ORGANIC
LETTERS

1999
Vol. 1, No. 2
179−181

# Reduction of Azides to Primary Amines in Substrates Bearing Labile Ester Functionality. Synthesis of a PEG-Solubilized, “Y”-Shaped Iminodiacetic Acid Reagent for Preparation of Folate-Tethered Drugs[1]

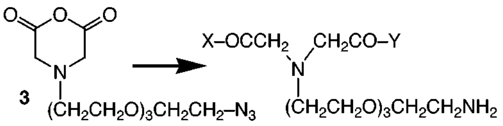
**Jae Wook Lee and Philip L. Fuchs***

*Department of Chemistry, Purdue University, West Lafayette, Indiana 47907*

*pfuchs@chem.purdue.edu*

**Received March 16, 1999**

## ABSTRACT



**Anhydride 3 is a useful reagent for the synthesis of triply linked drug conjugates. Examples using paclitaxel are provided. Conversion of the azido moiety to a primary amine in the presence of substrates bearing labile ester functionality requires the use of a tin/mercaptan reducing system which includes methanol exchange equilibration to effect nitrogen–tin bond scission.**

In conjunction with our program to utilize the folate receptor as a cell-specific mediator for the delivery of various oncolytic agents,[1] we required a general intermediate capable of accommodating covalent attachment of folic acid, an anticancer agent, and a polycarboxylic acid to increase water solubility (Figure 1, arrows a−c). In addition, we wished to exploit our synthesis of folates which employs the readily prepared pteroyl azide as a chemospecific reagent for “late” construction of the (typically insoluble) folate function via nitrogen acylation of the glutamate moiety (Figure 1, arrow d).[1b] Our initial target was iminodiacetic acid (IDA) anhydride **3** which enables sequential monoacylation of selected nucleophiles and still bears the azido moiety which serves as a latent amine. We selected **3** since the termini-differentiated tetraethylene glycol amino azide **4** can be inexpensively and easily prepared by the excellent method of Schwabacher.[2]

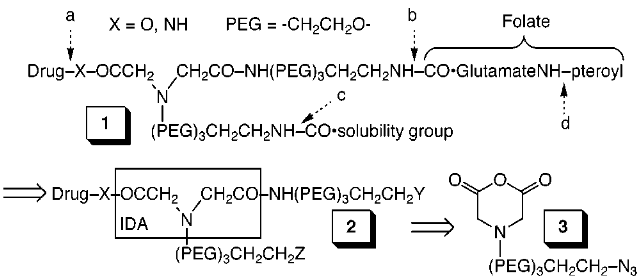


**Figure 1.**

Reaction of the Schwabacher amino azide **4**[2] with 2 equiv of allyl bromoacetate in the presence of potassium carbonate in acetonitrile for 30 h at 25 °C gave dialkylated product **5**[3] in 97% yield which was deprotected[4] by phenylsilane in the

---

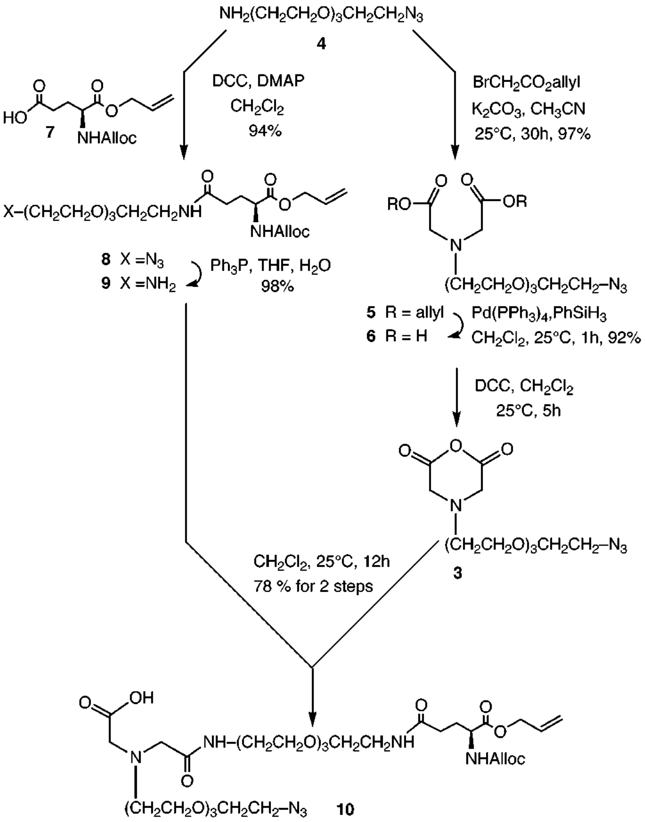
[1] Folate-Mediated Multiple Drug Delivery 3. For papers 1 and 2, see: (a) Wang, S.; Luo, J.; Lantrip, D. A.; Waters, D. J.; Mathias, C. J.; Green, M. A.; Fuchs, P. L.; Low, P. S. *Bioconjugate Chem.* **1997**, *8*, 673. (b) Luo, J.; Smith, M. D.; Lantrip, D. A.; Wang, S.; Fuchs, P. L. *J. Am. Chem. Soc.* **1997**, *119*, 10004.

