Synthesis and Incorporation of *C(5′)*-Ethynylated Uracil-Derived Phosphoramidites into RNA

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The D-*allo*- and L-*talo*-hept-6-ynofuranosyluracil-derived phosphoramidites **11A** and **11T** were prepared in 9–10% yield over eight steps from the previously described propargylic alcohols **1A** and **1T**, respectively. The corresponding nucleotides were incorporated into rU_{14} by standard solid-phase synthesis. While the duplex consisting of rU_{14} with one L-*talo*-hept-6-ynofuranosyluracil in the middle of the strand and rA_{14} (**I**·**V**) had the same melting point as the reference duplex $rU_{14}\cdot rA_{14}$ (**I**·**II**), the duplex with one D-*allo*-hept-6-ynofuranosyluracil in the middle of rU_{14} and rA_{14} (**I**·**III**) melted 1.5° lower than the reference duplex. The duplex **I**·**VI** consisting of rU_{14} with six L-*talo*-hept-6-ynofuranosyluracils distributed over the entire strand and rA_{14} showed a melting point that is 11° lower than the reference duplex. The corresponding duplex **I**·**IV** of rU_{14} possessing six D-*allo*-hept-6-ynofuranosyluracils and rA_{14} showed a melting point which is more than 20° below the one of the reference duplex. These results are in qualitative agreement with the predictions based on the conformational analysis of the nucleosides and the interference of the ethynyl moiety with the hydration of the oligonucleotides.

Introduction. – We are exploring the chemistry of ethynylene-linked oligonucleotide analogues[1]) to further test our hypothesis that the structural differentiation between backbone and nucleobases is not required for duplex formation [3]. In this context, we prepared the monomers **1A** and **1T** [4]. Their availability leads to the question about the influence of the ethynyl substituents at C(5′) on the conformation and pairing of RNA analogues derived from these monomers (*Fig. 1*). Information about the influence of substituents at C(5′) in DNA and RNA analogues on pairing is of interest in the context of antisense or antigene therapy [5–14] and in view of preparing RNA analogues possessing new functional properties [15–18]. Only a few nucleosides modified at C(5′)[2]) were incorporated into RNA or DNA, and had either little effect on duplex stability [35][36] or destabilized it [18][37][38] (compare [28][39][40]).

Results and Discussion. – 1. *Conformational Analysis and Synthesis of the Phosphoramidites.* An evaluation of the influence of the *C(5′)*-ethynyl substituent on duplex formation for RNA analogues derived from **1A** and **1T** must take into account the distribution of conformers resulting from rotation about the C(4′)–C(5′) bond, the steric interaction of the ethynyl group with the nucleobases and the backbone, and the influence of the ethynyl group on solvation. To evaluate the population of the three staggered conformations resulting from rotation about the C(4′)–C(5′) bond, we indicate a *gauche* (*g*) interaction of two substituents by a slash (C/O) and an *antiperiplanar* (*t*) arrangement between H and O by a double slash (H//O), using the

[1]) See [1][2] and earlier papers of this series.

[2]) For pertinent examples, see [19–34].

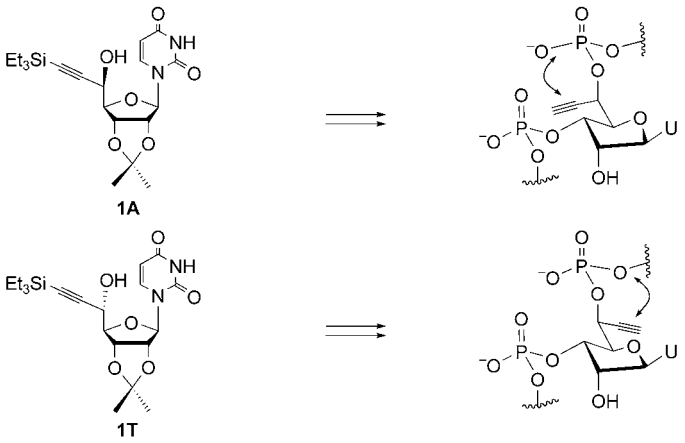


Fig. 1. *Modified rU_n oligonucleotides derived from* β-D-allo- *and* α-L-talo-*hept-6-ynofuranosyluracil* **1A** *and* **1T**

following interaction energies [41–44]: (O/C_{sp^3}) = 0.45 kcal/mol; (C_{sp^3}/C_{sp}) = 0.2 kcal/mol; O/O = 0.45 kcal/mol. The value for O/C_{sp} was estimated by considering that the value for O/C_{sp^3} is half the one for C_{sp^3}/C_{sp^3}. In a first approximation, we assumed O/C_{sp} to be half the one for C_{sp^3}/C_{sp}, *i.e.*, 0.1 kcal/mol.

A H//O interaction of −0.85 kcal/mol was deduced from the 57:29:14 distribution of the *gg*/*gt*/*tg* conformers resulting from rotation about the C(4′)–C(5′) bond of the β-D-ribofuranosyl-nucleosides [45]. On the basis of these values, one predicts the distribution of the conformers of β-D-*allo*-hept-6-ynofuranosyl- and α-L-*talo*-hept-6-ynofuranosyl-nucleosides shown in *Fig. 2*. The preferred conformation differs for the three nucleoside types. The β-D-ribofuranosyl-nucleosides prefer the *gg*, the β-D-*allo*-hept-6-ynofuranosyl-nucleosides the *gt-gg*, and the α-L-*talo*-hept-6-ynofuranosyl-nucleosides the *gg-gt* conformation. The results are strongly influenced by the value for H//O, although, qualitatively, they are not affected even by considerable deviations from it. Thus, assuming a value for H//O of −0.45 instead of −0.85 kcal/mol leads to a *gg-tg/gt-gg/tg-gt* conformer ratio for the D-*allo*-nucleosides of 29:48:23, and to a *gg-gt/gt-tg/tg-gg* ratio for the L-*talo* nucleosides of 65:21:14. The endocyclic orientation of the ethynyl group in the (preferred) *gt-gg* conformer of a D-*allo*-nucleoside and in the (disfavoured) *tg-gg* conformation of the L-*talo*-isomer may lead to a destabilizing steric interaction with the uracil moiety. This is suggested by the C(5′)···H–C(6) distance of 3.52 Å in *C(5′)*-ethynylated derivatives, the *C(3′) endo*-conformation of the furanosyl unit and an *anti*-conformation of the nucleobases, as it is found in the A form of RNA duplexes.

Inspection of an RNA strand in a A-helix [46] shows that a *C(5′)*-substituent must lead to a 1,5-interaction independently of the configuration at C(5′) (*Fig. 1*). A β-D-*allo*-hept-6-ynofuranosyl-nucleoside leads to a 1,5-interaction between the ethynyl group and P–O^-. An α-L-*talo*-hept-6-ynofuranosyl-nucleoside leads to a 1,5-interaction between the ethynyl group and P–*O*C(3′). Considering the small A value of the ethynyl group (0.18 kcal/mol [47]) and the long P–O bond, one expects that these 1,5-diaxial interactions will result in only minor destabilizations of the duplex. This

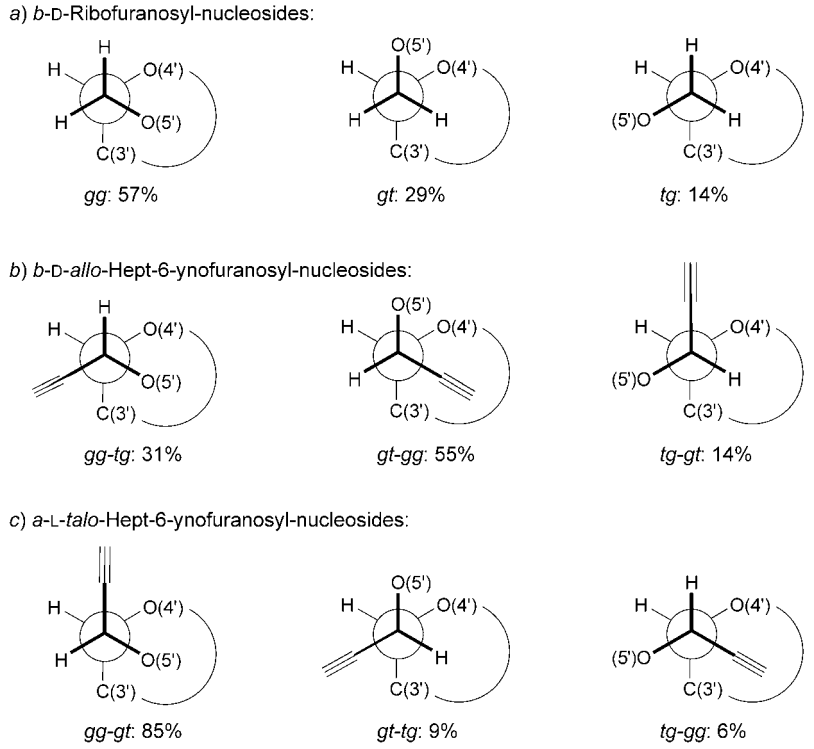


Fig. 2. *Calculated population of the conformations obtained by rotation about the C(5′) – C(4′) bond of* β-D-ribo-, β-D-allo-*hept-6-yno-, and* α-L-talo-*hept-6-ynofuranosyl-nucleosides*

expectation is supported by the observation that *Eschenmoser*'s p-RNA forms highly stable duplexes in spite of unavoidable 1,5-interactions [48 – 50].

An evaluation of the influence of the ethynyl group must also take the influence of the ethynyl group on the first hydration shell into account. H_2O Molecules of the first hydration shell are grouped into three classes [46] with decreasing binding affinity for phosphate, phosphodiester/sugar, and base. The ethynyl group of the β-D-*allo*-hept-6-ynofuranosyl-nucleosides points to the phosphate hydration site and should considerably disturb the hydration shell of the anionic centre. The ethynyl group of the α-L-*talo*-hept-6-ynofuranosyl-nucleosides points to the less-important phosphodiester/sugar hydration site and should result in a smaller destabilization of hydration.

The *gg-tg* conformation of β-D-*allo*-hept-6-ynofuranosyl-nucleosides required for duplex formation is disfavoured and leads to a destabilizing interaction of the ethynyl group with the first hydration shell. The *gg-gt* conformation of the α-L-*talo*-hept-6-ynofuranosyl-nucleosides required for duplex formation is preferred and leads to a destabilizing interaction with the less-important second hydration shell. Thus, duplexes containing β-D-*allo*-nucleotides are expected to be less stable then natural duplexes and also less stable than duplexes containing α-L-*talo*-nucleotides.

To synthesise the required D-*allo*-configured phosphoramidite **11A**, we transformed the propargylic alcohol **1A** [4] into the 2-nitrobenzoate **2A** (*Scheme*). De-isopropylidenation of **2A** in CF_3COOH/H_2O 1:1 gave the diol **3A**, which was converted to the tribenzoyl derivative **4A** (78%) and the dibenzoate **5A** (5%). The selective removal of the 2-nitrobenzoyl group of **5A** with Zn and NH_4Cl in $MeCN/H_2O$ 1:1 [51] to provide the propargylic alcohol **7A** proceeded well on a small scale, as suggested by TLC and a crude ^{1}H-NMR spectrum (*Table 1*, *Entry 3*). Performing the reaction on a gram scale led, however, to considerable amounts of the 2-aminobenzoate **6A**. Using activated Zn powder and NH_4OAc did not significantly affect the yield and the product distribution (*Entry 4*). Zn/CF_3COOH or Zn/CH_3COOH (*Entries 6* and *7*) led to **6A** as the main product. Starting from 1–5 g of **4A**, the propargylic alcohol **7A** was finally obtained in reproducible yields of 60–85% by portionwise addition of a Zn/NH_4Cl mixture (*Entries 11* and *12*). The propargylic alcohol **7A** was converted to the phosporamidites **11A** according to a procedure described by *Pitsch et al.* [52][53]. Thus, **7A** was dimethoxytritylated [54], and the crude product was *O*-debenzoylated under standard conditions to give the diol **8A** in 59% yield. The 2′-O-[(triisopropylsilyloxy)methyl] (=TOM) protecting group was introduced by stannylation with Bu_2SnCl_2 and $(i\text{-}Pr)_2NEt$, followed by treatment with TOMCl at 80°, leading to the desired 2′-*O*-alkylated nucleoside **9A** as the major product (33%) that was separated by chromatography from the 3′-*O*-alkylated isomer **10A** (16%) and from starting material **8A** (28%). The alcohol **9A** was transformed into the phosphoramidites **11A** (81%; 3:2 mixture of diastereoisomers) according to standard procedures [55].

