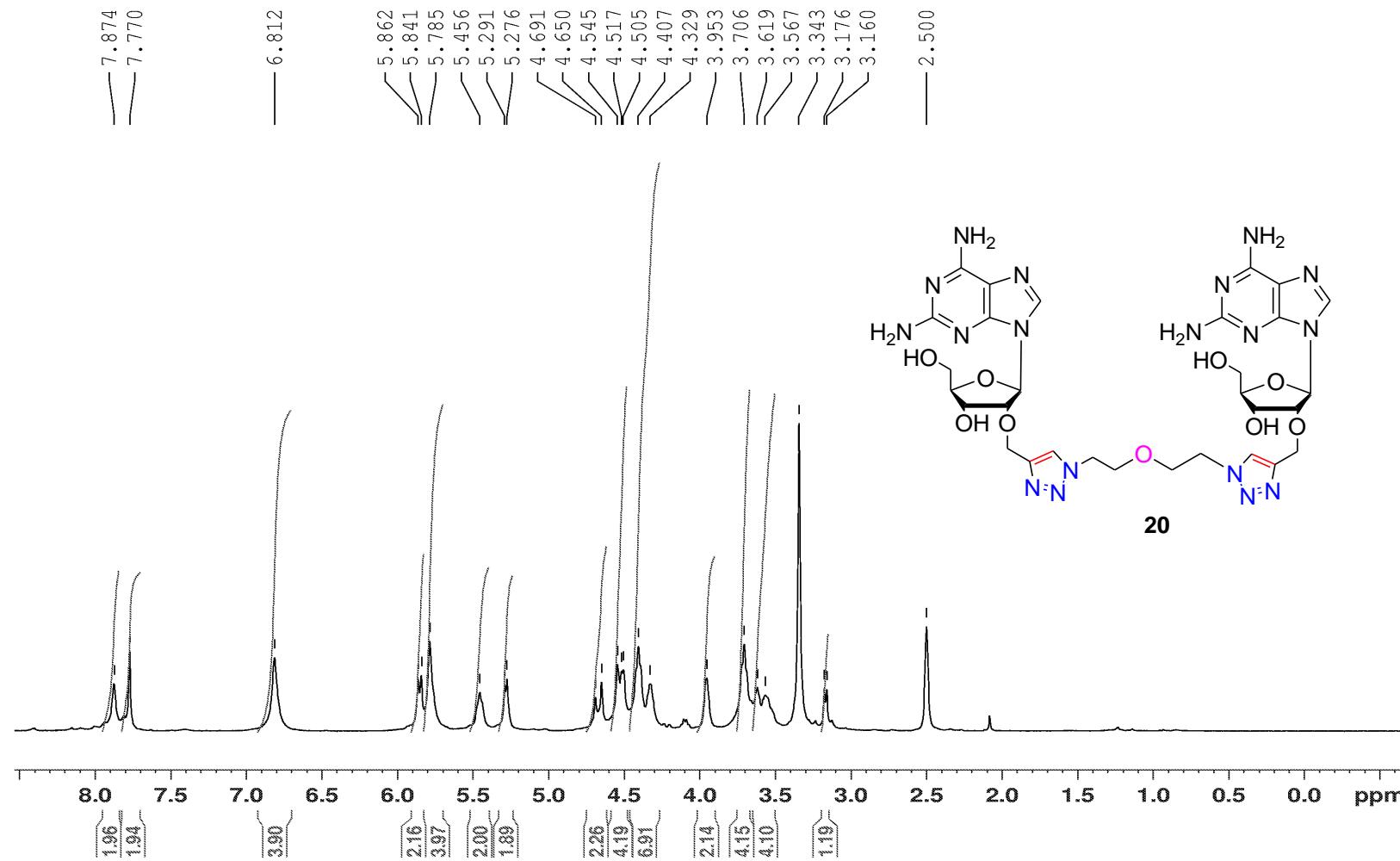
**Figure S55. DEPT-135 spectrum of compound 19.**



