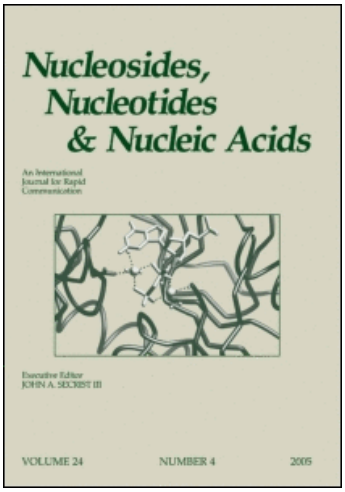

Nucleosides, Nucleotides and Nucleic Acids
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597286

SYNTHESIS OF 5-(2,2'-BIPYRIDINYL AND 2,2'-BIPYRIDINEDIIUMYL)-2'-DEOXYURIDINE NUCLEOSIDES: PRECURSORS TO METALLO-DNA CONJUGATES

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Online publication date: 28 October 2002

To cite this Article Gaballah, Samir T. , Kerr, Charles E. , Eaton, Bruce E. and Netzel, Thomas L.(2002) 'SYNTHESIS OF 5-(2,2'-BIPYRIDINYL AND 2,2'-BIPYRIDINEDIIUMYL)-2'-DEOXYURIDINE NUCLEOSIDES: PRECURSORS TO METALLO-DNA CONJUGATES', Nucleosides, Nucleotides and Nucleic Acids, 21: 8, 547 — 560
To link to this Article: DOI: 10.1081/NCN-120015068
URL: http://dx.doi.org/10.1081/NCN-120015068

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NUCLEOSIDES, NUCLEOTIDES & NUCLEIC ACIDS
Vol. 21, Nos. 8 & 9, pp. 547–560, 2002

SYNTHESIS OF 5-(2,2′-BIPYRIDINYL AND 2,2′-BIPYRIDINEDIIUMYL)-2′-DEOXYURIDINE NUCLEOSIDES: PRECURSORS TO METALLO-DNA CONJUGATES

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ABSTRACT

The synthesis of 2,2′-bipyridinyl-2′-deoxyuridine metal-chelator nucleosides (Bipy-dU) with either ethynyl or ethylenyl linkers was now been accomplished. These new nucleosides will permit the construction of a number of corresponding metallo-DNA conjugates where many types of metals can be complexed to the 2,2′-bipyridinyl chelator group and the resulting metallo-dU conjugates incorporated into DNA oligonucleotides. Additionally this paper also reports the synthesis of a di-*N*-alkylated bipyridinediiumyl-2′-deoxyuridine nucleoside ($Bipy^{2+}$-dU) with an ethylenyl linker. The $Bipy^{2+}$-dU nucleoside was found to decompose under basic conditions precluding its use in standard automated DNA-synthesis by the phosphoramidite method. No such restrictions apply to the two Bipy-dU nucleosides reported here for use as metal chelators.

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DOI: 10.1081/NCN-120015068 1525-7770 (Print); 1532-2335 (Online)

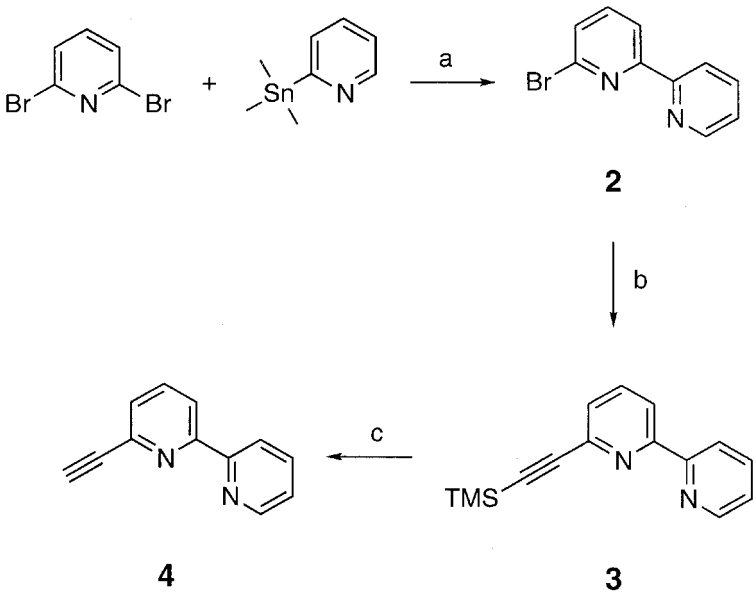
INTRODUCTION

Bipyridines have been widely used as ligands in coordination chemistry. Because of its stability, ease of functionalization, and chelating metal-coordination, 2,2′-bipyridine, in particular, has been heavily used to link metals to other molecular subunits.[1] Additionally, bipyridines have played important roles in constructing supramolecular assemblies,[1–3] developing pharmaceuticals[4–10] and sensors,[11,12] and mimicking photosynthetic light conversion.[13] Recently, bipyridines have also been used to make functionalized polymers[14–17] and photocatalysts.[18]

Two desires motivated our efforts to link 2,2′-bipyridine to 2′-deoxyuridine. First we were interested in synthesizing new bipyridine-deoxyuridine conjugates where the linker between the metal chelator and uracil was short, two carbons, and could be either flexible or rigid. It was also desired to prepare these new bipyridine-deoxyuridine conjugates so that they could be incorporated into DNA as metal-chelators. A number of research groups have constructed DNA duplexes with one or more covalently attached metal complexes.[19–25] These groups were interested in the physical properties of novel metallo-DNA constructs, in questions of the distance dependence of the rates of energy and electron transfer between the attached metallosubunits themselves and between the metallosubunits and particular DNA bases, and in the development of DNA hybridization probes and sensors. Thus routes to constructing new metallo-DNA conjugates appear to be of broad interest. This work reports the synthesis of 2,2′-bipyridinyl-dU conjugates with either ethynyl (**6**, Sch. 2) or ethylenyl (**1**, Sch. 3) linkers that will permit the construction of a number of corresponding metallo-DNA conjugates, since many types of metals can be complexed to the 2,2′-bipyridinyl chelator group and the resulting metallo-dU conjugates can then be incorporated into DNA oligonucleotides in a variety of ways.[26–30]