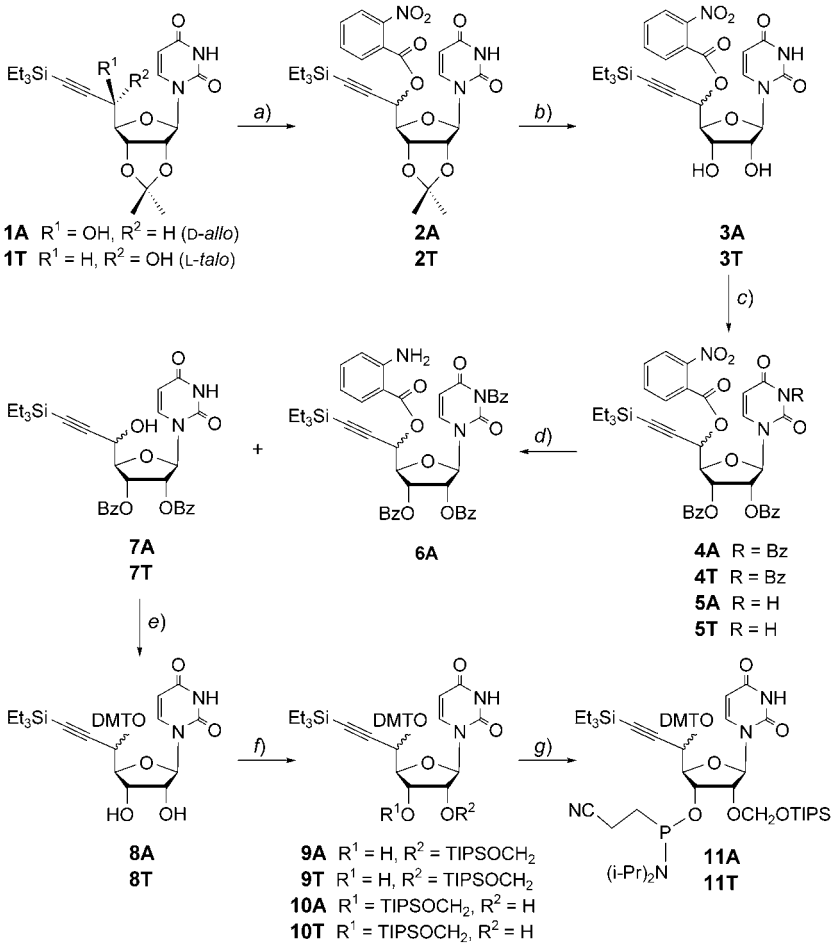
Table 1. *Conditions for the De(2-nitrobenzoylation) of* **4A** (*Entries 1–10: ca.* 20-mg batch, *Entries 11* and *12:* 1–5-g batch)

Entry	Reagents (each 5 equiv.)	Temp. and time	Solvent	Yield of **7A**
1	Zn/NH_4Cl (in 1 portion)	23°, 8 h	THF/H_2O 1:1	*ca.* 90%
2	Zn/NH_4Cl (in 2 portions)	23°, 8 h	THF/H_2O 1:1	*ca.* 90%
3	Zn/NH_4Cl (in 1 portion)	23°, 8 h	$MeCN/H_2O$ 1:1	*ca.* 90%
4	Zn/NH_4OAc (in 1 portion)	23°, 8 h	$MeCN/H_2O$ 1:1	*ca.* 90%
5	Zn/NH_4Cl (in 1 portion)	0°, 5 h	$MeCN/H_2O$ 1:1	[a]
6	Zn/AcOH (in 1 portion)	23°, 7 h	$MeCN/H_2O$ 1:1	[a]
7	Zn/CF_3CO_2H (in 1 portion)	23°, 7 h	$MeCN/H_2O$ 1:1	[a]
8	Zn/NH_4Cl (in 1 portion)[b]	23°, 7 h	$MeCN/H_2O$ 1:1	50%
9	Zn/NH_4Cl (in 1 portion)[c]	23°, 7 h	$MeCN/H_2O$ 1:1	60%
10	Zn/NH_4Cl (in 1 portion)[d]	23°, 7 h	$MeCN/H_2O$ 1:1	60%
11	Zn/NH_4Cl (in 5 portions)	23°, 7 h	$MeCN/H_2O$ 1:1	60%
12	Zn/NH_4Cl (in 5 portions)	23°, 7 h	$MeCN/H_2O$ 1:1	85%

[a]) Formation of **6A** and **7A** (not quantified); additional products in *Entry 5*. [b]) Excess NH_4Cl. [c]) Zn treated with 1M NaOH. [d]) Zn treated with 1M HCl.

Similarly, the α-L-*talo*-hept-6-ynofuranosyl-uracil **1T** was transformed *via* the intermediates **2T**–**4T** (13% of **5T** were obtained as side product) to the dimethoxytritylated diol **7T** (72% overall yield). The TOM protecting group was introduced similarly to the D-*allo*-series, providing mostly the *O(2′)*-protected alcohol **9T** (32%)

Scheme



a) 2-Nitrobenzoyl chloride, DMAP, pyridine, CH_2Cl_2; 78% of **2A**; 93% of **2T**. *b*) H_2O/CF_3COOH 1:1; quant. *c*) BzCl, 4-pyrrolidinopyridine, pyridine/CH_2Cl_2 1:1; 78% of **4A** and 5% of **5A**; 78% of **4T** and 13% of **5T**. *d*) Zn powder, NH_4Cl, MeCN/H_2O 1:1; 62% of **7A** (see also *Table 1*); >98% of **7T**. *e*) DMTCl, $AgNO_3$, *sym*-collidine, CH_2Cl_2, then NaOH, THF/MeOH/H_2O 5:4:1; 59% of **8A**; 57% of **8T**. *f*) Bu_2SnCl_2, $(i\text{-}Pr)_2NEt$, $(CH_2Cl)_2$, then TOMCl; 33% of **9A** and 16% of **10A**; 32% of **9T** and 10% of **10T**. *g*) 2-Cyanoethyl diisopropylphosphoramidochloridite, $(i\text{-}Pr)_2NEt$, CH_2Cl_2; 81% of **11A**; 76% of **11T**. DMAP = 4-(Dimethylamino)pyridine, DMT = 4,4′-dimethoxytrityl; TIPS = triisopropylsilyl; TOM = [(triisopropylsilyl)oxy]methyl.

besides its *O(3′)*-protected isomer **10T** (10%). The alcohol **9T** was transformed to the phosphoramidites **11T** (76%; 3:2 mixture of diastereoisomers), similarly as described for the synthesis of the D-*allo*-isomer **2**.

The signals for H–C(2′), H–C(3′), and H–C(5′) geminal to BzO groups are shifted downfield by 1.2–1.5 ppm as compared to those of H–C signals geminal to OH groups (*Tables 4* and *6* in the *Exper. Part*). The H–C(1′) and H–C(4′) signals are shifted downfield by 0.2–0.6 ppm upon benzoylation of the *cis* HO–C(2′) and

HO–C(3′). The upfield shift for H–C(2′) and H–C(3′) of **3A** and **3T** (4.24–4.36 ppm), and of H–C(5′) of **7A** and **7T** (4.83–4.86 ppm) show that the 2-nitrobenzoyl and the Bz group did not migrate. $J(1',2')/J(3',4') = 1.0-1.06$ suggests a *ca.* 1:1 equilibrium of the (*N*)- and (*S*)-conformers of the isopropylidene acetals **2A** and **2T**, whereas the (*N*)-conformer is slightly preferred in the diols **3A** and **3T** ($J(1',2')/J(3',4') = 0.73-0.8$). An increasing preference for the (*S*)-conformation is observed in the D-*allo*-series **8A**, **4A/5A/9A/11A**, **6A**, and **7A/10A** ($J(1',2')/J(3',4') = 1.5$, 2.1–2.3, 3.0, and 3.6–4.2, resp.) and in the L-*talo*-series **4T/8T/11T**, **7T/9T**, and **10T** ($J(1',2')/J(3',4') = 2.9-3.0$, 3.9–4.3, and 6.2, resp.). The L-*talo*-derivatives show stronger preference for the (*S*)-conformation than the corresponding D-*allo*-isomers with the exception of **7A/7T**. $J(4',5')$ for the L-*talo* derivatives **2T**–**4T** and **8T**–**10T** is 1.6–2.8 Hz larger than $J(4',5')$ for the corresponding D-*allo*-configured products, evidencing a stronger preference for the antiperiplanar orientation of H–C(4′) and H–C(5′) in these L-*talo*-isomers [56]. However, $J(4',5')$ of **7T** and **11T** is similar to $J(4',5')$ of **7A** and **11A** ($\Delta J \leq 0.3$ Hz). The value of $J(4',5')$ of the L-*talo*-isomers **2T**–**4T** and **7T**–**11T** (3.0–6.5 Hz) indicates a higher population of the *gt-tg* conformation than expected by the above calculation (9%).

The structure of the regioisomers **9A/10A** and **9T/10T** is revealed by the coupling constants of H–C(2′) and H–C(3′); the assignment was confirmed by H/D exchange and selective irradiation experiments. The large $J(2,OH)$ value of **9A** and **9T** (7.8–8.0 Hz) and the small $J(3,OH)$ value of **10A** and **10T** (1.6–2.8 Hz) evidence a intramolecular H-bond to the TOMO substituent (*cf.* [57][58]) and corroborate the (*S*)-conformation with a pseudoequatorial *O*-substituent at C(2′) and a pseudoaxial *O*-substituent at C(3′).

2. *Solid-Phase Synthesis of* C(5′)*-Ethynylated Ribonucleic Acids.* For the pairing studies, we chose the tetradecameric rU_{14} and rA_{14} strands. To study the influence of the ethynyl group, we incorporated one modified unit derived from **11A** or **11T** in the middle of the strand. In addition to the singly modified strands, we synthesized rU_{14} strands with six modifications distributed over the entire strand. The solid-phase synthesis of the oligomers **I**–**VI** (*Table 2*) were carried out on a 1.3-μmol scale essentially according to the protocol developed for the synthesis of pyranosyl-RNA (p-RNA) [59]. The standard phosphoramidites were protected with a DMT group at *O*–C(5′), a TOM group at *O*–C(2′), a cyanoethyl group at *O*–P, and an Ac group at N^6 of adenosine. Coupling was performed with 6 equiv. of phosphoramidite in the presence of 80 equiv. of 1-(benzylsulfanyl)-1*H*-tetrazole. Considering the slower detritylation of secondary alcohols, the detritylation of the hept-6-ynofuranosyl units incorporated into the oligonucleotides was performed with 6% $Cl_2CHCOOH$ in $(CH_2Cl)_2$ for 2.5 min. The time for coupling the phosphoramidites to the resulting secondary propargyl alcohols was doubled from 5 to 10 min. These conditions led to coupling yields of 90–95%. The base- and phosphate-protecting groups, and the solid support were cleaved off with 10M $MeNH_2$ in $EtOH/H_2O$ 1:1 at 23° within 2 h. Following this, the TOM and Et_3Si protecting groups were removed by treatment with 1M Bu_4NF (TBAF) · 3 H_2O in THF [3]). The products were treated with *Tris* · HCl buffer (pH 7.4), desalted by chromatography on a *Sephadex* G-10 column, and purified by

[3]) *N*-Methylpyrrolidinone was added to dissolve rA_{14} (**I**) in THF.

chromatography on a *SAX* anion exchange column at pH 11.5 with a NaCl gradient. The chromatograms showed a composition in agreement with the detritylation assay. Finally, the products were desalted on a *RP C-18* column and analyzed by MALDI-TOF mass spectroscopy [60] (*Table 2*).

