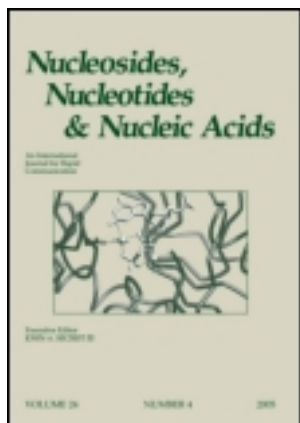


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### PHOSPHORAMIDITES AND OLIGONUCLEOTIDES CONTAINING 7-DEAZAPURINES AND PYRIMIDINES CARRYING AMINOPROPARGYL SIDE CHAINS

F. Seela<sup>a</sup>, N. Ramzaeva<sup>a</sup>, P. Leonard<sup>a</sup>, Y. Chen<sup>a</sup>, H. Debelak<sup>a</sup>, E. Feiling<sup>a</sup>, R. Kröschel<sup>a</sup>, M. Zulauf<sup>a</sup>, T. Wenzel, T. Fröhlich & M. Kostrzewa

<sup>a</sup> Institut für Chemie, Universität Osnabrück, Laboratorium für Organische und Bioorganische Chemie, Germany

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NUCLEOSIDES, NUCLEOTIDES & NUCLEIC ACIDS, 20(4–7), 1421–1424 (2001)

## PHOSPHORAMIDITES AND OLIGONUCLEOTIDES CONTAINING 7-DEAZAPURINES AND PYRIMIDINES CARRYING AMINOPROPARGYL SIDE CHAINS

F. Seela,\* N. Ramzaeva,<sup>1</sup> P. Leonard,<sup>1</sup> Y. Chen,<sup>1</sup>  
H. Debelak,<sup>1</sup> E. Feiling,<sup>1</sup> R. Kröschel,<sup>1</sup> M. Zulauf,<sup>1</sup>  
T. Wenzel,<sup>2</sup> T. Fröhlich,<sup>2</sup> and M. Kostrzewa<sup>2</sup>

<sup>1</sup>Laboratorium für Organische und Bioorganische Chemie,  
Institut für Chemie, Universität Osnabrück, Germany

<sup>2</sup>Bruker Saxonia Analytik GmbH, Leipzig, Germany

### ABSTRACT

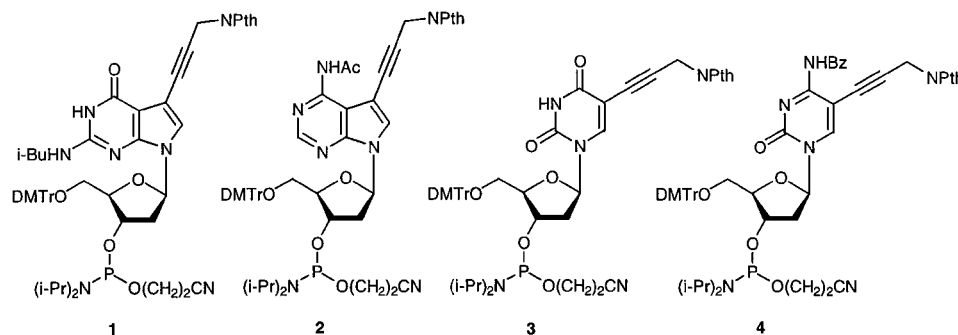
The synthesis of phosphoramidites containing 7-deazaguanine, 7-deazaadenine, uracil and cytosine carrying aminopropargyl chains is described. The corresponding oligonucleotides are stabilized in duplexes thermally as well as against degradation by exonucleases.

### INTRODUCTION

In a series of manuscripts our laboratory has reported on oligonucleotides containing alkynyl- or aminoalkynyl side chains linked to the 7-position of 7-deazapurines or 8-aza-7-deazapurines (purine numbering is used throughout the manuscript) (1–3). A positively charged side chain transforms a negatively charged oligonucleotide in a zwitterion or even a positively charged species. As a result favorable properties are generated, such as duplex stabilization, resistance against enzymatic degradation, or an increased sensitivity of oligonucleotide detection by MALDI-TOF spectrometry.

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\*Corresponding author.