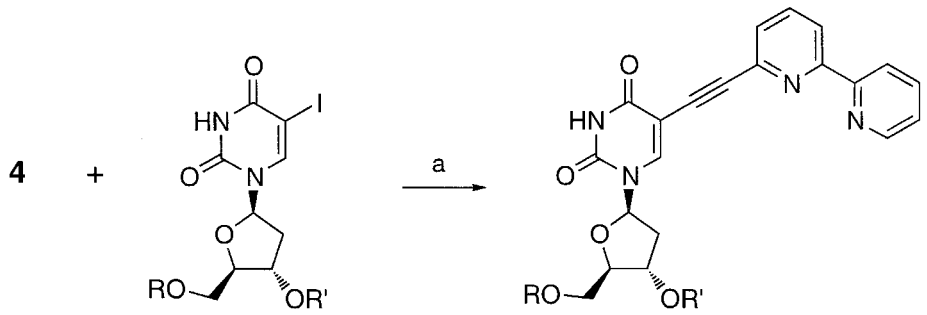
Secondarily, it was also of interest to convert these new conjugates into methyl viologen analogs, 5-(2,2′-bipyridinediiumyl)-2′-deoxyuridine nucleosides ($Bipy^{2+}$-dU). By analogy to 4,4′-bipyridinediium (methyl viologen),[31,32] we reasoned that $Bipy^{2+}$-dU nucleoside conjugates could function as reversible electron acceptors when incorporated into DNA duplexes. Importantly, singly reduced $Bipy^{2+}$ radicals are known to have strong absorbances in the UV-vis region and therefore should serve as good indicators of excess electron trapping in kinetics studies of electron transport in DNA.[33]

To date our efforts in the area of nucleoside conjugate chemistry have produced a number of pyrenyl-dU (Py-dU) conjugates that are capable of photoinjecting an excess electron into a DNA π-stack.[34–38] In these conjugates the lowest electronic, excited singlet-state of pyrene reduces the attached uracil in less than 30 ps, and in methanol the resulting primary electron transfer (ET) product, $Py^{\bullet+}/dU^{\bullet-}$, lives from 6 ps to 2.1 ns, depending upon the type of linker that joins the pyrenyl and uracil subunits.



Scheme 1. (a) PdL_4, toluene, reflux, overnight, (b) PdL_2Cl_2, CuI, Et_3N, THF, TMSA, (c) KF, THF, MeOH.

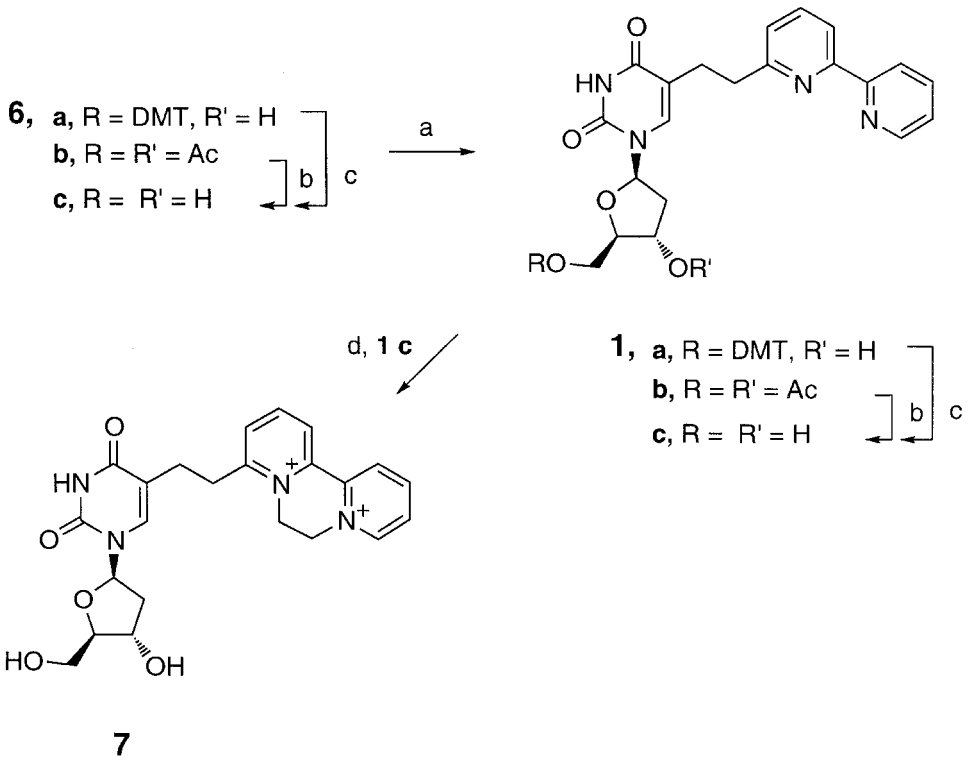
To further increase the lifetime of a photogenerated uracil anion in a DNA duplex, we have also synthesized several dimethylanilino-dU conjugates (DMA-dU) to serve as secondary electron donors.[39] In duplexes labeled with both Py-dU and DMA-dU, secondary ET from DMA to $Py^{\bullet+}/dU^{\bullet-}$ should produce the $DMA^{\bullet+}/Py/dU^{\bullet-}$ ET product that is expected to be



5, a, R = DMT, R' = H
b, R = R' = Ac
c, R = R' = H

6, a, R = DMT, R' = H
b, R = R' = Ac
c, R = R' = H

Scheme 2. (a) PdL_4, CuI, Et_3N, THF.



Scheme 3. (a) H_2 (50 psi), Pd/C, MeOH, (b) NH_4OH, MeOH, overnight, (c) CF_3COOH, CH_2Cl_2, (d) $BrCH_2CH_2Br$, 100°C, 24 h.

significantly longer lived than $Py^{\bullet+}/dU^{\bullet-}$. Thus routes to producing electron-trap nucleosides are of current interest. In addition to the syntheses of two kinds of Bipy-dU nucleosides (**1** and **6**), this paper also reports the synthesis of a $Bipy^{2+}$-dU nucleoside with an ethylenyl linker (**7**, Sch. 3). However, the nucleoside **7** was found to react rapidly with pyridine, and thus will not survive the base deprotection conditions required after standard automated DNA-strand synthesis by the phosphoramidite method. In contrast no such restrictions apply to the Bipy-dU nucleosides reported here for use as metal chelators.

To make both Bipy-dU and $Bipy^{2+}$-dU conjugates, we focused on connecting bipyridine via short alkynyl or alkyl linkers to the 5-position of deoxyuridine as a direct way of positioning the chelating and electron accepting subunits in the DNA major groove. These linkers were selected both to provide strong electronic coupling between uracil and the polypyridinyl subunits and to accurately locate the polypyridinyl subunits along the surface of a DNA duplex. Additionally, locating the polypyridinyl subunits in the major groove was less likely to interfere with normal DNA base pairing than would locating them in the minor groove. These considerations

in turn made both ethynylbipyridine and ethynyl-dU attractive precursors to final ethynyl- and ethylenyl-linked nucleoside conjugates.