Table 2. *Synthesis and Characterization of the Oligonucleotides* **I**–**VI**

	Sequence (X = **11A**, Y = **11B**)	Yield		Mol. formula	$[M-H]^-$	
		[b])	[c])		calc.	obs.[a])
I	5′-r(AAAAAAAAAAAAAA)-3′	99%	14%	$C_{140}H_{169}N_{70}O_{82}P_{13}$	4547.1	4547.9
II	5′-r(UUUUUUUUUUUUUU)-3′	88%	5% [d])	$C_{126}H_{155}N_{28}O_{110}P_{13}$	4224.8	4224.7
III	5′-r(UUUUUXUUUUUUUU)-3′	84%	8% [d])	$C_{128}H_{155}N_{28}O_{110}P_{13}$	4248.7	4248.7
IV	5′-r(UXUXUXUXUXUXUU)-3′	64%	13%	$C_{138}H_{155}N_{28}O_{110}P_{13}$	4368.9	4368.9
V	5′-r(UUUUUYUUUUUUUU)-3′	85%	16%	$C_{128}H_{155}N_{28}O_{110}P_{13}$	4224.8	4223.5
VI	5′-r(UYUYUYUYUYUYUU)-3′	67%	8%	$C_{138}H_{155}N_{28}O_{110}P_{13}$	4368.9	4367.3

[a]) By MALDI-TOF mass spectrometry according to [60]. [b]) Calculated from the yield of the coupling steps, as determined by the detritylation assay. [c]) Yield after purification. [d]) Losses during purification.

3. *Pairing Studies.* The melting points of the duplexes consisting of oligonucleotide **I** and the complementary oligonucleotides **II**–**VI** were measured at single strand concentrations of 5 μmol at pH 7 in an aqueous Na_2HPO_4/NaH_2PO_4 buffer and at a 0.15M NaCl concentration (*Table 3*). While the duplex **I**·**V** substituted with one α-L-*talo*-hept-6-ynofuranosyluracil showed the same melting point as the unmodified reference duplex **I**·**II** (20°), the duplex **I**·**III** with one β-D-*allo*-hept-6-ynofuranosyluracil showed a melting point that is 1.5° lower than that of the reference **I**·**II** (*Fig. 3*, *a*).

Table 3. *Melting Points* T_m [°C] *of* **I**·**II**, **I**·**III**, **I**·**IV**, **I**·**V**, *and* **I**·**VI** *Obtained from Temperature-Dependent UV Spectroscopy*

Entry	Duplex	Sequence (X = **11A**, Y = **11B**)	T_m	$T_m - T_m$(**I**·**II**)
1	**I**·**II**	5′-(AAAAAAAAAAAAAA)-3′		
		3′-(UUUUUUUUUUUUUU)-5′	20°	
2	**I**·**III**	5′-(AAAAAAAAAAAAAA)-3′		
		3′-(UUUUUUUUXUUUUU)-5′	18.5°	−1.5°
3	**I**·**IV**	5′-(AAAAAAAAAAAAAA)-3′		
		3′-(UUXUXUXUXUXUXU)-5′	< 0°	> −20°
4	**I**·**V**	5′-(AAAAAAAAAAAAAA)-3′		
		3′-(UUUUUUUUYUUUUU)-5′	20°	0°
5	**I**·**VI**	5′-(AAAAAAAAAAAAAA)-3′		
		3′-(UUYUYUYUYUYUYU)-5′	9°	−11°

The duplex **I**·**VI** substituted with six α-L-*talo*-hept-6-ynofuranosyluracils showed a melting point of *ca.* 9° (*Fig. 3*, *b*). The partially sigmoidal curve allows only to derive an approximate melting point. The corresponding duplex **I**·**IV** substituted with six β-D-*allo*-hept-6-ynofuranosyluracils showed only the upper half of the sigmoidal curve. The melting point of this duplex is estimated to be below 0°.

We also determined the melting behaviour of the single strands. While the absorption spectra of rU_{14} (**II**) shows only a minor dependence on the temperature, the

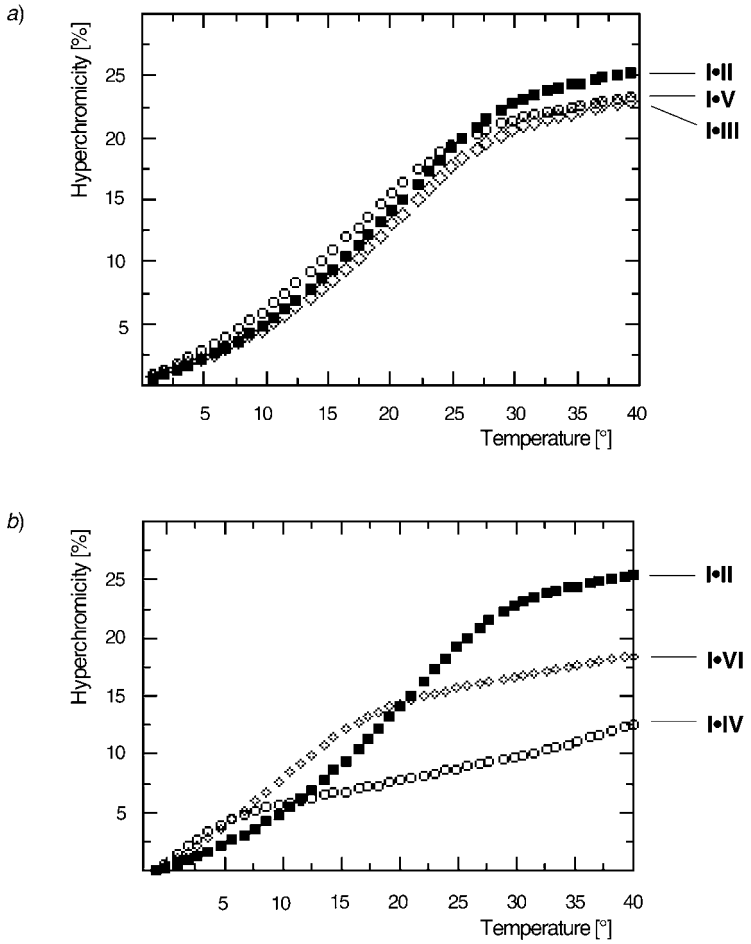


Fig. 3. a) *Temperature-dependent UV spectra* ('melting curves') *of the non-modified and modified* $rA_{14}\cdot rU_{14}$ *duplexes* **I**·**II**, **I**·**III**, *and* **I**·**V**, *and* b) *of the non-modified and modified* $rA_{14}\cdot rU_{14}$ *duplexes* **I**·**II**, **I**·**IV**, *and* **I**·**VI**

absorption spectra of rA_{14} (**I**) depends considerably on the temperature. This explains the additional rise of the hyperchromicity after the melting point.

To see whether the modified duplexes possess the same conformation as the reference duplex **I**·**II**, we measured temperature dependent CD spectra (*Fig. 4*). As the UV spectra, the CD spectra of the rA_{14} strand show a considerable dependence on the temperature while the CD spectra of the rU_{14} strand show almost no temperature dependence. One also observes a much larger ellipticity for rA_{14} than for rU_{14}. The CD spectra of rA_{14} (**I**), the duplex $rA_{14}\cdot rU_{14}$ (**I**·**II**), and the duplex with one *β*-D-*allo*-hept-6-ynofuranosyluracil (**I**·**III**) are similar to each other and differ only by the change of ellipticity between the melting-point temperatures, suggesting that all duplexes adopt the same conformation.

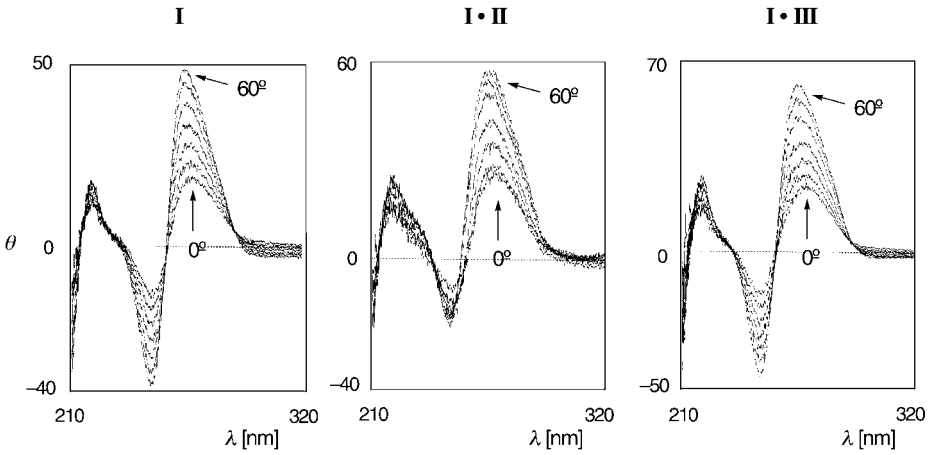


Fig. 4. *CD Spectra of* rA_{14} (**I**), *the duplex* $rA_{14}\cdot rU_{14}$ (**I**·**II**), *and the modified duplex* **I**·**III**

According to the temperature-dependent UV and CD spectra, incorporation of one α-L-*talo*-hept-6-ynofuranosyluracil does not destabilize the resulting duplex, while incorporation of one β-D-*allo*-hept-6-ynofuranosyluracil caused a slight destabilization. The incorporation of six hept-6-ynofuranosyluracils, however, strongly destabilizes the corresponding duplexes, and this much more so upon incorporating β-D-*allo*- than α-L-*talo*-residues, in agreement with the results of the conformational analysis and the relative importance of the different hydration sites.

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Experimental Part

General. See [2]. Ion-exchange HPLC: *Macherey-Nagel Nucleogel-SAX 1000–8/77* (anal.) and *1000–8/46* (prep.); eluent *A*, 10 mM Na_2HPO_4 in H_2O (pH 11.5); eluent *B*, 10 mM Na_2HPO_4/1M NaCl in H_2O (pH 11.5; detection at 260 nm, elution at 23°. MS: fast-atom-bombardment (FAB; 3-nitrobenzyl alcohol (NOBA)), or matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF; 0.05M 1*H*-indole-3-acrylic acid (IAA) in THF or 0.05M α-cyano-4-hydroxycinnamic acid (CCA) in MeCN/EtOH/H_2O), and high-resolution MALDI (HR-MALDI; 0.05M 2,5-dihydroxybenzoic acid (DHB) in THF). All measurements with oligonucleotides were made in a phosphate buffer (10 mM aq. NaH_2PO_4 buffer containing 150 mM NaCl at pH 7.0) with a total oligonucleotide concentration of 5 μM. Concentrations of oligonucleotide solns. were calculated from the UV absorbance of the soln. at 260 nm and pH 7 using the following molar extinction coefficients: $\varepsilon(r(A)) = 15100$, $\varepsilon(r(U)) = 9900$.