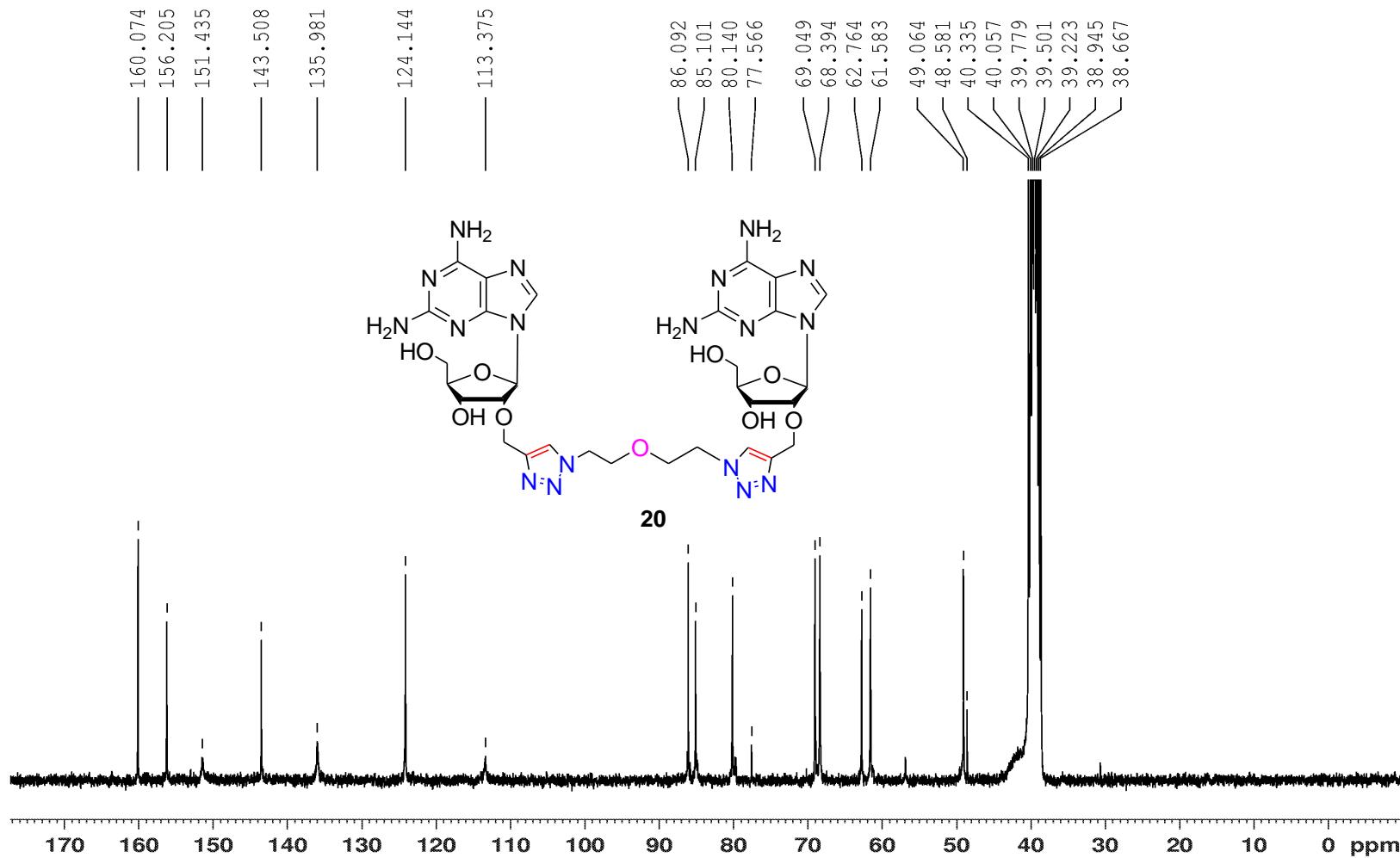
**Figure S56.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 19.