Our synthetic scheme used a common bromobipyridine precursor (**2**, Sch. 1), two Sonogashira couplings to effect conjugation of the bipyridine to the deoxyuridine (Schs. 1 and 2), and a final alkyne reduction to convert **6** to **1** (Sch. 3). Several different methods of preparing bromobipyridines have been reported.[40–45] After investigating several of these, we found that adaptation of a pyridylstannane-based synthesis of 2,2′:6′,2″-terpyridine[42] produced **2** in good overall yield. Palladium catalyzed coupling of **2** to trimethylsilylacetylene (TMSA) followed by desilylation with KF was performed successfully on a multi-gram scale (see Sch. 1).

With 6-ethynyl-2,2′-bipyridine (**4**) in hand, the Sonogashira coupling was performed, giving the ethynyl-linked Bipy-dU conjugate (**6**, Sch. 2). To eliminate the π-conjugation between the deoxyuridine and the bipyridine subunits, the alkyne linker was selectively reduced via catalytic hydrogenation giving the ethylenyl-linked Bipy-dU conjugate (**1**, Sch. 3). Subsequent quaternization of (**1a**) with 1,2-dibromoethane produced the ethylenyl-linked $Bipy^{2+}$-dU conjugate (**7**, Sch. 3).

RESULTS AND DISCUSSION

A great variety of methods to prepare functionalized 2,2′-bipyridines have been described in the literature,[40,41,43,44,46–48] and forming 6-bromo-2,2′-bipyridine (**2**, Sch. 1) was key to synthesizing 6-ethynylbipyridine (**4**). The key compound **2** had been synthesized previously using two different approaches. One approach synthesized **2** from 6,6′-dibromo-2,2′-bipyridine and phenyllithium.[40] A second approach used Stille-type cross-coupling.[47,49,50] This latter method had been used to make terpyridines and bipyridines using pyridyl stannane in conjunction with, respectively, dibromopyridine and bromopyridine. It was found that reproducible yields of **2** (67% isolated) could be obtained following the second approach starting with 2-(trimethylstannanyl)pyridine and 2,6-dibromopyridine in refluxing toluene in the presence of PdL_4. Compound **3** was synthesized from **2** and TMSA through a Pd(0)-cross coupling reaction.[51] The Pd-catalysed reaction could be carried out using PdL_4 (ca. 5 mol%) in dry tetrahydrofuran (THF) or dimethyformamide (DMF) in the presence of CuI and triethylamine (Et_3N) at 60°C. Under these conditions using PdL_4 catalyst, the observed yield was 68%. Also, the Pd-catalyst could be created in situ from PdL_2Cl_2.[42] These latter conditions using PdL_2Cl_2 catalyst gave the best yield of the desired coupling product (89%, see Sch. 1). Removing the trimethylsilyl group of **3** with KF in THF/methanol (MeOH) (1:1 by vol.) solution at room temperature produced compound **4** in 98% yield.

The nucleoside **6c** could be prepared directly by performing the Pd-coupling with **4** and **5c** (see Sch. 2). However, purification of the crude reaction product was problematic. It was more convenient and reproducible to perform the palladium coupling reaction and subsequent purifications on either the 5′-*O*-DMT protected **6a** or the 3′-*O*, 5′-*O*- diacetate protected **6b**. Three steps were employed to synthesize **1a** in Sch. 3. First, the 5′-OH of **5c** was protected with DMTCl in dry pyridine to afford 5′-*O*-DMT-IdU, **5a**.[52] Second, **5a** was coupled to **4** in dry THF or DMF at 60°C using Pd(0)-cross coupling to form **6a**. To produce **1a**, compound **6a** was hydrogenated on 10% Pd/C in dry MeOH by stirring at room temperature for 24–48 h in a sealed glass vessel pressurized with hydrogen (50 psi).[53] The glass vessel was sealed with either stainless steel or brass fittings and topped with a pressure gauge. Before adding **6a**, the Pd/C-catalyst in MeOH was activated by pressurizing the hydrogenation vessel with hydrogen (50 psi) and stirring the slurry for 15–20 m.

We were also interested in synthesizing the diquat nucleoside conjugate **7** in Sch. 3, ethylenyl-linked $Bipy^{2+}$-dU. (Note that attempts to quanternize **6c**, the alkynyl-linked Bipy-dU nucleoside, using similar reaction conditions were not successful. This reaction was carried out in dry freshly distilled 1,2-dibromoethane and a precipitate formed as in the quaternization of **1c**. However, NMR analysis showed that the precipitate was not the desired diquat. Also, TLC analysis of this product showed multiple spots, and no useful amount of the desired diquat was recovered.) The alkylation conditions required forming the diquat conjugate **7** from the free nucleoside **1c**. Alkylation of the two nitrogen atoms of the bipyridine moiety was carried out in dry freshly distilled 1,2-dibromoethane. Compound **7** was formed in 40% yield after refluxing **1c** in a solution of 1,2-dibromoethane for 24 h. While **7** was stable and readily isolated under neutral conditions, when it was added to neat pyridine the resulting solution turned dark green within ten minutes and eventually became black. TLC analysis of this black solution showed numerous, uncharged degradation products. Related studies on the stability of several model diquats, including methylviologen, to 10% diethyl amine in MeOH and 0.05 M K_2CO_3 in MeOH showed that all of them reacted rapidly to produce strongly colored solutions and uncharged degradation products (as determined by TLC analysis).[54]

SUMMARY

Reproducible synthetic methods are now available to prepare bipyridinyl-deoxyuridine conjugates with short, two-atom linkers that are either stiff or flexible. These conjugates will locate the bipyridinyl subunits in the major groove of DNA and will be valuable building blocks for creating DNA structures for studies of the properties of metallo-DNA where precise positioning of the bipyridinyl chelator is important. We have also shown

that **7**, a viologen analog, can be prepared via a straightforward dialkylation of the **6c** nucleoside with 1,2-dibromoethane, and is stable under neutral conditions. Conjugate **7**, however, is not stable under the standard base deprotection conditions that are encountered in automated DNA synthesis. Our current efforts continue to explore new methods for preparing viologen-related deoxyuridine nucleosides that will survive automated DNA synthesis and therefore will be able to serve as traps in studies of excess electron transport in DNA.