1-[6,7-Dideoxy-2,3-O-*isopropylidene-5*-O-(*2-nitrobenzoyl*)-*7*-C-*(triethylsilyl)-β*-D-allo-*hept-6-ynofuranosyl]-uracil* (**2A**). A soln. of pyridine (1.7 ml, 21.1 mmol), DMAP (0.15 g, 1.2 mmol), and **1A** [4] (2.9 g, 6.9 mmol) in CH_2Cl_2 (70 ml) was treated dropwise with 2-nitrobenzoyl chloride (1.04 ml, 7.9 mmol), stirred at reflux for 24 h, diluted with CH_2Cl_2 (30 ml), washed with sat. aq. NH_4Cl soln., dried (Na_2SO_4), and evaporated. FC (AcOEt/hexane 1:2 → 1:1) gave **2A** (3.06 g 78%). Colourless foam. R_f (AcOEt/hexane 1:1) 0.32. $[\alpha]_D^{25} = -60.2$ (c = 0.82, $CHCl_3$). UV (MeOH): 258 (12200). IR ($CHCl_3$): 3387*w*, 2958*w*, 2876*w*, 2200*w*, 1755*m*, 1728*m*, 1695*s*, 1645*w*, 1537*m*, 1457*w*, 1385*w*, 1351*w*, 1251*w*, 1156*w*, 1093*m*, 1070*m*, 1017*w*, 972*w*, 857*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 4*; additionally, 8.65 (br. *s*, NH); 7.95–7.90 (*m*, 1 arom. H); 7.70–7.63 (*m*, 3 arom. H); 1.59, 1.37 (2*s*, Me_2C); 1.00 (*t*, J = 7.8, ($MeCH_2$)$_3$Si); 0.64 (*q*, J = 7.8, ($MeCH_2$)$_3$Si). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 5*; additionally, 162.9 (*s*, OC=O); 148.7 (*s*, C(2) of 2-NO_2Bz); 133.3 (*d*, C(5) of 2-NO_2Bz); 132.8 (*d*, C(4) of 2-NO_2Bz); 130.6 (*d*, C(6) of 2-NO_2Bz); 126.5 (*s*, C(1) of 2-NO_2Bz); 124.2 (*d*, C(3) of 2-NO_2Bz); 115.4 (*s*, Me_2C);

Table 4. *Selected ^{1}H-NMR Chemical Shifts* [ppm] *and Coupling Constants* [Hz] *of the* D-allo-*Hept-6-ynofuranosyluridines* **2A**–**11A** *in $CDCl_3$ Solution*

	2A	**3A**	**4A**	**5A**	**6A**	**7A**	**8A**	**9A**	**10A**	**11A**[a])
H–C(5)	5.51	5.66	5.89	5.76	5.67	5.96	5.46	5.28	5.26	5.51/5.46
H–C(6)	7.33	7.66	[b])	[b])	[b])	[b])	7.77	7.60	[b])	7.66
H–C(1′)	5.92	5.87	6.47	6.48	6.53	6.48	5.84	6.13	5.99	6.18/6.14
H–C(2′)	4.75	4.44	5.65	5.62	5.72	5.79	4.28	4.20	4.25[c])	4.38/4.34
H–C(3′)	4.92	4.36	5.94	5.90	6.15	6.11	4.34	4.51[d])	4.51	4.34
H–C(4′)	4.46	4.24	4.73	4.70	4.74	4.56	3.98	3.99	4.07	4.13/4.06
H–C(5′)	5.83	5.91	6.12	6.11	6.13	4.86[e])	4.15	4.08	4.03	4.20/4.03
J(5,6)	7.9	8.0	8.1	8.1	8.3	8.3	8.3	8.3	8.2	8.1/8.1
J(1′,2′)	3.7	3.6	7.2	7.2	7.5	7.6	5.0	6.2	7.5	6.3/6.0
J(2′,3′)	6.6	5.8	6.5	6.2	5.8	5.7	6.0	5.4	5.9	5.8/5.8
J(3′,4′)	3.5	4.8	3.1	3.4	2.5	1.8	3.4	2.9	2.1	2.7/3.0
J(4′,5′)	3.3	2.9	2.8	2.8	2.9	3.0	2.8	2.5	2.7	2.7/2.8

[a]) 3 : 2 Mixture of diastereoisomers. [b]) Signal hidden by the signals of the aromatic H-atoms at 7.0–8.0 ppm. [c]) δ(HO–C(2′)) = 3.72 ppm, J(2′,OH) = 8.0 Hz. [d]) δ(HO–C(3′)) = 2.99 ppm, J(3′,OH) = 2.8 Hz. [e]) δ(HO–C(5′)) = 3.46 ppm, J(5′,OH) = 3.7 Hz.

Table 5. *Selected ^{13}C-NMR Chemical Shifts* [ppm] *of the* D-allo-*Hept-6-ynofuranosyluridines* **2A**–**5A** *and* **7A**–**11A** *in $CDCl_3$ Solution*

	2A	**3A**	**4A**	**5A**	**7A**	**8A**	**9A**	**10A**	**11A**[a])
C(2)	163.8	164.4	162.0	163.7	162.3	164.0	163.3	162.6	162.8
C(4)	150.1	151.8	149.7	150.7	150.0	151.8	150.6	150.4	150.2/150.0
C(5)	102.7	102.9	103.6	103.7	103.6	102.5	102.6	102.5	102.5
C(6)	140.3	140.3	148.7	139.7	140.3	140.7	140.5	140.1	140.5/140.3
C(1′)	92.0	89.6	86.4	86.2	86.8	89.7	86.8	87.4	86.2/86.1
C(2′)	84.0	74.8	73.2	73.0	73.9	75.3	82.5	73.5	77.8
C(3′)	80.5	70.9	70.7	70.6	71.6	71.0	70.5	78.9	71.7/70.4
C(4′)	85.3	84.3	82.6	82.5	85.9	87.2	86.3	85.8	85.9/85.7
C(5′)	66.1	66.6	65.8	65.9	63.0	65.5	65.8	65.3	65.7/65.6
C(6′)	98.1	98.7	98.1	98.1	102.5	103.4	103.2	103.1	103.5/103.2
C(7′)	96.2	93.1	93.9	93.7	91.6	91.6	91.6	91.4	91.8/91.1

[a]) 3 : 2 Mixture of diastereoisomers.

27.1, 25.1 (2*q*, *Me*$_2$C); 7.3 (*q*, (*Me*CH$_2$)$_3$Si); 4.1 (*t*, (MeCH$_2$)$_3$Si). HR-MALDI-MS (DHB): 594.188 ([*M* + Na]$^+$, $C_{27}H_{33}N_3NaO_9Si^+$; calc. 594.187). Anal calc. for $C_{27}H_{33}N_3O_9Si$ (571.66): C 56.73, H 5.82, N 7.35; found: C 56.53, H 5.82, N 7.32.

*1-[6,7-Dideoxy-5-*O*-(2-nitrobenzoyl)-7-*C*-(triethylsilyl)-β-*D*-allo-hept-6-ynofuranosyl]uracil* (**3A**). A suspension of **2A** (3.0 g, 5.2 mmol) in H_2O (35 ml) was treated with CF_3COOH (35 ml), stirred at 80° for 1 h, diluted with H_2O (180 ml), and extracted with CH_2Cl_2. The combined org. layers were washed with brine, dried (Na_2SO_4), and evaporated to give crude **3A** (3.1 g, quant.). Red foam. R_f (AcOEt/MeOH 20 : 1) 0.56. $[\alpha]_D^{25} = -43.9$ ($c = 0.7$, $CHCl_3$). UV (MeOH): 259 (11500). IR ($CHCl_3$): 3391*w*, 2958*w*, 2876*w*, 2185*w*, 1745*m*, 1693*s*, 1640*w*, 1537*m*, 1458*w*, 1350*m*, 1252*m*, 1114*m*, 1068*m*, 1005*w*, 964*w*, 847*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 4*; additionally 9.73 (br. *s*, NH); 7.97–7.92 (*m*, 1 arom. H); 7.74–7.66 (*m*, 3 arom. H); 3.9–3.3 (br. *s*, 2 OH); 0.92 (*t*, $J = 8.1$, (*Me*CH$_2$)$_3$Si); 0.58 (*q*, $J = 8.1$, (MeC*H*$_2$)$_3$Si). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 5*; additionally, 162.9 (*s*, OC=O); 148.7 (*s*, C(2) of 2-NO_2Bz); 133.3 (*d*, C(5) of 2-NO_2Bz); 132.6 (*d*, C(4) of 2-NO_2Bz); 130.2 (*d*, C(6) of 2-NO_2Bz); 127.1 (*s*, C(1) of 2-NO_2Bz); 124.5 (*d*, C(3) of 2-NO_2Bz); 7.5 (*q*, (*Me*CH$_2$)$_3$Si); 4.1 (*t*, (MeCH$_2$)$_3$Si). HR-MALDI-MS (DHB): 554.157 ([*M* + Na]$^+$, $C_{24}H_{29}N_3NaO_9Si^+$; calc. 554.157). Anal. calc. for $C_{24}H_{29}N_3O_9Si$ (531.59): C 54.23, H 5.50, N 7.90; found: C 54.04, H 5.56, N 7.82.

Benzoylation of **3A**. A soln. of pyridine (40 ml), 4-pyrrolidinopyridine (0.62 g, 4.2 mmol), and **3A** (2.2 g, 4.15 mmol) in CH_2Cl_2 (40 ml) was treated dropwise with BzCl (2.4 ml, 21 mmol), stirred at 23° for 24 h, diluted with CH_2Cl_2, washed with brine, dried (Na_2SO_4), and evaporated. FC (AcOEt/hexane 1:4 → 1:2) gave **4A** (2.7 g, 78%) and **5A** (164 mg, 5%).

3-Benzoyl-1-[2,3-di-O-*benzoyl-6,7-dideoxy-5*-O-(*2-nitrobenzoyl*)-7-C-*(triethylsilyl)-β*-D-allo-*hept-6-ynofuranosyl]uracil* (**4A**). Colourless foam. R_f (AcOEt/hexane 1:1) 0.66. $[\alpha]_D^{25} = -64.2$ ($c = 1.1$, $CHCl_3$). UV (MeOH): 254 (19200). IR ($CHCl_3$): 3032*w*, 2958*w*, 2876*w*, 2160*w*, 1732*s*, 1718*m*, 1678*s*, 1601*w*, 1584*w*, 1537*m*, 1450*m*, 1375*m*, 1350*m*, 1316*m*, 1265*s*, 1122*m*, 1107*m*, 1069*m*, 1025*w*, 973*w*, 920*w*, 847*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 4*; additionally, 8.31–7.23 (*m*, 19 arom. H, H–C(6)); 1.00 (*t*, $J = 8.1$, $(MeCH_2)_3Si$); 0.65 (*q*, $J = 8.1$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 5*; additionally, 168.5 (*s*, NC=O); 165.6, 165.4, 164.1 (3*s*, 3 OC=O); 148.3 (*s*, C(2) of 2-NO_2Bz); 135.3, 134.1, 134.0 (3*d*, C(4) of 3 Bz); 133.4 (*d*, C(4) of 2-NO_2Bz); 132.7 (*d*, C(5) of 2-NO_2Bz); 131.5 (*s*, C(1) of BzN); 130.7 (*d*, C(2) and C(6) of BzN); 130.3 (*d*, C(6) of 2-NO_2Bz); 130.2, 130.1 (2*d*, C(2) and C(6) of 2 BzO); 129.35 (*d*, C(3) and C(5) of BzN); 129.30 (*s*, C(1) of BzO); 128.9, 128.7 (2*d*, C(3) and C(5) of 2 BzO); 128.4 (*s*, C(1) of BzO); 126.7 (*s*, C(1) of 2-NO_2Bz); 124.4 (*d*, C(3) of 2-NO_2Bz); 7.4 (*q*, $(MeCH_2)_3Si$); 4.0 (*t*, $(MeCH_2)_3Si$). HR-MALDI-MS (DHB): 866.235 ($[M+Na]^+$, $C_{45}H_{41}N_3NaO_{12}Si^+$; calc. 866.236). Anal. calc. for $C_{45}H_{41}N_3O_{12}Si$ (843.92): C 64.05, H 4.90, N 4.98; found: C 64.23, H 5.16, N 4.73.

1-[2,3-Di-O-*benzoyl-6,7-dideoxy-5*-O-(*2-nitrobenzoyl*)-7-C-*(triethylsilyl)-β*-D-allo-*hept-6-ynofuranosyl]uracil* (**5A**). Colourless foam. R_f (AcOEt/hexane 1:1) 0.36. $[\alpha]_D^{25} = -55.9$ ($c = 0.8$, $CHCl_3$). UV (MeOH): 248 (20200). IR ($CHCl_3$): 3384*w*, 3032*w*, 2959*w*, 2871*w*, 2160*w*, 1732*s*, 1698*s*, 1602*w*, 1584*w*, 1537*m*, 1452*m*, 1350*m*, 1316*w*, 1261*s*, 1122*m*, 1107*m*, 1068*m*, 1025*w*, 973*w*, 847*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 4*; additionally, 8.65 (br. *s*, NH); 8.10–7.23 (*m*, 14 arom. H, H–C(6)); 1.00 (*t*, $J = 8.1$, $(MeCH_2)_3Si$); 0.65 (*q*, $J = 8.1$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 5*; additionally, 165.4, 165.3, 164.1 (3*s*, 3 OC=O); 148.3 (*s*, C(2) of 2-NO_2BzO); 134.0, 133.9 (2*d*, C(4) of 2 BzO); 133.5 (*d*, C(4) of 2-NO_2BzO); 132.6 (*d*, C(5) of 2-NO_2BzO); 130.4 (*d*, C(6) of 2-NO_2BzO); 130.2 (*d*, C(2) and C(6) of 2 BzO); 128.9 (*s*, C(1) of BzO); 128.8, 128.7 (2*d*, C(3) and C(5) of 2 BzO); 128.5 (*s*, C(1) of BzO); 126.8 (*s*, C(1) of 2-NO_2BzO); 124.4 (*d*, C(3) of 2-NO_2BzO); 7.3 (*q*, $(MeCH_2)_3Si$); 4.0 (*t*, $(MeCH_2)_3Si$).