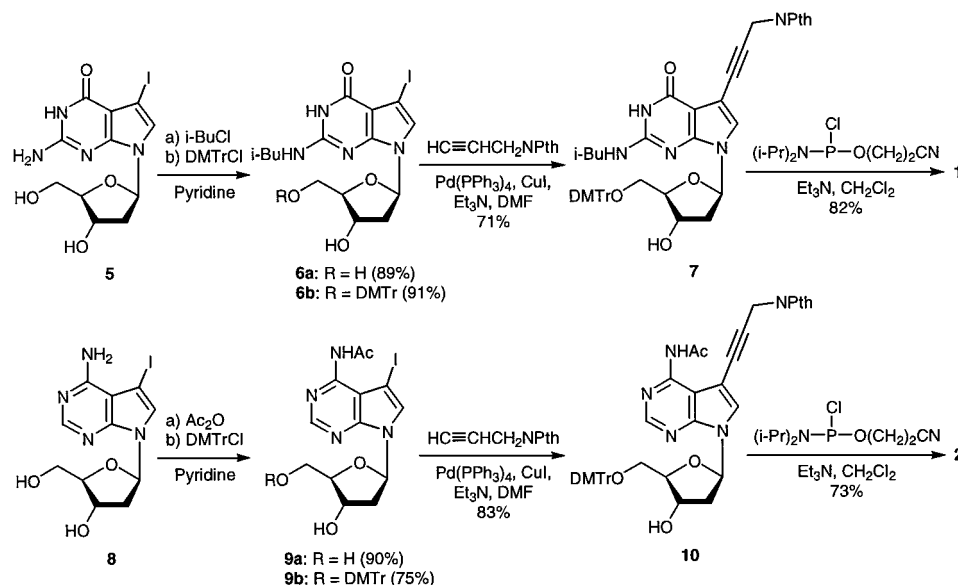
*Scheme 1.*

This manuscript reports on the synthesis of the phosphoramidites **1–4** (Scheme 1) carrying an aminopropargyl side chain protected by phthaloyl residues (4–6). This protecting group is removed with ammonia under standard conditions (25% ammonia, 60°C, 24 h) and is superior over the rather labile trifluoroacetyl group (7,8) which gives rise to side reaction (9).

## RESULTS AND DISCUSSION

### 1. Synthesis of the 7-deazapurine Phosphoramidites **1** and **2**

The synthesis of the phosphoramidites **1** and **2** was performed using the iodo compounds **5** (10) and **8** (11) as precursors (Scheme 2). While the 7-deazaguanine



*Scheme 2.*





base was protected with an isobutyryl residue, the 7-deazaadenine moiety was acetylated. Next, the DMTr residues were introduced. Compounds **6b** and **9b** were coupled with phthaloyl-propargylamine employing the Sonogashira reaction (Pd(PPh<sub>3</sub>)<sub>4</sub>, CuI, Et<sub>3</sub>N in DMF). The fully protected nucleosides **7** and **10** gave the phosphoramidites **1** and **2** applying standard methodology.

**Table.** T<sub>m</sub>-Values and Thermodynamic Data of Base Modified Oligonucleotides

Duplex		T <sub>m</sub> [°C]	ΔH° [kcal/mol]	ΔS° [cal/mol · K]	ΔG° <sub>298</sub> [kcal/mol]
5'-d(T-A-G-G-T-C-A-A-T-A-C-T)	<b>17</b>	50 <sup>a</sup>	-90	-252	-11.8
3'-d(A-T-C-C-A-G-T-T-A-T-G-A)	<b>18</b>	47 <sup>b</sup>	-89	-253	-10.9
5'-d(T-A-G-G-T-C-A-A-T-A-C-T)	<b>17</b>	54 <sup>a</sup>	-80	-220	-12.0
3'-d(A-T-C-C-A-G*-T-T-A-T-G*-A)	<b>19</b>	52 <sup>b</sup>	-95	-268	-12.0
5'-d(T-A-G*-G*-T-C-A-A-T-A-C-T)	<b>20</b>	54 <sup>a</sup>	-71	-189	-11.9
3'-d(A-T-C-C-A-G*-T-T-A-T-G*-A)	<b>19</b>	54 <sup>b</sup>	-98	-272	-13.0
5'-d(T-A-G-G-T-C-A-A-T-A-C-T)	<b>17</b>	55 <sup>a</sup>	-84	-232	-12.4
3'-d(A-T-C-C-A*-G-T-T-A*-T-G-A)	<b>21</b>	53 <sup>b</sup>	-85	-237	-11.8
5'-d(T-A*-G-G-T-C-A*-A*-T-A*-C-T)	<b>22</b>	59 <sup>a</sup>	-82	-222	-12.9
3'-d(A-T-C-C-A*-G-T-T-A*-T-G-A)	<b>21</b>	56 <sup>b</sup>	-84	-230	-12.7
5'-d(T-A-G-G-U*-C-A-A-U*-A-C-T)	<b>23</b>	53 <sup>a</sup>	-86	-239	-12.0
3'-d(A-T-C-C-A-G-T-T-A-T-G-A)	<b>18</b>	50 <sup>b</sup>	-91	-254	-11.8
5'-d(T-A-G-G-T-C*-A-A-T-A-C*-T)	<b>24</b>	57 <sup>a</sup>	-70	-187	-12.3
3'-d(A-T-C*-C*-A-G-T-T-A-T-G-A)	<b>25</b>	56 <sup>b</sup>	-87	-239	-13.1
5'-d(T-A-G-G-T-C*-A-A-T-A-C*-T)	<b>24</b>	54 <sup>a</sup>	-81	-222	-12.0
3'-d(A-T-C-C-A-G-T-T-A-T-G-A)	<b>18</b>	53 <sup>b</sup>	-84	-234	-11.8