EXPERIMENTAL

General Synthetic Procedures

All manipulations of tetrakis(triphenylphosphine)palladium(0) (PdL_4) and 4,4′-dimethoxytrityl chloride (DMTCl) were performed in a Vacuum Atmospheres M040–2 glove box pressurized with dry nitrogen gas. All other reactions were carried out on the benchtop under a dry nitrogen atmosphere. Chromatography was carried out on a Biotage Flash-40^{TM} system using either prepackaged KP-Sil^{TM} cartridges, or Flash-40^{TM} cartridge housings repacked with Whatman^{TM} flash silica (80 Å pore, 230–400 mesh). Silica gel used for pad filtrations and dry powder chromatography sample loading was also Whatman^{TM} flash silica. PdL_4 was prepared according to the literature[55] and stored in a glove-box freezer (−33°C). The following solvents were dried and redistilled in continuous circulation distillation apparati: tetrahydrofuran (THF, dried with benzophenone /Na^0), triethylamine (Et_3N, dried with CaH_2), *N,N*-dimethylformamide (DMF, dried with CaH_2), and methanol (MeOH, dried with Mg turnings). Trimethylsilylacetylene (TMSA) was obtained from GFS Chemicals and used after vacuum transfer from CaH_2. 1,2-dibromoethane was purchased from Aldrich and was distilled with CaH_2 after treating with 5% $NaHCO_3$ and drying over anhydrous sodium sulfate. All copper salts used in preparing bipyridines or palladium couplings were obtained from Strem Chemicals. 5-Iodo-2′-deoxyuridine (IdU) obtained from Aldrich was used as received. Phenyllithium and butyllithium were obtained from common sources and titrated prior to use with 2-butanol and 9,10-phenanthroline in hydrocarbon solution. Other reagents and solvents were obtained from common suppliers and were usually used without further purification. Mass spectrometry was performed at the Georgia Institute of Technology. NMR Spectra were obtained from a Varian Unity+300 NMR spectrometer operating at either 75 or 300 MHz frequencies located at Georgia State University.

6-Bromo-2,2′-bipyridine, 2. 6-Bromo-2,2′-bipyridine was prepared from 2-trimethylstannylpyridine and 2,6-dibromopyridine by adaptation of a literature method,[47,48] or less conveniently from 6,6′-dibromo-2,2′

bipyridine also by adaptation of a literature method.[40] From the stannane, the procedure was as follows: 2-Trimethylstannylpyridine (44.35 g, 183 mmol), 2,6-dibromopyridine (45.60 g, 192 mmol), PdL_4 (10.60 g, 9.2 mmol), and 500 mL dry toluene were combined and refluxed for 18 h. Toluene was removed in vacuo. The resulting crude material was loaded onto a flash silica column and chromatographed. Elution with hexanes then benzene afforded both pure bromobipyridine (10.6 g as a white solid after recrystallization from hexanes) and impure bromobipyridine. The impure material was repurified by recrystallization from hexanes giving 18.4 g. The total yield of **2** was 67% based on 2,6-dibromopyridine and gave analytical data identical to those previously reported.

6-(Trimethylsilanylethynyl)-2,2′-bipyridine, 3. To a solution of **2** (1.82 g, 7.73 mmol) in THF (5 mL) were added, successively, PdL_2Cl_2 (0.27 g, 0.38 mmol), CuI (0.15 g, 0.77 mmol), Et_3N (3 mL), THF (10 mL), and, finally, TMSA (4.37 mL, 30.9 mmol) in a nitrogen atmosphere. The yellow solution was heated at 60°C until the starting material was completely consumed (2–3 h). The solvent was evaporated to give a crude product that was purified by pad filtration with ethyl acetate/CH_2Cl_2 (1:2 by vol.) then crystallized from hexanes to give 0.99 g of **3** (89% yield).^{1}H NMR ($CDCl_3$, 300 MHz) δ: 0.303 (9H, s, $(CH_3)_3$), 7.30 (1H, m, Ar), 7.49 (1H, dd, $J = 0.9$, 7.5 Hz, Ar), 7.74–7.84 (2H, m, Ar), 8.36 (1H, dd, $J = 0.9$, 8.1 Hz, Ar), 8.47 (1H, dt, $J = 0.9$, 8.1 Hz, Ar), 8.66 (1H, dq, $J = 0.9$, 4.8 Hz, Ar).^{13}C NMR ($CDCl_3$, 75 MHz) δ: −0.21, 94.48, 103.98, 120.54, 121.62, 123.96, 127.54, 136.88, 136.95, 142.44, 149.04, 155.41, 156.44. HRMS (EI) m/z for $C_{15}H_{16}N_2Si$ (M^+): calc'd. 252.1082, found 252.1075.

6-Ethynyl-2,2′-bipyridine, 4. A mixture of **3** (0.985 g, 3.90 mmol) and KF (0.27 g, 4.70 mmol) in MeOH/THF (50 mL) was stirred for 2 h at room temperature. The crude product was purified using silica gel pad filtration with ethyl acetate/CH_2Cl_2 (1:2 by vol.) to produce 0.68 g of **4** (98% yield).^{1}H NMR ($CDCl_3$, 300 MHz) δ: 3.22 (1H, s, ≡CH), 7.29 (1H, qd, $J = 0.9$, 7.5 Hz, Ar), 7.49 (1H, dd, $J = 0.9$, 7.5 Hz, Ar), 7.75–7.82 (2H, m, Ar), 8.40 (1H, dd, $J = 0.9$, 7.9 Hz, Ar), 8.46 (1H, dt, $J = 0.9$, 7.9 Hz, Ar), 8.65 (1H, dq, $J = 0.9$, 4.8 Hz, Ar).^{13}C NMR ($CDCl_3$, 75 MHz) δ: 77.44, 82.93, 121.00, 121.34, 123.94, 127.33, 136.80, 136.97, 141.48, 148.92, 155.04, 156.36. HRMS (EI) m/z for $C_{12}H_8N_2$ (M^+): calc'd. 180.0687, found 180.0674.