Partial Deprotection of **4A**. *a*) A soln. of **4A** (1.0 g, 1.19 mmol) in H_2O (15 ml) and MeCN (15 ml) was treated at 23° with four batches of NH_4Cl (0.25 g, 4.7 mmol) and Zn powder (0.16 g, 2.4 mmol) in intervals of 20 min. The suspension was stirred for 7 h and filtered through silica gel (elution with AcOEt). The filtrate was washed with sat. aq. $NaHCO_3$ soln. and brine, dried (Na_2SO_4), and evaporated. FC (toluene/MeOH 100:1 → 10:1) gave **7A** (1.8 g, 62%).

b) As in *a*), but heating after 3 h to 80° and stirring at 80° for 2 h. FC (toluene/MeOH 100:1) gave **6A** (4%) and crude **7A** (*ca.* 24%) beside several decomposition products.

3-Benzoyl-1-[5-O-*(2-aminobenzoyl)2,3-di*-O-*benzoyl-6,7-dideoxy-7*-C-*(triethylsilyl)-β*-D-talo-*hept-6-ynofuranosyl]uracil* (**6A**). Colourless foam. R_f (toluene/MeOH 4:1) 0.54. ^{1}H-NMR (200 MHz, $CDCl_3$): see *Table 4*; additionally, 8.07–7.98 (*m*, 3 arom. H,); 7.92–7.76 (*m*, 2 arom. H); 7.61–7.22 (*m*, 11 arom. H, H–C(6)); 6.84–6.71 (*m*, 2 arom. H); 5.79 (br. *s*, exchanged with CD_3OD, NH_2); 0.95 (*t*, $J = 8.0$, $(MeCH_2)_3Si$); 0.60 (*q*, $J = 8.0$, $(MeCH_2)_3Si$). HR-MALDI-MS (DHB): 836.259 ($[M+Na]^+$, $C_{45}H_{43}N_3NaO_{16}Si^+$; calc. 836.261).

3-Benzoyl-1-[2,3-di-O-*benzoyl-6,7-dideoxy-7*-C-*(triethylsilyl)-β*-D-allo-*hept-6-ynofuranosyl]uracil* (**7A**). Colourless foam. R_f (AcOEt/hexane 1:2) 0.38. $[\alpha]_D^{25} = -76.4$ ($c = 1.1$, $CHCl_3$). UV (MeOH): 252 (19100). IR ($CHCl_3$): 3350*w* (br.), 3035*w*, 2957*w*, 2876*w*, 2280*w*, 1755*m*, 1730*s*, 1711*m*, 1675*s*, 1601*w*, 1450*m*, 1378*m*, 1316*w*, 1270*s*, 1126*m*, 1092*m*, 1070*m*, 1024*w*, 974*w*, 921*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 4*; additionally, 8.80 (br. *s*, NH); 8.08–7.21 (*m*, 15 arom. H, H–C(6)); 0.94 (*t*, $J = 7.8$, $(MeCH_2)_3Si$); 0.59 (*q*, $J = 7.8$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 5*; additionally, 168.7 (*s*, NC=O); 165.8, 165.3 (2*s*, 2 OC=O); 135.3 (*d*, C(4) of BzN); 134.0, 133.9 (2*d*, C(4) of 2 BzO); 131.6 (*s*, C(1) of BzN); 130.8 (*d*, C(2) and C(6) of BzN); 130.12, 130.09 (2*d*, C(2) and C(6) of 2 BzO); 129.4 (*d*, C(3) and C(5) of BzN); 129.3 (*s*, C(1) of BzO); 128.9, 128.7 (2*d*, C(3) and C(5) of 2 BzO); 128.6 (*s*, C(1) of BzO); 7.4 (*q*, $(MeCH_2)_3Si$); 4.1 (*t*, $(MeCH_2)_3Si$). HR-MALDI-MS (DHB): 717.224 ($[M+Na]^+$, $C_{38}H_{38}N_2NaO_9Si^+$; calc. 717.224). Anal. calc. for $C_{38}H_{38}N_2O_9Si$ (694.81): C 65.69, H 5.51, N 4.03; found: C 65.51, H 5.63, N 4.16.

1-[6,7-Dideoxy-5-O-*(4,4′-dimethoxytrityl)-7*-C-*(triethylsilyl)-β*-D-allo-*hept-6-ynofuranosyl]uracil* (**8A**). A soln. of 4,4′-dimethoxytrityl chloride (DMTCl; 1.0 g, 2.95 mmol) in CH_2Cl_2 (11 ml) was treated with *sym*-collidine (2.4 ml), stirred at 23° for 15 min, treated with $AgNO_3$ (0.5 g, 2.95 mmol), and stirred for 1 h. An aliquot (8.2 ml, 1.80 mmol of the reagent) of this suspension was filtered into a soln. of **7A** (500 mg, 0.72 mmol) in CH_2Cl_2 (5 ml). The mixture was stirred at 23° for 30 min and filtered through silica gel (elution with AcOEt/Et_3N 99:1). The filtrate was evaporated. A soln. of the residue in THF/MeOH/H_2O 5:4:1 (50 ml) was cooled to

0°, treated dropwise with an aq. 10M NaOH soln. (0.72 ml, 7.2 mmol), stirred at 0°, treated with sat. aq. NH_4Cl soln., and extracted with AcOEt. The combined org. layers were dried (Na_2SO_4), filtered, and evaporated. FC (hexane/AcOEt/Et_3N 1:1:0.01 → 1:2:0.01) gave **8A** (0.29 g, 59%). Colourless foam. R_f (AcOEt/hexane 1:1) 0.12. $[\alpha]_D^{25} = -59.5$ ($c = 0.9$, $CHCl_3$). UV (MeOH): 257 (14500). IR ($CHCl_3$): 3391*w*, 3005*w*, 2957*m*, 2875*w*, 2838*w*, 2280*w*, 1691*s*, 1608*m*, 1509*s*, 1463*m*, 1395*w*, 1300*m*, 1252*s*, 1110*w*, 1092*w*, 1070*w*, 1036*m*, 975*w*, 910*w*, 829*m*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 4*; additionally, 7.51–7.17 (*m*, 9 arom. H); 6.82 (*d*, $J = 9.0$, 4 arom. H); 3.78, 3.77 (2*s*, 2 MeO); 0.88 (*t*, $J = 7.8$, $(MeCH_2)_3Si$); 0.46 (*q*, $J = 7.8$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 5*; additionally, 159.1, 159.07 (2*s*, C(4) of 2 $MeOC_6H_4$); 145.4 (*s*, C(1) of Ph); 135.9, 135.6 (2*s*, C(1) of 2 $MeOC_6H_4$); 130.9 (*d*, C(2) and C(6) of 2 $MeOC_6H_4$); 128.6 (*d*, C(2) and C(6) of Ph); 128.0 (*d*, C(3) and C(5) of Ph); 127.2 (*d*, C(4) of Ph); 113.4, 113.3 (2*d*, C(3) and C(5) of 2 $MeOC_6H_4$); 88.4 (*s*, Ar_3C); 55.3 (*q*, 2 MeO); 7.4 (*q*, $(MeCH_2)_3Si$); 4.0 (*t*, $(MeCH_2)_3Si$). Anal. calc. for $C_{38}H_{44}N_2O_8Si$ (684.86): C 66.64, H 6.48, N 4.09; found: C 66.50, H 6.54, N 4.12.

(Silyloxy)methylation of **8A**. A soln. of **8A** (0.68 g, 0.99 mmol) and $(i\text{-}Pr)_2NEt$ (0.85 ml, 4.97 mmol) in $(CH_2Cl)_2$ (4 ml) was treated with Bu_2SnCl_2 (0.33 g, 1.09 mmol), stirred for 90 min at 23°, treated with (triisopropylsilyl)oxymethyl chloride (TOMCl; 0.26 g, 1.19 mmol), heated to 80°, stirred for 70 min, diluted with sat. aq. $NaHCO_3$ soln., and extracted with CH_2Cl_2. The org. phase was washed with brine, dried (Na_2SO_4), and evaporated. FC (hexane/AcOEt/Et_3N 4:1:0.01 → 2:1:0.01) gave **9A** (284 mg, 33%), **10A** (122 mg, 16%), and **8A** (189 mg, 28%).

*1-(6,7-Dideoxy-5-*O*-(4,4′-dimethoxytrityl)-7-*C*-(triethylsilyl)-2-*O*-{[(triisopropylsilyl)oxy]methyl}-β-*D*-allo-hept-6-ynofuranosyl)uracil* (**9A**). Colourless foam. R_f (AcOEt/hexane 1:2) 0.29. $[\alpha]_D^{25} = -36.8$ ($c = 0.9$, $CHCl_3$). UV (MeOH): 257 (12900). IR ($CHCl_3$): 3391*w*, 3005*w*, 2954*m*, 2870*m*, 2180*w*, 1693*s*, 1608*w*, 1509*m*, 1461*m*, 1386*w*, 1255*m*, 1079*m*, 1036*m*, 1015*m*, 882*w*, 827*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 4*; additionally, 8.08 (br. *s*, NH); 7.51–7.21 (*m*, 9 arom. H); 6.81–6.76 (*m*, 4 arom. H); 5.20, 4.93 (2*d*, $J = 4.7$, OCH_2O); 3.79 (*s*, 2 MeO); 1.13–1.08 (*m*, $(Me_2CH)_3Si$); 0.88 (*t*, $J = 7.5$, $(MeCH_2)_3Si$); 0.45 (*q*, $J = 7.5$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 5*; additionally 159.2, 159.1 (2*s*, C(4) of 2 $MeOC_6H_4$); 145.4 (*s*, C(1) of Ph); 135.8, 135.4 (2*s*, C(1) of 2 $MeOC_6H_4$); 131.0 (*d*, C(2) and C(6) of 2 $MeOC_6H_4$); 128.6 (*d*, C(2) and C(6) of Ph); 128.0 (*d*, C(3) and C(5) of Ph); 127.3 (*d*, C(4) of Ph); 113.4, 113.3 (2*d*, C(3) and C(5) of 2 $MeOC_6H_4$); 91.1 (*t*, OCH_2O); 88.6 (*s*, Ar_3C); 55.3 (*q*, 2 MeO); 17.85 (*q*, $(Me_2CH)_3Si$); 11.9 (*d*, $(Me_2CH)_3Si$); 7.4 (*q*, $(MeCH_2)_3Si$); 4.0 (*t*, $(MeCH_2)_3Si$).

*1-(6,7-Dideoxy-5-*O*-(4,4′-dimethoxytrityl)-7-*C*-(triethylsilyl)-3-*O*-{[(triisopropylsilyl)oxy]methyl}-β-*D*-allo-hept-6-ynofuranosyl)uracil* (**10A**). Colourless foam. R_f (AcOEt/hexane 1:2) 0.14. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 4*; additionally, 7.51–7.21 (*m*, 9 arom. H, H–C(6)); 6.85–6.75 (*m*, 4 arom. H); 5.14, 4.94 (2*d*, $J = 4.8$, OCH_2O); 3.792, 3.790 (2*s*, 2 MeO); 1.15–1.08 (*m*, $(Me_2CH)_3Si$); 0.87 (*t*, $J = 7.5$, $(MeCH_2)_3Si$); 0.45 (*q*, $J = 7.5$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 5*; additionally, 158.9, 158.7 (2*s*, C(4) of 2 $MeOC_6H_4$); 145.1 (*s*, C(1) of Ph); 135.5, 135.0 (2*s*, C(1) of 2 $MeOC_6H_4$); 130.7, 130.69 (2*d*, C(2) and C(6) of 2 $MeOC_6H_4$); 128.3 (*d*, C(2) and C(6) of Ph); 127.8 (*d*, C(3) and C(5) of Ph); 127.1 (*d*, C(4) of Ph); 113.2, 113.1 (2*d*, C(3) and C(5) of 2 $MeOC_6H_4$); 91.2 (*t*, OCH_2O); 88.5 (*s*, Ar_3C); 55.24, 55.21 (2*q*, 2 MeO); 17.8 (*q*, $(Me_2CH)_3Si$); 11.9 (*d*, $(Me_2CH)_3Si$); 7.4 (*q*, $(MeCH_2)_3Si$); 4.0 (*t*, $(MeCH_2)_3Si$).