[2] Schwabacher, A. W.; Lane, J. W.; Schiesher, M. W.; Leigh, K. M.; Johnson, C. W. *J. Org. Chem.* **1998**, *63*, 1727. We find that the use of 5% HCl in place of phosphoric acid improves the yield of **4** to 90%. For an earlier preparation of **4**, see: Bertozzi, C. R.; Bednarski, M. D. *J. Org. Chem.* **1991**, *56*, 4326−4329.

10.1021/ol9905248 CCC: $18.00

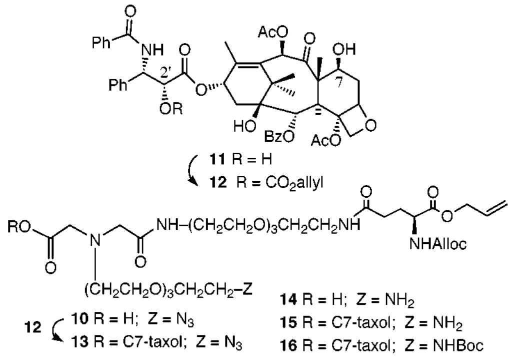
presence of 5% $Pd(PPh_3)_4$ to afford iminodiacetic acid **6** in 92% yield. Iminodiacetic acid **6** was initially treated with 1.0 equiv of DCC at room temperature to generate anhydride **3**. In a separate sequence, amino azide **4** was acylated with the readily available bis-protected glutamate **7**[1b] in methylene chloride with 1.1 equiv of DCC and a catalytic amount of DMAP for 12 h at 25 °C to provide azide **8** in 94% yield (cf. 47% yield in THF). Reaction of **8** with triphenylphosphine[2,5] in THF and $H_2O$ gave the desired primary amine **9** in 98% yield. Addition of amine **9** to the preformed anhydride **3** and stirring for an additional 12 h at 25 °C smoothly provided glutamate **10** (Scheme 1).

**Scheme 1**



We needed to protect the C-2′ position of paclitaxel **11** to selectively functionalize the C-7 site.[6] Since we were employing allyl esters on the glutamate moiety, we elected to also protect the C-2′ position as an allyloxycarbonate to provide for eventual simultaneous deblocking of all three centers. 2′-Alloc paclitaxel **12** was obtained in 98% yield from reaction of paclitaxel **11** with allyl chloroformate and Hünig's base at 25 °C in methylene chloride. Similar reaction in THF only afforded **12** in 58% yield. Reaction of 2′-alloc paclitaxel **12** with carboxylic acid **10** in the presence of DCC and a catalytic amount of DMAP in methylene chloride for 12 h at 25 °C provided Y-shaped intermediate **13** in 93% yield after column chromatography (Scheme 2).

**Scheme 2**



Initially, we tried to reduce azide **13** to amine **15** using the triphenylphosphine method, but we could not isolate the pure compound. It was found that the phosphine imine intermediate formed satisfactorily, but hydrolysis was very slow and amine **14** was consumed at a rate competitive with its formation over the course of 48 h. Products arising from transamination and ester hydrolysis were observed. At this stage, a variety of azide reduction methods[7−10] (NaHTe, $LiMe_2NBH_3$, tin hydrides, and TMS-Cl/NaI) were surveyed with essentially no success. To preserve our supply of **13**, we returned to azide **8** as a model compound. The TMSCl/NaI method was ineffective, and $HSCH_2CH_2CH_2SH/Et_3N/MeOH$ underwent reaction with the ester functionality. The first encouraging reaction utilized $SnCl_2/PhSH/Et_3N$,[11] which quickly and completely consumed the starting material and gave product **9** without affecting the ester group (Scheme 1).

Application of this method also rapidly reduced compound **10** to amine **14** but revealed the problem of effective product isolation. The recommended protocol[11] employs a strongly basic workup procedure (2 N NaOH) to destroy the tin complex, but is not applicable to our substrates because of the labile ester functionality. The workup procedure was modified, utilizing an exchange method to transfer the tin from nitrogen to the oxygen of the methanol solvent.[9a] After the reaction of compound **10** with $SnCl_2/PhSH/Et_3N$ in acetonitrile, a weakly basic workup (saturated $NaHCO_3$ solution) served to neutralize the ammonium salt. The

---

(3) All new compounds have been characterized by proton NMR, carbon NMR, exact mass, and/or elemental analysis.

(4) Dessolin, M.; Guillerez, M.-G.; Thieriet, N.; Guibe, F.; Loffet, A. *Tetrahedron Lett.* **1995**, *36*, 5741.

(5) Frisch, B.; Boeckler, C.; Schuber, F. *Bioconjugate Chem.* **1996**, *7*, 180. (b) Wentworth, P., Jr.; Vandersteen, A. M.; Janda, K. D. *Chem. Commun.* **1997**, 759. (c) Urpi, F.; Vilarrasa, J. *Tetrahedron Lett.* **1986**, *27*, 4623. (d) Schoetzau, T.; Holletz, T.; Cech, D. *Chem. Commun.* **1996**, 387.