5′-*O*-(4,4′-Dimethoxytrityl)-5-((2,2′-bipyridin-6-yl)-ethynyl)-2′-deoxyuridine, 6a. To a solution of **5a** (1.97 g, 3.0 mmol), PdL_4 (0.173 g, 0.15 mmol), CuI (0.057 g, 0.3 mmol), and Et_3N (2.09 mL, 15 mmol) in THF (18 mL) was added **4** (0.702 g, 1.3 mmol) in a glove box. The reaction mixture was stirred at 50–60°C for 3 h. The reaction mixture was chromatographed

on a silica gel column pre-equilibrated with 1% Et_3N in CH_2Cl_2 (by vol.) and eluted with 10:0.0, 9.9:0.1, 9.8:0.2, 9.7:0.3, and 9.6:0.4 CH_2Cl_2/MeOH (by vol.). A yellow foam of **6a** was obtained (1.80 g, 84% yield).^{1}H NMR (DMSO-d_6, 300 MHz) δ: 2.29–2.43 (2H, m, $H_{2'\beta}/H_{2'\alpha}$), 3.22–3.33 (2H, m, $H_{5'\alpha}/H_{5'\beta}$), 3.67 (6H, s, DMT-$(CH_3O)_2$), 4.03 (1H, m, $H_{4'}$), 4.37 (1H, m, $H_{3'}$), 5.42 (1H, d, $J = 4.5$ Hz, $OH_{3'}$), 6.21 (1H, t, $J = 6.3$ Hz, $H_{1'}$), 6.89 (4H, dd, $J = 3.9$, 8.94 Hz, Ar), 7.09 (1H, dd, $J = 0.9$, 7.5 Hz, Ar), 7.19 (1H, t, $J = 7.2$ Hz, Ar), 7.31–7.38 (6H, m, Ar), 7.46–7.56 (3H, m, Ar), 7.89 (1H, t, $J = 7.8$ Hz, Ar), 7.97 (1H, dt, $J = 1.5$, 7.65 Hz, Ar), 8.27 (1H, s, H_6), 8.33–8.39 (2H, m, Ar), 8.75 (1H, m, Ar), 11.90 (1H, br, H_{N3}).^{13}C NMR ($CDCl_3$, 75 MHz) δ: 41.54, 55.07, 63.57, 72.23, 79.81, 86.11, 86.66, 86.96, 93.04, 99.57, 113.27, 120.18, 121.60, 123.89, 126.90, 127.43, 127.83, 127.97, 129.87, 129.96, 135.38, 135.52, 136.67, 136.87, 142.05, 143.43, 144.43, 148.88, 149.42, 155.31, 155.99, 158.6, 158.48, 161.42. HRMS (FAB) m/z for $C_{42}H_{37}N_4O_7$ $(M + H)^+$: calc'd. 709.2662, found 709.2679.

5′-*O*-(4,4′-Dimethoxytrityl)-5-((2,2'-bipyridin-6-yl)-ethylenyl)-2′-deoxyuridine, 1a. Dry **6a** (800 mg, 1.13 mmol) was dissolved in 300 mL of dry MeOH and added to 30 mL MeOH solution containing 400 mg of Pd/C previously activated by stirring under H_2 (50 psi) for 20 m at room temperature. The reaction mixture was further stirred under H_2 (50 psi) at room temperature until complete consumption of the starting materials (2 days). The solution was then filtered, reduced in volume to 0.5 mL, and purified by chromatography on a silica gel column pre-equilibrated with 1% Et_3N (by vol.) in CH_2Cl_2 and eluted with 10:0.0, 9.9:0.1, 9.8:0.2, 9.7:0.3, and 9.6:0.4 CH_2Cl_2/MeOH (v/v). A pale yellow foam of **1a** was obtained (706 mg, 87% yield).^{1}H NMR (CD_2Cl_2, 300 MHz) δ: 1.78 (1H, m, $H_{2'\beta}$), 2.15 (1H, m, $H_{2'\alpha}$), 2.45 (2H, t, $J = 7.9$ Hz, CH_2), 2.83 (2H, t, $J = 7.9$ Hz, CH_2), 3.02 (1H, m, $H_{5'\beta}$), 3.16 (1H, m, $H_{5'\alpha}$), 3.33 (1H, d, $J = 3$ Hz, $OH_{3'}$), 3.61 (3H, s, DMT-OCH_3), 3.62 (3H, s, DMT-OCH_3), 3.80 (1H, q, $J = 3.3$ Hz, $H_{4'}$), 3.95 (1H, m, $H_{3'}$), 6.15 (1H, t, $J = 6.3$ Hz, $H_{1'}$), 6.68–6.82 (5H, m, Ar), 7.00 (1H, s, H_6), 7.08–7.28 (10H, m, Ar), 7.54 (1H, t, $J = 7.8$ Hz, Ar), 7.70 (1H, dt, $J = 1.8$, 7.5 Hz, Ar), 8.45 (1H, m, Ar), 8.16 (1H, m, Ar), 8.47 (1H, m, Ar), 9.08 (1H, br, H_{N3}).^{13}C (CD_2Cl_2, 75 MHz) δ: 27.05, 36.88, 40.82, 46.34, 55.49, 63.58, 71.00, 84.33, 85.38, 86.76, 113.45, 114.37, 118.92, 121.88, 123.68, 123.99, 127.21, 128.22, 128.40, 130.34, 135.84, 135.87, 136.56, 137.29, 137.40, 144.91, 149.21, 150.66, 155.88, 156.81, 160.75, 163.71. HRMS (FAB) m/z for $C_{42}H_{41}N_4O_7$ $(M + H)^+$: calc'd. 713.2975, found 713.3003.

3′,5′-Di-*O*-acetyl-5-((2,2′-bipyridin-6-yl)-ethynyl)-2′-deoxyuridine, 6b. In a glove box, **4** (2.80 g, 15.50 mmol), THF (44 mL), Et_3N (11 mL), PdL_4 (1.79 g, 1.55 mmol), CuI (0.59 g, 3.10 mmol), and **5b** (6.80 g, 15.50 mmol) were combined in a flask, and the flask sealed with a septum. The flask was removed from the glove box, and the mixture was stirred and