*1-(6,7-Dideoxy-5-*O*-(4,4′-dimethoxytrityl)-7-*C*-(triethylsilyl)-2-*O*-{[(triisopropylsilyl)oxy]methyl}-β-*D*-allo-hept-6-ynofuranosyl)uracil 3-[(2-Cyanoethyl) (Diisopropyl)phosphoramidite]* (**11A**). A soln. of **9A** (165 mg, 0.184 mmol) and $(i\text{-}Pr)_2NEt$ (0.08 ml, 0.47 mmol) in CH_2Cl_2 (1.2 ml) was treated dropwise with 2-cyanoethyl (diisopropyl)phosphoramidochloridite (0.051 ml, 0.23 mmol) and stirred at 23° for 24 h. Evaporation and FC (hexane/AcOEt (1% Et_3N) 3:1 → 1:1) gave **11A** (160 mg, 81%). Colourless foam. R_f (AcOEt/hexane 1:2) 0.39 and 0.34 (2 diastereoisomers). ^{1}H-NMR (500 MHz, $CDCl_3$; 3:2 mixture of diastereoisomers): see *Table 4*; additionally, 7.50–7.20 (*m*, 9 arom. H); 6.83–6.79 (*m*, 4 arom. H); 5.00 (*d*, $J = 5.1$, 0.6 H), 4.96 (*d*, $J = 5.1$, 0.4 H), 4.94 (*d*, $J = 5.2$, 0.6 H), 4.85 (*d*, $J = 5.2$, 0.4 H) (OCH_2O); 3.93–3.80 (*m*, $NCCH_2CH_2O$); 3.792, 3.790, 3.785, 3.782 (4*s*, 2 MeO); 3.69–3.62 (*m*, $NCCH_2CH_2O$); 3.59–3.50 (*m*, $(Me_2CH)_2N$); 2.67–2.61 (*m*, NCCH); 2.35 (*t*, $J = 6.7$, NCCH); 1.20–1.00 (*m*, $(Me_2CH)_2N$, $(Me_2CH)_3Si$); 0.88 (*t*, $J = 8.1$, $(MeCH_2)_3Si$); 0.46 (*q*, $J = 8.1$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$; 3:2 mixture of diastereoisomers): see *Table 5*; additionally, 158.75, 158.72 (2*s*, C(4) of 2 $MeOC_6H_4$); 145.14, 145.11 (2*s*, C(1) of Ph); 135.8, 135.7, 135.5, 135.3 (4*s*, C(1) of 2 $MeOC_6H_4$); 130.67 (2 C), 130.66, 130.62 (3*d*, C(2) and C(6) of 2 $MeOC_6H_4$); 128.45, 128.36 (2*d*, C(2) and C(6) of Ph); 127.75, 127.71 (2*d*, C(3) and C(5) of Ph); 126.95 (*d*, C(4) of Ph); 117.71, 117.20 (2*s*, CN); 113.10 (2 C), 113.03, 112.99 (3*d*, C(3) and C(5) of 2 $MeOC_6H_4$); 89.48 (*dt*, ^5J(C,P) = 4.5, OCH_2O); 89.04 (*dt*, ^5J(C,P) = 2.8, OCH_2O); 88.14, 88.05 (2*s*, Ar_3C); 59.0, 57.9 (2*dt*, ^2J(C,P) = 17, $NCCH_2CH_2O$); 55.21, 55.20 (2*q*, 2 MeO); 43.40, 43.10 (2*dd*, ^2J(C,P) = 13, $(Me_2CH)_2N$); 24.61, 24.55 (2 C), 24.48 (3*q*, $(Me_2CH)_2N$); 20.40, 19.97 (2*dt*, ^3J(C,P) = 5.7, $NCCH_2CH_2O$); 17.80, 17.79, 17.74, 17.73 (4*q*, $(Me_2CH)_3Si$); 11.93, 11.89 (2*d*, $(Me_2CH)_3Si$); 7.41, 7.37 (2*q*,

(*Me*CH_2)$_3$Si); 4.02, 3.96 (2*t*, (Me*C*H_2)$_3$Si). ^{31}P-NMR (121 MHz, $CDCl_3$; 3:2 mixture of diastereoisomers): 151.3 (0.4 P); 150.0 (0.6 P). FAB-MS (NOBA): 1071.6 (11, $[M+H]^+$), 303.1 (100, $DMTr^+$).

*1-[6,7-Dideoxy-2,3-*O*-isopropylidene-5-*O*-(2-nitrobenzoyl)-7-*C*-(triethylsilyl)-α-*L*-talo-hept-6-ynofuranosyl]-uracil* (**2T**). A soln. of pyridine (3.5 ml, 43.5 mmol), DMAP (0.30 g, 2.4 mmol), and **1T** [4] (5.9 g, 14.0 mmol) in CH_2Cl_2 (65 ml) was treated dropwise with 2-nitrobenzoyl chloride (2.12 ml, 16.1 mmol), stirred at 23° for 60 h, diluted with CH_2Cl_2 (30 ml), washed with sat. aq. NH_4Cl soln., dried (Na_2SO_4), and evaporated. FC (AcOEt/hexane 2:3 → 1:1) gave **2T** (7.42 g 93%). Colourless foam. R_f (AcOEt/hexane 1:1) 0.32. $[\alpha]_D^{25} = +26.6$ (c = 0.49, $CHCl_3$). UV (MeOH): 257 (11800). IR ($CHCl_3$): 3375*w*, 2957*w*, 2850*w*, 2160*w*, 1750*m*, 1695*s*, 1630*w*, 1537*m*, 1457*w*, 1385*w*, 1351*m*, 1272*m*, 1150*w*, 1115*w*, 1092*m*, 1067*m*, 1005*w*, 961*w*, 859*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 6*; additionally, 8.44 (br. *s*, NH); 7.96–7.91 (*m*, 1 arom. H); 7.70–7.61 (*m*, 3 arom. H); 1.57, 1.36 (2*s*, Me_2C); 1.01 (*t*, J = 7.8, (*Me*CH_2)$_3$Si); 0.65 (*q*, J = 7.8, (MeCH_2)$_3$Si). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 7*; additionally, 164.0 (*s*, OC=O); 148.1 (*s*, C(2) of 2-NO_2Bz); 133.4 (*d*, C(5) of 2-NO_2Bz); 132.4 (*d*, C(4) of 2-NO_2Bz); 129.9 (*d*, C(6) of 2-NO_2Bz); 127.2 (*s*, C(1) of 2-NO_2Bz); 124.3 (*d*, C(3) of 2-NO_2Bz); 114.4 (*s*, Me_2*C*); 26.9, 25.0 (2*q*, *Me*$_2$C); 7.3 (*q*, (*Me*CH_2)$_3$Si); 4.1 (*t*, (Me*C*H_2)$_3$Si). HR-MALDI-MS (DHB): 594.188 ($[M+Na]^+$, $C_{27}H_{33}N_3NaO_9Si^+$; calc. 594.189). Anal. calc. for $C_{27}H_{33}N_3O_9Si$ (571.66): C 56.73, H 5.82, N 7.35; found: C 56.90, H 5.88, N 7.31.

Table 6. *Selected ^{1}H-NMR Chemical Shifts* [ppm] *and Coupling Constants* [Hz] *of the* L-talo-*Hept-6-ynofuranosyluridines* **2T**–**4T** *and* **7T**–**11T** *in $CDCl_3$ Solution*

	2T	**3T**	**4T**	**7T**	**8T**	**9T**	**10T**	**11T**[a])
H–C(5)	5.64	5.65	5.94	5.95	5.65	5.64	5.66	5.61/5.60
H–C(6)	7.47	7.63	[b])	[b])	7.66	7.73	7.55	7.66/7.62
H–C(1′)	5.72	5.84	6.44	6.47	5.75	6.09	5.88	6.08/6.12
H–C(2′)	4.94	4.36	5.74	5.74	4.29	4.10	4.14[c])	4.33/4.38
H–C(3′)	4.98	4.36	5.97	5.86	4.48	4.64[d])	4.57	4.68/4.61
H–C(4′)	4.48	4.26	4.69	4.49	3.64	3.64	3.43	3.94/3.91
H–C(5′)	5.89	5.95	6.03	4.83[e])	4.24	4.29	4.31	4.28/4.22
J(5,6)	8.0	8.2	8.1	8.1	8.1	8.4	8.1	8.2/8.2
J(1′,2′)	2.2	4.1	7.2	7.5	6.2	7.8	8.1	8.2/8.2
J(2′,3′)	6.6	[f])	6.1	5.9	5.7	5.4	6.2	5.3/5.1
J(3′,4′)	2.2	5.6	2.5	1.9	2.2	1.8	1.3	2.7/3.0
J(4′,5′)	6.5	5.6	5.0	3.0	4.7	4.4	4.3	4.3/3.9

[a]) 3:2 Mixture of diastereoisomers. [b]) Signal hidden by the signals of the aromatic H-atoms at 7.0–8.0 ppm. [c]) δ(HO–C(2′)) = 3.56 ppm, J(2′,OH) = 7.8 Hz. [d]) δ(HO–C(3′)) = 2.96 ppm, J(3′,OH) = 1.6 Hz. [e]) δ(HO–C(5′)) = 3.15 ppm, J(5′,OH) = 3.8 Hz. [f]) Not assigned.

Table 7. *Selected ^{13}C-NMR Chemical Shifts* [ppm] *of the* L-talo-*Hept-6-ynofuranosyluridines* **2T**–**4T** *and* **7T**–**11T** *in $CDCl_3$ Solution*

	2T	**3T**	**4T**	**7T**	**8T**	**9T**	**10T**	**11T**[a])
C(2)	163.9	164.3	162.1	162.3	163.8	163.2	163.3	162.7
C(4)	150.5	151.8	149.8	150.0	151.8	150.6	151.0	150.2/150.3
C(5)	102.3	102.8	103.7	103.5	102.7	103.2	103.2	102.7/102.8
C(6)	142.3	140.4	148.2	140.3	140.5	140.2	140.1	140.1
C(1′)	95.5	89.2	87.0	87.0	88.3	85.2	86.8	84.3/84.6
C(2′)	84.7	75.1	73.7	73.9	75.3	82.9	73.8	77.8
C(3′)	81.8	70.7	71.3	72.4	70.9	70.4	77.2	71.7/70.4
C(4′)	87.9	84.6	82.5	85.6	86.1	84.9	84.0	85.6/86.2
C(5′)	66.4	65.4	65.2	63.1	64.7	64.6	64.8	65.1/64.8
C(6′)	99.1	99.4	98.9	103.8	105.2	106.3	105.4	105.5/105.0
C(7′)	92.6	92.6	93.8	90.4	90.4	91.2	90.5	90.4/90.9

[a]) 3:2 Mixture of diastereoisomers.