<sup>a</sup>) 1 M NaCl, 100 mM MgCl<sub>2</sub>, 60 mM Na-cacodylate (pH 7.0). <sup>b</sup>) 100 mM NaCl, 10 mM MgCl<sub>2</sub>, and 10 mM Na-cacodylate (pH 7.0) with 5 μM single strand concentration. The residues with a\* represent the modified constituents.

### 2. Synthesis of the Pyrimidine Phosphoramidites **3** and **4**

The reaction sequence performed on the 7-deazapurine nucleosides was also used for the preparation of the phosphoramidite **3** (Scheme 3). The phosphoramidite **4** was prepared in a slightly different way. The 5-iodo-2'-deoxycytidine (**14**) was directly used for the cross coupling reaction yielding the derivative **15**. The crude reaction product was tritylated and benzoylated in a one-pot reaction to give the intermediate **16**. Afterwards, the phosphoramidite **4** was prepared as described before.

### 3. Synthesis and Properties of Oligonucleotides

The oligonucleotides shown in the Table were prepared by solid-phase synthesis in a 1- $\mu$ mole scale using the phosphoramidites **1–4**. The coupling yields were always higher than 96%. The oligonucleotides were deprotected and purified as DMTr derivatives. They were detritylated with 3% aq. trifluoroacetic acid on OPC cartridges. In the case of the dU<sup>\*</sup>-derivatives a side product was formed during the solid-phase synthesis when multiple residues were incorporated. The T<sub>m</sub>-values of the Table show that the positively charged aminopropargyl chain stabilizes the duplexes. Also degradation with exonucleases is retarded. The favorable properties of such oligonucleotides were used in MALDI-TOF spectrometry to increase the oligonucleotide detection sensitivity (12).

## REFERENCES

1. Seela, F.; Zulauf, M. *Helv. Chim. Acta*, **1999**, 82, 1878–1898.
2. Ramzaeva, N.; Mittelbach, C.; Seela, F. *Helv. Chim. Acta*, **1997**, 80, 1809–1822.
3. Seela, F.; Zulauf, M. *J. Chem. Soc., Perkin Trans. 1*, **1999**, 479–488.
4. Ramzaeva, N.; Mittelbach, C.; Seela, F. *Nucleosides Nucleotides*, **1999**, 18, 1439–1440.
5. Gibson, K.J.; Benkovic, S.J. *Nucleic Acids Res.*, **1987**, 15, 6455–6467.
6. Meyer, R.B. in "Methods in Molecular Biology", **1994**, Vol 26, pp. 73–91, Humana Press Inc. Totowa, NJ, USA.
7. Hobbs, Jr., F.W. *J. Org. Chem.*, **1989**, 54, 3420–3422.
8. Ahmadian, M.; Zhang, P.; Bergstrom, D.E. *Nucleic Acids Res.*, **1988**, 26, 3127–3135.
9. Ferrer, E.; Neubauer, G.; Mann, M.; Eritja, R. *J. Chem. Soc., Perkin Trans. 1*, **1997**, 2051–2057.
10. Ramzaeva, N.; Seela, F. *Helv. Chim. Acta*, **1995**, 78, 1083–1090.
11. Seela, F.; Zulauf, M. *Synthesis*, **1996**, 726–730.
12. Sauer, S.; Lechner, D.; Berlin, K.; Lehrach, H.; Escary, J.L.; Fox, N.; Gut, I.G. *Nucleic Acids Res.*, **2000**, 28, e13.

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