heated to 60°C for 3 h in an oil bath. The contents of the flask were dried onto silica gel. A silica gel pad filtration was done to remove metals and salts using $CHCl_3$, 25% $CHCl_3$ (by vol.) in ethyl acetate, and 5% MeOH (by vol.) in ethyl acetate as eluents. The eluate was concentrated and loaded onto a flash silica column. Elution using CH_2Cl_2, 50% ethyl acetate (by vol.) in CH_2Cl_2, and ethyl acetate separated the desired product from the bulk material. Concentration gave a foam that was recrystallized from ethanol to give **6b** (4.11 g, 8.38 mmol, 54% yield) as a white solid.[1]H NMR ($CDCl_3$, 300 MHz) δ: 2.13 (3H, s, CH_3), 2.26 (3H, s, CH_3), 2.27 (1H, m, $H_{2'\beta}$), 2.59 (1H, ddd, $J = 3.0$, 6.0, 14.0 Hz, $H_{2'\alpha}$), 4.33 (1H, q, $J = 3.0$ Hz, $H_{4'}$), 4.40 (2H, m, $H_{5'\beta}/H_{5'\alpha}$), 5.27 (1H, dt, $J = 3.0$, 6.0 Hz, $H_{3'}$), 6.35 (1H, dd, $J = 6.0$, 8.0 Hz, $H_{1'}$), 7.33 (1H, m, Ar), 7.56 (1H, m, Ar), 7.82 (2H, m, Ar), 8.09 (1H, s, H_6), 8.41 (2H, m, Ar), 8.68 (1H, m, Ar), 10.41 (1H, br, H_{N3}).[13]C NMR ($CDCl_3$, 75 MHz) δ: 20.84, 20.93, 38.30, 63.78, 73.89, 79.60, 82.62, 85.51, 93.27, 100.12, 120.63, 121.34, 124.02, 127.33, 136.94, 137.07, 141.91, 142.71, 149.09, 149.12, 155.27, 156.46, 160.76, 170.27, 170.34. HRMS (FAB) m/z for $C_{25}H_{23}N_4O_7$ $(M + H)^+$: calc'd. 491.15667, found 491.15445.

3′,5′-Di-*O*-acetyl-5-((2,2′-bipyridin-6-yl)-ethylenyl)-2′-deoxyuridine, 1b. Compound 6b (545 mg, 1.11 mmol) and 10% Pd/C (254 mg) were placed in a 250 mL pressure vessel, and the vessel was flushed with nitrogen. MeOH (20 mL) was added, and the vessel was flushed repeatedly with hydrogen before finally being pressurized to 50 psi. After stirring under pressure for 2 d, the vessel was carefully depressurized, and the reaction mixture was filtered. The retentate was washed with copious amounts of chloroform and benzene. The filtrate was concentrated onto silica gel and chromatographed using 0, 2.5 and 5% MeOH (by vol.) in CH_2Cl_2. The product **1b** was obtained as a white solid (414 mg, 0.84 mmol, 75.3% yield).[1]H NMR ($CDCl_3$, 300 MHz) δ: 1.85 (1H, ddd, $J = 4$, 6, 21 Hz, $H_{2'\beta}$), 2.00 (3H, s, CH_3), 2.05 (3H, s, CH_3), 2.25 (1H, ddd, $J = 2$, 6, 14 Hz, $H_{2'\alpha}$), 2.85 (2H, m, CH_2), 3.09 (2H, t, $J = 7$ Hz, CH_2), 4.08 (2H, m, $H_{5'\beta}/H_{5'\alpha}$), 4.18 (1H, m, $H_{4'}$), 4.99 (1H, m, $H_{3'}$), 6.22 (1H, dd, $J = 6$, 9 Hz, $H_{1'}$), 7.11 (2H, m, Ar), 7.27 (1H, ddd, $J = 1$, 5, 8 Hz, Ar), 7.68 (1H, td, $J = 8$, 2 Hz, Ar), 8.18 (1H, d, $J = 8$ Hz, Ar), 8.38 (1H, d, $J = 8$ Hz, Ar), 8.64 (1H, m, Ar), 9.83 (1H, br, H_{N3}).[13]C NMR ($CDCl_3$, 75 MHz) δ: 20.68, 20.77, 27.07, 36.27, 37.03, 63.69, 74.02, 81.92, 84.49, 114.60, 118.49, 120.93, 123.20, 123.57, 135.19, 136.77, 137.17, 149.09, 150.36, 155.40, 156.16, 159.76, 163.33, 169.99, 170.21. HRMS (FAB) m/z for $C_{25}H_{27}N_4O_7$ $(M + H)^+$: calc'd. 495.1879, found 495.1889.

5-((2,2′-Bipyridin-6-yl)-ethynyl)-2′-deoxyuridine, 6c. To a solution of **5c** (1.062 g, 3.00 mmol), PdL_4 (0.173 g, 0.15 mmol), CuI (0.057 g, 0.30 mmol), and Et_3N (2.1 mL, 15 mmol) in DMF (15 mL) was added **4**

(0.702 g, 1.30 mmol) in a glove box. The reaction mixture was stirred at 50–60°C for 4 h and then chromatographed on a silica gel column eluted with 10:0.0, 9.9:0.1, 9.8:0.2, 9.7:0.3, 9.6:0.4, 9.3:0.07, and 9.0:0.1 CH_2Cl_2/MeOH (v/v). A yellow foam of **6c** was obtained (1.12 g, 91% yield).^{1}H NMR (DMSO-d_6, 300 MHz) δ: 2.11–2.25 (2H, m, $H_{2'\beta}/H_{2'\alpha}$), 3.55–3.70 (2H, m, $H_{5'\beta}/H_{5'\alpha}$), 3.82 (1H, q, $J = 3.3$ Hz, $H_{4'}$), 4.26 (1H, quintet, $J = 4.5$ Hz, $H_{3'}$), 5.21 (1H, t, $J = 4.8$ Hz, $OH_{5'}$), 5.28 (1H, d, $J = 4.5$ Hz, $OH_{3'}$), 6.13 (1H, t, $J = 6.6$ Hz, $H_{1'}$), 7.53 (1H, m, Ar), 7.6 (2H, d, $J = 7.5$ Hz, Ar), 7.92–8.0 (2H, m, Ar), 8.35 (2H, d, $J =$ 7.2, Ar), 8.50 (1H, s, H_6), 8.75, (1H, d, $J =$ 4.5 Hz, Ar), 11.77 (1H, s, H_{N3}).^{13}C NMR (DMSO-d_6, 75 MHz) δ: 60.81, 69.90, 82.28, 85.04, 87.66, 91.56, 97.30, 120.07, 120.74, 124.62, 127.47, 137.50, 138.05, 142.03, 145.13, 149.42, 149.44, 154.46, 155.76, 161.44. HRMS (FAB) m/z for $C_{21}H_{19}N_4O_5$ $(M + H)^+$: calc'd. 407.1355, found 407.1373.