1-[6,7-Dideoxy-5-O-(*2-nitrobenzoyl*)-*7*-C-*(triethylsilyl)-α*-L-talo-*hept-6-ynofuranosyl]uracil* (**3T**). A suspension of **2T** (7.29 g, 12.75 mmol) in H_2O (100 ml) was treated with CF_3COOH (100 ml), stirred at 80° for 12 h, diluted with H_2O (200 ml), and extracted with CH_2Cl_2. The combined org. layers were washed with brine, dried (Na_2SO_4), and evaporated to give crude **3T** (7.0 g, quant.). Red foam. R_f (AcOEt/MeOH 20:1) 0.55. $[\alpha]_D^{25} = -43.9$ ($c = 1.2$, $CHCl_3$). UV ($CHCl_3$): 259 (11500). IR ($CHCl_3$): 3391*w*, 2958*w*, 2876*w*, 2160*w*, 1746*m*, 1694*s*, 1537*m*, 1458*w*, 1395*w*, 1351*m*, 1280*m*, 1113*w*, 1069*m*, 1005*w*, 963*w*, 867*w*, 840*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 6*; additionally, 9.73 (br. *s*, NH); 7.95–7.91 (*m*, 1 arom. H); 7.73–7.60 (*m*, 3 arom. H); 3.24 (br. *s*, 2 OH); 0.99 (*t*, $J = 8.0$, $(MeCH_2)_3Si$); 0.64 (*q*, $J = 8.0$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 7*; additionally, 164.2 (*s*, OC=O); 148.2 (*s*, C(2) of 2-NO_2Bz); 133.6 (*d*, C(5) of 2-NO_2Bz); 132.7 (*d*, C(4) of 2-NO_2Bz); 130.3 (*d*, C(6) of 2-NO_2Bz); 126.9 (*s*, C(1) of 2-NO_2Bz); 124.5 (*d*, C(3) of 2-NO_2Bz); 7.5 (*q*, $(MeCH_2)_3Si$); 4.1 (*t*, $(MeCH_2)_3Si$). HR-MALDI-MS (DHB): 554.156 ($[M+Na]^+$, $C_{24}H_{29}N_3NaO_9Si^+$; calc. 554.157). Anal. calc. for $C_{24}H_{29}N_3O_9Si$ (531.59): C 54.23, H 5.50, N 7.90; found: C 54.22, H 5.50, N 7.70.

Benzoylation of **3T**. A soln. of pyridine (75 ml), 4-pyrrolidinopyridine (1.90 g, 12.8 mmol), and **3T** (6.8 g, 12.8 mmol) in CH_2Cl_2 (75 ml) was treated dropwise with BzCl (7.4 ml, 64.0 mmol), stirred at 23° for 36 h, diluted with CH_2Cl_2, washed with brine, dried (Na_2SO_4), and evaporated. FC (AcOEt/hexane 1:4 → 1:2) gave **4T** (8.2 g, 78%) and **5T** (1.28 g, 13%).

3-Benzoyl-1-[2,3-di-O-*benzoyl-6,7-dideoxy-5*-O-(*2-nitrobenzoyl*)-*7*-C-*(triethylsilyl)-α*-L-talo-*hept-6-ynofuranosyl]uracil* (**4T**). Colourless foam. R_f (AcOEt/hexane 1:1) 0.66. $[\alpha]_D^{25} = -64.2$ ($c = 1.1$, $CHCl_3$). UV (MeOH): 254 (19200). IR ($CHCl_3$): 3034*w*, 2957*w*, 2876*w*, 2150*w*, 1752*s*, 1712*m*, 1678*s*, 1601*w*, 1584*w*, 1538*m*, 1451*m*, 1376*m*, 1351*m*, 1316*m*, 1268*s*, 1121*m*, 1091*m*, 1070*m*, 1024*w*, 973*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 6*; additionally, 8.13–7.23 (*m*, 19 arom. H, H–C(6)); 1.02 (*t*, $J = 7.8$, $(MeCH_2)_3Si$); 0.68 (*q*, $J = 7.8$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 7*; additionally, 168.6 (*s*, NC=O); 165.7, 165.3 (2*s*, 2 OC=O); 164.2 (*s*, OC=O of 2-NO_2BzO); 148.2 (*s*, C(2) of 2-NO_2Bz); 139.6, 137.1 (2*d*, C(4) of 2 BzO); 135.4 (*d*, C(4) of BzN); 133.7 (*d*, C(4) of 2-NO_2BzO); 132.7 (*d*, C(5) of 2-NO_2BzO); 131.6 (*s*, C(1) of BzN); 130.9 (*d*, C(2) and C(6) of BzN); 130.3 (*d*, C(6) of 2-NO_2BzO); 130.2 (*d*, C(2) and C(6) of 2 BzO); 129.5 (*d*, C(3 and C(5) of BzN); 129.1 (*s*, C(1) of BzO); 128.9, 128.7 (2*d*, C(3) and C(5) of 2 BzO); 128.5 (*s*, C(1) of BzO); 126.8 (*s*, C(1) of 2-NO_2BzO); 124.5 (*d*, C(3) of 2-NO_2BzO); 7.3 (*q*, $(MeCH_2)_3Si$); 4.1 (*t*, $(MeCH_2)_3Si$). HR-MALDI-MS (DHB): 866.235 ($[M+Na]^+$, $C_{45}H_{41}N_3NaO_{12}Si^+$; calc. 866.236). Anal. calc. for $C_{45}H_{41}N_3O_{12}Si$ (843.92): C 64.05, H 4.90, N 4.98; found: C 64.10, H 5.04, N 4.92.

1-[2,3-Di-O-*benzoyl-6,7-dideoxy-5*-O-(*2-nitrobenzoyl*)-*7*-C-*(triethylsilyl)-α*-L-talo-*hept-6-ynofuranosyl]uracil* (**5T**). R_f (AcOEt/hexane 1:1) 0.36.

3-Benzoyl-1-[2,3-O-*dibenzoyl-6,7-dideoxy-7*-C-*(triethylsilyl)-α*-L-talo-*hept-6-ynofuranosyl]uracil* (**7T**). A soln. of **4T** (4.1 g, 4.86 mmol) in H_2O (150 ml) and MeCN (150 ml) was treated at 23° with three batches of NH_4Cl (0.78 g, 14.6 mmol) and Zn powder (0.48 g, 7.3 mmol) in intervals of 90 min, stirred for 6 h, and filtered through silica gel (elution with AcOEt). The filtrate was washed with sat. aq. $NaHCO_3$ soln. and brine, dried (Na_2SO_4), and evaporated. FC (toluene/MeOH 100:1 → 10:1) gave **7T** (3.3 g, quant.). Colourless foam. R_f (AcOEt/hexane 1:2) 0.38. $[\alpha]_D^{25} = -45.8$ ($c = 0.78$, $CHCl_3$). UV (MeOH): 254 (13400). IR ($CHCl_3$): 3035*w*, 2958*w*, 2876*w*, 2150*w*, 1752*m*, 1730*s*, 1712*m*, 1676*s*, 1601*w*, 1452*m*, 1378*m*, 1316*m*, 1266*s*, 1126*m*, 1094*m*, 1071*m*, 1024*w*, 974*w*, 921*w*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 6*; additionally, 8.64 (br. *s*, NH); 8.03–7.12 (*m*, 15 arom. H, H–C(6)); 1.01 (*t*, $J = 7.9$, $(MeCH_2)_3Si$); 0.65 (*q*, $J = 7.9$, $(MeCH_2)_3Si$). ^{13}C-NMR (75 MHz, $CDCl_3$): see *Table 7;* additionally, 168.7 (*s*, NC=O); 166.1, 165.8 (2*s*, 2 OC=O); 135.4 (*d*, C(4) of BzN); 134.3, 134.1 (2*d*, C(4) of 2 BzO); 131.5 (*s*, C(1) of BzN); 130.8 (*d*, C(2) and C(6) of BzN); 130.20, 130.15 (2*d*, C(2) and C(6) of 2 BzO); 129.4 (*d*, C(3) and C(5) of BzN); 129.0 (*s*, C(1) of BzO); 128.9, 128.7 (2*d*, C(3) and C(5) of 2 BzO); 128.6 (*s*, C(1) of BzO); 7.5 (*q*, $(MeCH_2)_3Si$); 4.1 (*t*, $(MeCH_2)_3Si$). HR-MALDI-MS (DHB): 717.223 ($[M+Na]^+$, $C_{38}H_{38}N_2NaO_9Si^+$; calc. 717.224).

1-[6,7-Dideoxy-5-O-*(4,4′-dimethoxytrityl)-7*-C-*(triethylsilyl)-α*-L-talo-*hept-6-ynofuranosyl]uracil* (**8T**). A soln. of DMTCl (6.0 g, 17.7 mmol) in CH_2Cl_2 (70 ml) was treated with *sym*-collidine (14.2 ml), stirred at 23° for 15 min, treated with $AgNO_3$ (3.0 g, 17.7 mmol), and stirred for 1 h. An aliquot of this suspension (69.1 ml, 14.4 mmol of the reagent) was filtered into a soln. of **7T** (4.0 g, 5.8 mmol) in CH_2Cl_2 (40 ml). The mixture was stirred at 23° for 75 min and filtered through silica gel (elution with AcOEt/Et_3N 99:1). After evaporation, a soln. of the residue in THF/MeOH/H_2O 5:4:1 (400 ml) was cooled to 0°, treated dropwise with 10M aq. NaOH soln. (5.8 ml, 58.0 mmol), stirred at 0° for 2 h, treated with sat. aq. NH_4Cl soln., and extracted with AcOEt/Et_3N 99:1. The org. phase was dried (Na_2SO_4), filtered, and evaporated. FC (hexane/AcOEt/Et_3N 1:1:0.01 → 1:2:0.01) gave **8T** (2.3 g, 57%). Colourless foam. R_f (AcOEt/hexane 1:1) 0.12. $[\alpha]_D^{25} = +26.0$ ($c = 0.8$, $CHCl_3$). UV (MeOH): 260 (7380). IR ($CHCl_3$): 3390*w*, 3006*w*, 2958*m*, 2875*w*, 2838*w*, 2125*w*, 1693*s*, 1608*m*, 1510*m*, 1463*m*, 1395*w*, 1280*m*, 1253*s*, 1116*w*, 1055*w*, 1037*m*, 965*w*, 890*w*, 829*m*. ^{1}H-NMR (300 MHz, $CDCl_3$): see *Table 6*;

additionally, 7.51–7.23 (*m*, 9 arom. H); 6.84–6.80 (*m*, 4 arom. H); 3.80 (*s*, 2 MeO); 2.60 (*s*, 2 OH); 0.93 (*t*, J = 7.8, (MeCH$_2$)$_3$Si); 0.51 (*q*, J = 7.8, (MeCH_2)$_3$Si). ^{13}C-NMR (75 MHz, CDCl$_3$): see *Table 7*; additionally, 159.1 (*s*, C(4) of 2 MeOC$_6$H$_4$); 145.1 (*s*, C(1) of Ph); 136.0, 135.7 (2*s*, C(1) of 2 MeOC$_6$H$_4$); 130.9, 130.7 (2*d*, C(2) and C(6) of 2 MeOC$_6$H$_4$); 128.6 (*d*, C(2) and C(6) of Ph); 128.1 (*d*, C(3) and C(5) of Ph); 127.4 (*d*, C(4) of Ph); 113.40, 113.35 (2*d*, C(3) and C(5) of 2 MeOC$_6$H$_4$); 88.6 (*s*, Ar$_3$$C$); 55.3 (*q*, 2 MeO); 7.4 (*q*, (MeCH$_2$)$_3$Si); 4.1 (*t*, (MeCH$_2$)$_3$Si). Anal. calc. for C$_{38}$H$_{44}$N$_2$O$_8$Si (684.86): C 66.64, H 6.48, N 4.09; found: C 66.73, H 6.41, N 3.87.

(Silyloxy)methylation of **8T**. A soln. of **7T** (1.78 g, 2.6 mmol) and (i-Pr)$_2$NEt (2.2 ml, 13.0 mmol) in (CH$_2$Cl)$_2$ (12 ml) was treated with Bu$_2$SnCl$_2$ (0.95 g, 3.12 mmol), stirred for 90 min at 23°, treated with TOMCl (0.81 g, 3.64 mmol), heated to 80°, stirred for 45 min, treated with sat. aq. NaHCO$_3$ soln., and extracted with CH$_2$Cl$_2$. The org. phase was washed with brine, dried (Na$_2$SO$_4$), and evaporated. FC (hexane/AcOEt/Et$_3$N 4:1:0.01 → 2:1:0.01) gave **9T** (716 mg, 32%) and **10T** (235 mg, 10%).