(6) Greenwald, R. B.; Gilbert, C. W.; Pendri, A.; Conover, C. D.; Xia, J.; Martinez, A. *J. Med. Chem.* **1996**, *39*, 424.

(7) Alvarez, S. G.; Fisher, G. B.; Singaram, B. *Tetrahedron Lett.* **1995**, *36*, 2567.

(8) Suzuki, H.; Kawaguchi, T.; Takaoka, K. *Bull. Chem. Soc. Jpn.* **1986**, *59*, 665.

(9) Hays, D. S.; Fu, G. C. *J. Org. Chem.* **1998**, *63*, 2796. (b) Samano, M. C.; Robins, M. J. *Tetrahedron Lett.* **1991**, *32*, 6293.

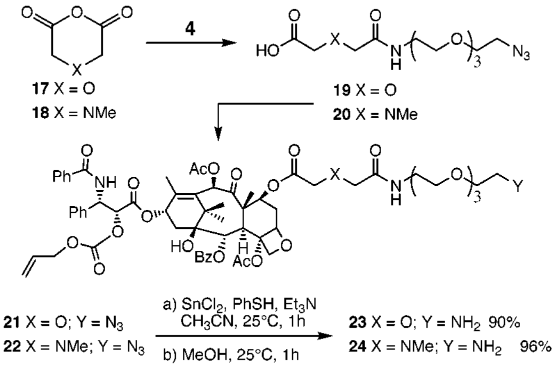
(10) Kamal, A.; Rao, N. V.; Laxman, E. *Tetrahedron Lett.* **1997**, *38*, 6945.

(11) Bartra, M.; Romea, P.; Urpi, F.; Vilarrasa, J. *Tetrahedron* **1990**, *46*, 587.

methylene chloride extract was evaporated followed by stirring the residue for 1 h at 25 °C in MeOH. Chromatography then provided model amine **14** in 75% yield. Repeating the protocol with paclitaxel-linked substrate **13** provided primary amine **15** in 96% yield with the methanol treatment. Addition of Boc anhydride at this stage of the methanol treatment afforded urethane **16** in 98% yield (Scheme 2).

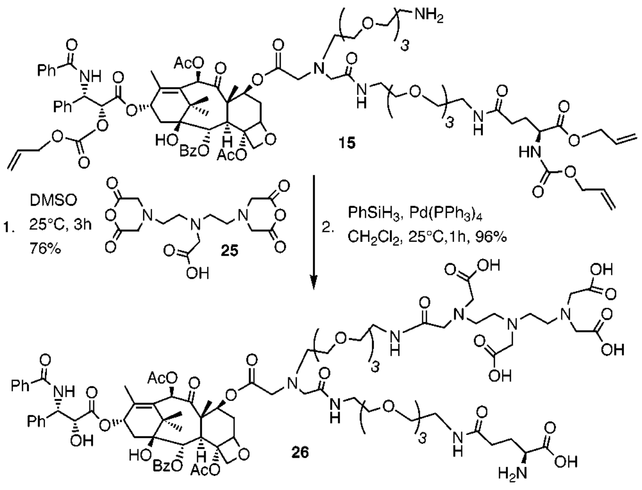
Two additional paclitaxel derivatives were prepared from reaction of the Schwabacher aminoazide **4** with 1.5 equiv of anhydrides **17** and **18** in $CH_2Cl_2$ for 26 h at 25 °C followed by additon of water and stirring for 10 h to consume the excess anhydride to afford the azidocarboxylic acids **19** and **20** in >98% yields. Acylation of paclitaxel derivative **12** using DCC and DMAP (catalytic) in $CH_2Cl_2$ for 12 h at 25 °C afforded key intermediates **21** and **22** in 92−94% yields. Application of the azide reduction methodology to these substrates smoothly provided primary amines **23** and **24** in >90% yield (Scheme 3).

**Scheme 3**



Installation of the polyacid functionality was accomplished by treatment of **15** with 7 equiv of DTPA dianhydride **25** in DMSO for 3 h at 25 °C. Water was added, and the reaction solution was stirred for several hours to destroy the excess anhydride. The resulting solution was lyophilized, and the residue was dissolved in $CH_2Cl_2$ and then filtered. The filtrate was concentrated and recrystallized from $CH_2Cl_2$/EtOAc/ether to provide a 76% yield of the tris-allyl-protected tetraacid (not shown) which was subjected to treatment with phenylsilane and palladium [0][4] to afford the desired triply deprotected amino pentaacid **26** in 96% yield (Scheme 4).

**Scheme 4**



The addition of the pteroyl unit to **26** to produce the target drug **1**, as well as pharmacological testing data, will be presented elsewhere.

**Acknowledgment.** We acknowledge Arlene Rothwell and Karl Wood for mass spectral data and Endocyte, Inc., for financial support of this project. Dr. Douglas Lantrip is thanked for assistance in manuscript preparation.

OL9905248