5-((2,2′-Bipyridin-6-yl)-ethylenyl)-2′-deoxyuridine, 1c. Dry **6c** (200 mg, 0.49 mmol) was dissolved in dry MeOH (100 mL) and added to 30 mL of MeOH solution containing 250 mg of 10% Pd/C previously activated by stirring under H_2 (50 psi) for 20 m at room temperature. The reaction mixture was further stirred under H_2 (50 psi) at room temperature until complete consumption of the starting material (2 days). The solution was then filtered, reduced in volume to 0.5 mL, and chromatographed on a silica gel column eluted with 10:0.0, 9.9:0.1, 9.8:0.2, 9.7:0.3, 9.6:0.4, 9.3:0.07, and 9.0:0.1 CH_2Cl_2/MeOH (v/v). A pale yellow foam of **1c** was obtained (125 mg, 62% yield).^{1}H NMR (DMSO-d_6, 300 MHz) δ: 1.88–1.95 (2H, m, $H_{2'\beta}/H_{2'\alpha}$), 2.60–2.78 (2H, m, CH_2), 2.99 (2H, t, $J = 7.8$, CH_2), 3.43–3.54 (2H, m, $H_{5'\beta}/H_{5'\alpha}$), 3.711 (1H, q, $J = 2.7$ Hz, $H_{4'}$), 4.14 (1H, m, $H_{3'}$), 5.01 (1H, t, $J = 4.8$ Hz, $OH_{5'}$), 5.195 (1H, d, $J = 3.9$ Hz, $OH_{3'}$), 6.10 (1H, t, $J = 7.2$ Hz, $H_{1'}$), 7.28 (1H, d, $J = 7.5$ Hz, Ar), 7.42 (1H, m, Ar), 7.62 (1H, s, H_6), 7.82 (1H, t, $J = 7.5$ Hz, Ar), 7.91 (1H, dt, $J = 1.2$, 7.5 Hz, Ar), 8.37 (1H, d, $J =$ 7.8, Ar), 8.65 (1H, d, $J =$ 6 Hz, Ar), 11.25 (1H, s, H_{N3}).^{13}C NMR (DMSO-d_6, 75 MHz) δ: 26.26, 35.86, 61.32, 70.45, 83.80, 87.29, 112.86, 117.95, 120.50, 123.28, 124.10, 136.52, 137.24, 137.60, 149.26, 150.32, 154.60, 155.43, 160.21, 163.43. HRMS (FAB) m/z for $C_{21}H_{22}N_4O_5$ $(M + H)^+$: calc'd. 411.1668, found 411.1665.

5-[2-(4-(6,7-Dihydrodipyrido[1,2-a;2′,1′-c]pyrazinediylium))-ethylenyl]- 2′-deoxyuridine^{2+}, 7. In a Schlenk tube, **6c** (100 mg, 0.024 mmol) was co-evaporated with dry THF three times in vacuo. Freshly distilled 1,2-dibromo-ethane (4 mL) was added to **6c** in a glove box. The mixture was degassed three times at –198°C (2 x 10^{-4} torr) on vacuum manifold. The solution was refluxed with stirring at 100°C for 24 h. The yellow precipitate of **7** was filtered off, washed with acetone, CH_2Cl_2, and THF and dried on a vacuum manifold (58 mg, 40% yield).^{1}H NMR (DMSO-d_6, 300 MHz) δ: 2.09 (2H, t, $J = 6.0$ Hz, $H_{2'\beta}/H_{2'\alpha}$), 2.71–2.76 (2H, m, CH_2), 3.49–3.60 (4H, m,

CH_2/ $H_{5'\beta}$/$H_{5'\alpha}$), 3.76 (1H, q, $J = 3.6$ Hz, $H_{4'}$), 4.21 (1H, m, $H_{3'}$), 4.96 (1H, t, $J = 5.1$ Hz, $OH_{5'}$), 5.22 (1H, d, $J = 4.2$ Hz, $OH_{3'}$), 5.29 (4H, s, CH_2CH_2), 6.13 (1H, t, $J = 6.6$ Hz, $H_{1'}$), 7.83 (1H, s, H_6), 8.31 (1H, d, $J = 7.8$ Hz, Ar), 8.45 (1H, t, $J = 7.5$ Hz, Ar), 8.83 (1H, t, $J = 8.1$ Hz, Ar), 8.91–9.00 (2H, m, Ar), 9.06 (1H, d, $J = 8.1$ Hz, Ar), 9.38 (1H, d, $J = 5.7$ Hz, Ar), 11.43 (1H, s, H_{N3}).[13]C NMR (CD_3OD, 75 MHz) δ: 26.31, 34.38, 41.36, 53.49, 62.72, 71.98, 86.56, 88.96, 111.77, 128.38, 129.75, 131.42, 133.79, 140.43, 142.30, 147.84, 148.01, 149.06, 151.99, 161.32, 165.90. HRMS (FAB) m/z for the singly reduced ($M^{\bullet+}$) diquat $(C_{23}H_{26}N_4O_5)^{2+}$:[56,57] calc'd. 438.1903, found 438.1935.

REFERENCES

1. Kaes, C.; Katz, A.; Hosseini, M. Chem. Rev. **2000**, *100*, 3553–3590.
2. Philip, D.; Stoddart, J.F. Angew. Chem., Int. Ed. Engl. **1996**, *35*, 1154–1196.
3. Achar, S.; Puddephatt, R. J. Angew. Chem., Int. Ed. **1994**, *33*, 847–849.
4. Buzek, J.; Kuderova, A.; Pexa, T.; Stankova, V.; Lauerova, L.; Palecek, E. J. Biomol. Struct. Dyn. **1999**, *17*, 41–50.
5. Dowell, R. I.; Hales, N. H.; Tucker, H. Eur. J. Med. Chem. **1993**, *28*, 513–516.
6. Kodaka, M.; Kubota, Y. J. Chem. Soc., Perkin Trans. 2 **1999**, 891–894.
7. Ryabov, A.D.; Firsova, Y.N.; Ershov, A.Y.; Dementiev, I.A. J. Biol. Inorg. **1999**, *4*, 175–182.
8. Ryabova, E.S.; Goral, V.N.; Csöregi, E.; Mattiasson, B.; Ryabov, A.D. Angew. Chem. Int. Ed. Engl. **1999**, *38*, 804–807.
9. Zhang, B.; Breslow, R. J. Am. Chem. Soc. **1997**, *119*, 1676–1681.
10. Zhou, X.-W.; Zhuang, Z.-L.; Chen, Q.-X. J. Protein Chem. **1999**, *18*, 735–740.
11. Beer, P. D. Acc. Chem. Res. **1998**, *31*, 71–80.
12. Chin, T.; Gao, Z.; Lelouche, I.; Shin, Y.-G.; Purandare, A.; Knapp, S.; Isied, S. S. J. Am. Chem. Soc. **1997**, *119*, 12,849–12,858.
13. Gust, D.; Moore, T. A. Science **1989**, *244*, 35–41.
14. Chujo, Y.; Sada, K.; Saegusa, T. Macromolecules **1993**, *26*, 6320–6323.
15. Eisenbach, C.D.; Goeldel, A.; Terskan-Reiold, M.; Schubert, U. S. In *Polymeric Materials Encyclopedia*; Salamone, J.C., Ed.; CRC Press: Boca Raton, 1996; Vol. 10, p 8162.
16. Dupray, L.M.; Devenney, M.; Striplin, D.R.; Meyer, T.J.J. Am. Chem. Soc. **1997**, *119*, 10,243–10,244.
17. Wang, B.; Wasielewski, M.R. J. Am. Chem. Soc. **1997**, *119*, 12–21.
18. Ziessel, R. In *Photosensitization and Photocatalysis Using Inorganic and Organometalic Compounds*; Kalyanasundaram, K., Gratzel, M., Eds.; Kluwer Academic Publishers: Dordecht, 1993, p 217.
19. Telser, J.; Cruickshank, K.A.; Schanze, K.S.; Netzel, T.L.J. Am. Chem. Soc. **1989**, *111*, 7221–7226.
20. Meade, T.J.; Kayyem, J.F. Angew. Chem., Int. Ed. **1995**, *34*, 352–354.
21. Hall, D.B.; Holmlin, R.E.; Barton, J.K. Nature **1996**, *382*, 731–735.
22. Stemp, E.D.; Barton, J.K. Met. Ions Biol. Syst. **1996**, *32*, 325–365.
23. Dandliker, P.J.; Holmlin, R.E.; Barton, J.K. Science **1997**, *275*, 1465–1468.