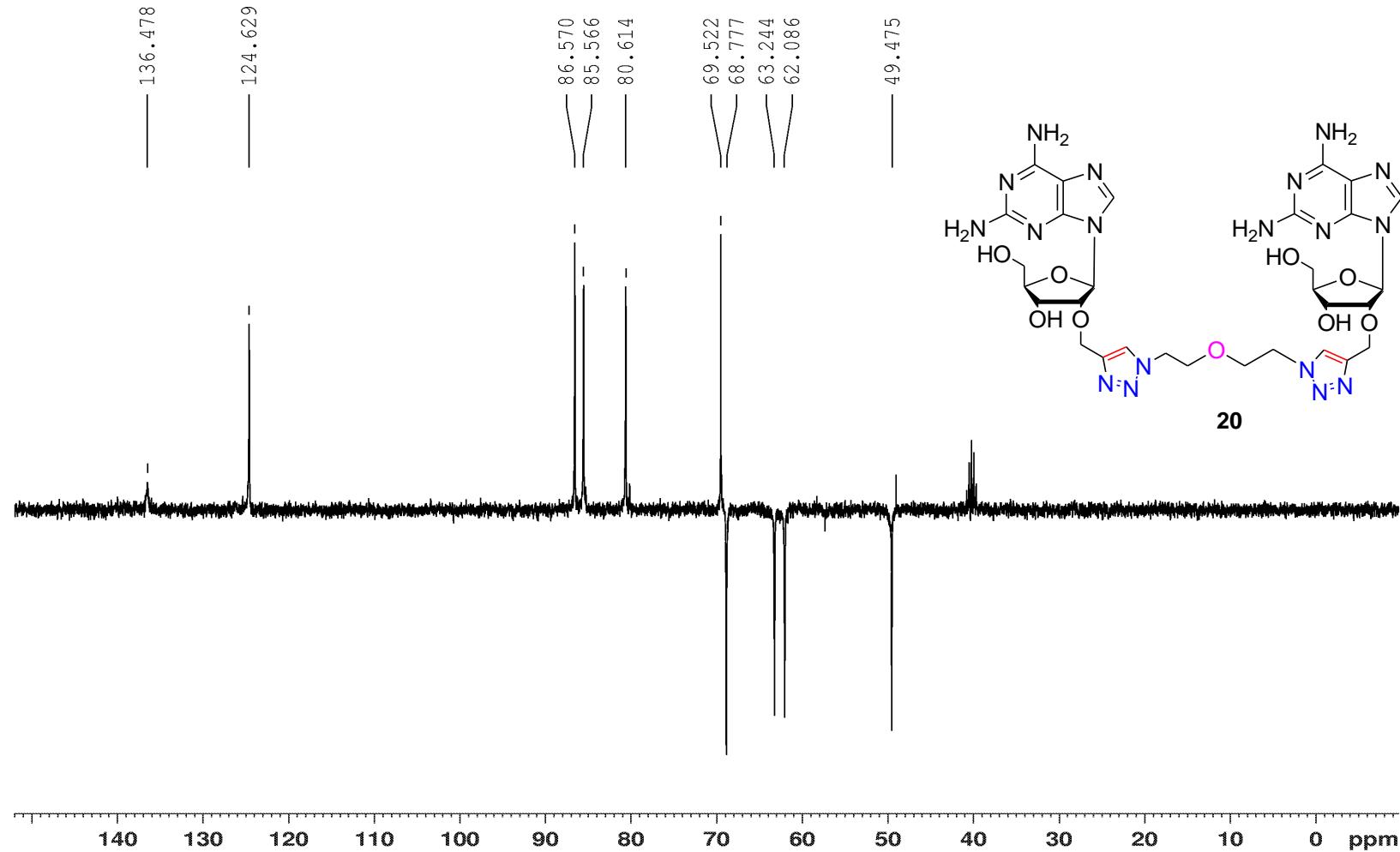
**Figure S57.**  $^1\text{H}$  NMR spectrum of compound 20.



**Figure S58.**  $^{13}\text{C}$  NMR spectrum of compound 20.



**Figure S59. DEPT-135 spectrum of compound 20.**



**Figure S60.**  $^1\text{H}$ - $^{13}\text{C}$  gated-decoupled spectrum of compound 20.