*1-(6,7-Dideoxy-5-*O*-(4,4′-dimethoxytrityl)-7-*C*-(triethylsilyl)-2-*O*-{[(triisopropylsilyl)oxy]methyl}-α-*L*-talo-hept-6-ynofuranosyl)uracil* (**9T**). Colourless foam. R_f (AcOEt/hexane 1:2) 0.29. $[\alpha]_D^{25}$ = 58.8 (c = 0.76, CHCl$_3$). UV (MeOH): 260 (18800). IR (CHCl$_3$): 3392*w*, 3007*w*, 2957*m*, 2871*m*, 2140*w*, 1695*s*, 1608*w*, 1510*m*, 1463*m*, 1385*w*, 1295*w*, 1255*m*, 1111*w*, 1075*m*, 1046*m*, 1011*m*, 883*w*, 829*w*. ^{1}H-NMR (300 MHz, CDCl$_3$): see *Table 6*; additionally, 8.02 (br. *s*, NH); 7.51–7.21 (*m*, 9 arom. H); 6.85–6.79 (*m*, 4 arom. H); 5.18, 4.86 (2*d*, J = 4.9, OCH$_2$O); 3.799, 3.796 (2*s*, 2 MeO); 1.11–1.07 (*m*, (Me$_2$CH)$_3$Si); 0.94 (*t*, J = 7.8, (MeCH$_2$)$_3$Si); 0.54 (*q*, J = 7.8, (MeCH_2)$_3$Si). ^{13}C-NMR (75 MHz, CDCl$_3$): see *Table 7*; additionally, 159.0 (*s*, C(4) of 2 MeOC$_6$H$_4$); 145.1 (*s*, C(1) of Ph); 136.0, 135.7 (2*s*, C(1) of 2 MeOC$_6$H$_4$); 130.8, 130.7 (2*d*, C(2) and C(6) of 2 MeOC$_6$H$_4$); 128.6 (*d*, C(2) and C(6) of Ph); 128.0 (*d*, C(3) and C(5) of Ph); 127.3 (*d*, C(4) of Ph); 113.4, 113.3 (2*d*, C(3) and C(5) of 2 MeOC$_6$H$_4$); 90.0 (*t*, OCH$_2$O); 88.4 (*s*, Ar$_3$$C$); 55.4 (*q*, 2 MeO); 18.0 (*q*, (Me_2CH)$_3$Si); 12.1 (*d*, (Me$_2$$C$H)$_3$Si); 7.7 (*q*, ($MeCH_2$)$_3$Si); 4.3 (*t*, (Me$CH_2$)$_3$Si). FAB-MS (NOBA): 870 (1, M^+), 303 (100, DMTr$^+$). Anal. calc. for C$_{48}$H$_{66}$N$_2$O$_9$Si$_2$ (871.22): C 66.17, H 7.64, N 3.22; found: C 65.93, H 7.60, N 3.23.

*1-(6,7-Dideoxy-5-*O*-(4,4′-dimethoxytrityl)-7-*C*-(triethylsilyl)-3-*O*-{[(triisopropylsilyl)oxy]methyl}-α-*L*-talo-hept-6-ynofuranosyl)uracil* (**10T**). Colourless foam. R_f (AcOEt/hexane 1:2) 0.14. ^{1}H-NMR (300 MHz, CDCl$_3$): see *Table 6*; additionally, 7.51–7.23 (*m*, 9 arom. H); 6.86–6.78 (*m*, 4 arom. H); 5.09, 4.95 (2*d*, J = 4.7, OCH$_2$O); 3.798, 3.796 (2*s*, 2 MeO); 1.14–1.06 (*m*, (Me$_2$CH)$_3$Si); 0.97 (*t*, J = 7.8, (MeCH$_2$)$_3$Si); 0.57 (*q*, J = 7.8, (MeCH_2)$_3$Si). ^{13}C-NMR (75 MHz, CDCl$_3$): see *Table 7*; additionally, 159.3 (*s*, C(4) of 2 MeOC$_6$H$_4$); 145.1 (*s*, C(1) of Ph); 136.0, 135.7 (2*s*, C(1) von 2 MeOC$_6$H$_4$); 130.8, 130.6 (2*d*, C(2) and C(6) of 2 MeOC$_6$H$_4$); 128.5 (*d*, C(2) and C(6) of Ph); 128.2 (*d*, C(3) and C(5) of Ph); 127.5 (*d*, C(4) of Ph); 113.5 (*d*, C(3) and C(5) of 2 MeOC$_6$H$_4$); 89.7 (*t*, OCH$_2$O); 88.8 (*s*, Ar$_3$$C$); 55.4 (*q*, 2 MeO); 17.9 (*q*, (Me_2CH)$_3$Si); 12.0 (*d*, (Me$_2$$C$H)$_3$Si); 7.5 (*q*, ($MeCH_2$)$_3$Si); 4.2 (*t*, (Me$CH_2$)$_3$Si).

*1-(6,7-Dideoxy-5-*O*-(4,4′-dimethoxytrityl)-7-*C*-(triethylsilyl)-2-*O*-{[(triisopropylsilyl)oxy]methyl}-α-*L*-talo-hept-6-ynofuranosyl)uracil 3-[(2-Cyanoethyl) (Diisopropyl)phosphoramidite]* (**11T**). A soln. of **9T** (400 mg, 0.46 mmol) and (i-Pr)$_2$NEt (0.24 ml, 1.38 mmol) in CH$_2$Cl$_2$ (2.5 ml) was treated dropwise with 2-cyanoethyl (diisopropyl)phosphoramidochloridite (0.14 ml, 0.64 mmol) and stirred at 23° for 3 h. Evaporation and FC (hexane/AcOEt/Et$_3$N 4:1:0.01 → 1:1:0.01) gave **11T** (374 mg, 76%). R_f (AcOEt/hexane 1:2) 0.48, 0.44 (2 diastereoisomers). $[\alpha]_D^{25}$ = 5.9 (c = 0.86, CHCl$_3$). UV (MeOH): 259 (12300). IR (CHCl$_3$): 3400*w*, 2960*m*, 2870*m*, 2250*w*, 1724*m*, 1695*s*, 1608*w*, 1510*m*, 1463*m*, 1382*w*, 1365*w*, 1301*w*, 1120*w*, 1041*m*, 981*w*, 829*w*. ^{1}H-NMR (500 MHz, CDCl$_3$; 3:2 mixture of diastereoisomers): see *Table 6*; additionally, 7.50–7.20 (*m*, 9 arom. H); 6.84–6.79 (*m*, 4 arom. H); 5.00 (*d*, J = 5.4, 0.4 H), 4.97 (*d*, J = 5.5, 0.6 H), 4.85 (*d*, J = 5.4, 0.4 H), 4.82 (*d*, J = 5.4, 0.6 H) (OCH$_2$O); 3.792, 3.790, 3.785, 3.782 (4*s*, 2 MeO); 3.67–3.55 (*m*, NCCH$_2$CH_2O, (Me$_2$CH)$_2$N); 2.65–2.52 (*m*, NCCH); 2.35 (*m*, NCCH); 1.21–1.17 (*m*, (Me_2CH)$_2$N, (Me$_2$CH)$_3$Si); 1.04–0.91 (*m*, (Me_2CH)$_3$Si, (MeCH$_2$)$_3$Si); 0.58–0.53 (*m*, (MeCH_2)$_3$Si). ^{13}C-NMR (75 MHz, CDCl$_3$; 3:2 mixture of diastereoisomers): see *Table 7*; additionally, 158.95 (2 C), 158.92, 158.86 (3*s*, C(4) of 2 MeOC$_6$H$_4$); 144.96, 144.82 (2*s*, C(1) of Ph); 135.66, 135.56, 135.39, 135.35 (4*s*, C(1) of 2 MeOC$_6$H$_4$); 130.85, 130.75 (2*d*, C(2) and C(6) of 2 MeOC$_6$H$_4$); 128.61–127.16 (several *d*); 117.63, 117.47 (2*s*, CN); 113.15, 113.11, 113.05, 113.00 (4*d*, C(3) and C(5) of 2 MeOC$_6$H$_4$); 89.35, 89.30 (2*t*, OCH$_2$O); 88.67 (*s*, Ar$_3$$C$); 58.75, 57.90 (2*dt*, ^2J(C,P) = 17, NCCH$_2$$CH_2$O); 55.22, 55.20 (2*q*, 2 MeO); 43.52, 43.41 (2 C), 43.30 (3*d*, 2 (Me$_2$$C$H)$_2$N); 24.69–24.50 (several *q*, (Me_2CH)$_2$N); 20.40 (*dt*, ^3J(C,P) = 6.3), 19.73 (*dt*, ^3J(C,P) = 7.5) (NCCH$_2$CH$_2$O); 17.71, 17.68 (4 C), 17.67 (3*q*, (Me_2CH)$_3$Si); 11.90 (*d*, (Me$_2$$C$H)$_3$Si); 7.40, 7.39 (2*q*, ($MeCH_2$)$_3$Si); 4.03, 3.97 (2*t*, (MeCH$_2$)$_3$Si). ^{31}P-NMR (121 MHz, CDCl$_3$; 3:2 mixture of diastereoisomers): 151.3 (0.6 P), 150.0 (0.4 P). FAB-MS (NOBA): 1071 (10, $[M+H]^+$), 303 (100, DMTr$^+$).

Oligonucleotide Synthesis. Oligonucleotides were synthesised on a *Pharmacia Gene Assembler* on a 1.3-μmol scale. The commercial phoshophoramidites and the CPG solid supports were obtained from *Xeragon*. Solvents and reagents were prepared essentially according to the protocol for the synthesis of p-RNA [59].

Detritylation was accomplished within 2.5 min by 6% $Cl_2CHCOOH$ in $(CH_2Cl)_2$. Couplings (0.12 ml of 0.1M phosphoramidite soln. in MeCN or in MeCN/THF 7:3 for phosphoramidite **11A** and **11T**, resp., and 0.36 ml of 0.3M 1-(benzylsulfanyl)-1*H*-tetrazole in MeCN) were performed within 5 min (RNA-phosphoramidites coupled on unmodified nucleosides), or within 10 min (modified phosphoramidites **11A** and **11T**, and RNA-phosphoramidites coupled with modified nucleotides). Capping and oxidation was accomplished under standard conditions (see user manual for *Gene Assembler Plus*).

Cleavage of Cyanoethyl and Nucleobase Protecting Groups with Concomitant Detachment from the CPG Solid Support and Cleavage of the TOM and Et_3Si Protecting Groups. After completion of the automated synthesis, the CPG support carrying the oligonucleotide was dried *in vacuo* for 30 min, placed in *Eppendorf* tubes (1.5 ml), and treated with 10M $MeNH_2$ in H_2O/EtOH 1:1 (1 ml) for 2 h at 23°, followed by centrifugation. The supernatant was removed, and the CPG support was washed with H_2O (3×0.5 ml). The combined washings and supernatant were evaporated with a *SpeedVac* evaporator. The residue was treated with 1M TBAF in THF (1 ml; for **II**–**VI**) or with 0.5M TBAF in THF/*N*-methylpyrrolidinone 1:1 (1 ml; for **I**) at 23° for 12 h. The mixture was treated with 1M Tris · HCl (pH 7.4, 1 ml) and concentrated to 1 ml (*SpeedVac* evaporator). Salts were removed by chromatography (*Sephadex G-10* column, elution with sterile H_2O, flow rate: 1 ml/min). The UV absorbing fractions eluting between 12–18 min were collected and evaporated.

HPLC Purification of the Oligonucleotides. Oligonucleotides were purified by ion-exchange HPLC. Column: *SAX 1000-8* (*Macherey & Nagel*); buffer *A:* 10 mM Na_2HPO_4 in H_2O, pH 10.5; buffer *B:* 10 mM Na_2HPO_4 in H_2O, 1M NaCl, pH 10.5. Product fractions were collected in 0.1M aq. $Et_3NH \cdot HCO_3$ buffer soln.

Desalting of the Oligonucleotides. The product fractions obtained by HPLC were combined, diluted with 0.1M aq. $Et_3NH \cdot HCO_3$ buffer to twice the volume, and applied to a previously conditioned reverse-phase *SepPak* cartridge (*C18 Waters*). Successive elution with 0.1M aq. $Et_3NH \cdot HCO_3$ buffer soln., H_2O, and MeCN/H_2O 1:1, followed by lyophillization of the MeCN/H_2O fractions containing the products afforded the salt-free oligonucleotides.

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