24. Arkin, M.R.; Stemp, E.D.A.; Pulver, S.C.; Barton, J.K. Chem. Biol. **1997**, *4*, 389–400.
25. Hurley, D.J.; Tor, Y.J. Am. Chem. Soc. **1998**, *120*, 2194–2195.
26. Weizman, H.; Tor, Y.J. Am. Chem. Soc. **2001**, *123*, 3375–3376.
27. Dreyer, G.B.; Dervan, P.B. Proc. Natl. Acad. Sci. U.S.A. **1985**, *82*, 968–972.
28. Chen, C.-H.B.; Sigman, D.S.J. Am. Chem. Soc. **1988**, *110*, 6570–6572.
29. Bashkin, J.K.; Frolova, E.I.; Smapath, U. J. Am. Chem. Soc. **1994**, *116*, 5981–5982.
30. Matsumura, K.; Endo, M.; Komiyama, M. J. Chem. Soc., Chem. Commun. **1994**, 2019.
31. Slama-Schwok, A.; Ottolenghi, M.; Avnir, D. Nature **1992**, *355*, 240–242.
32. Wantanabe, T.; Honda, K. J. Phys. Chem. **1982**, *86*, 2617–2619.
33. Farrington, J.A.; Ebert, M.; Land, E.J. J. Chem. Soc., Faraday Trans. 1 **1978**, *74*, 665–675.
34. Thornton, N.B.; Wojtowicz, H.; Netzel, T.L.; Dixon, D.W.J. Phys. Chem. B **1998**, *102*, 2101–2110.
35. Netzel, T. L.; Zhao, M.; Nafisi, K.; Headrick, J.; Sigman, M. S.; Eaton, B. E. J. Am. Chem. Soc. **1995**, *117*, 9119–9128.
36. Manoharan, M.; Tivel, K. L.; Zhao, M.; Nafisi, K.; Netzel, T.L.J. Phys. Chem. **1995**, *99*, 17,461–17,472.
37. Kerr, C. E.; Mitchell, C. D.; Headrick, J.; Eaton, B.E.; Netzel, T. L. J. Phys. Chem. B **2000**, *104*, 1637–1650.
38. Kerr, C.E.; Mitchell, C.D.; Ying, Y.-M.; Eaton, B.E.; Netzel, T.L.J. Phys. Chem. B **2000**, *104*, 2166–2175.
39. Kerr, C.E.; Eaton, B.E.; Netzel, T.L. Nucleosides, Nucleotides & Nuc. Acids **2000**, *19*, 851–866.
40. Garber, T.; Rillema, P. Synth. Commun. **1990**, *20*, 1233–1239.
41. Fraser, C.L.; Anastasi, N.R.; Lamba, J.J.S. J. Org. Chem. **1997**, *62*, 9314–9317.
42. Grosshenny, V.; Romero, F. M.; Ziessel, R. J. Org. Chem. **1997**, *62*, 1491–1500.
43. Parks, J.E.; Wagner, B.E.; Holm, R.H.J. Organomet. Chem. **1973**, *56*, 53–66.
44. Polin, J.; Schmohel, E.; Balzani, V. Synthesis **1998**, 321–324.
45. Romero, F.M.; Ziessel, R. Tetrahedron Lett. **1995**, *36*, 6471–6474.
46. Yamamoto, Y.; Azuma, Y.; Mitoh, H. Synthesis **1986**, *7*, 564–565.
47. Schubert, U.S.; Eschbaumer, C.; Heller, M. Org. Lett. **2000**, *2*, 3373–3376.
48. Schubert, U.S.; Eschbaumer, C. Org. Lett. **1999**, *1*, 1027–1029.
49. Stille, J.K. Angew. Chem., Int. Ed. **1986**, *25*, 508–524.
50. Lehmann, U.; Henze, O.; Schluter, D. Chem. Eur. J. **1999**, *5*, 854–859.
51. Sonogashira, K.; Tohda, Y.; Hagihara, N. Tetrahedron Lett. **1975**, *50*, 4467–4470.
52. Classon, B.; Samuelsson, B. Acta Chem. Scand. B **1985**, *39*, 501–504.
53. Siegel, S. Heterogeneous Catalytic Hydrogenation of C=C and C+C In *Comprehensive Organic Synthesis*; Trost, B., Flemming, I., Eds.; Pergamon Press: Oxford, 1991, pp 417–442.
54. Gaballah, S.T.; Netzel, T.L. Nucleosides, Nucleotides & Nuc. Acids **2002**, *21*, in press.

55. Coulson, D.R. Inorg. Synth. **1972**, *13*, 121–124.
56. Ryan, T.M.; Day, R.J.; Cooks, R.G. Anal. Chem. **1980**, *52*, 2054–2057.
57. Dang, T.A.; Day, R.J.; Hercules, D.M. Anal. Chem. **1984**, *1984*, 866–871.

Received March 12, 2002
Accepted June 24, 2002