ORGANIC LETTERS

2006
Vol. 8, No. 19
4327−4330

Synthesis of a β-(1→3)-D-Rhamnotetraose by a One-Pot, Multiple Radical Fragmentation

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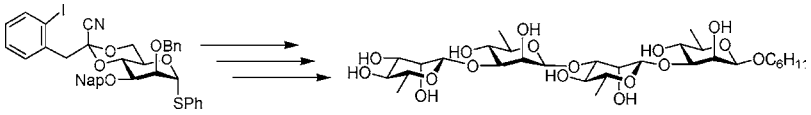
Received July 11, 2006

ABSTRACT



A naturally occurring β-(1→3)-D-rhamnotetraose has been constructed under conditions of sequential β-selective mannosylation controlled by the 4,6-*O*-[1-cyano-2-(2-iodophenyl)-ethylidene] protecting group. The route is concise, proceeding through a late-stage radical deoxygenation that successfully uncovers all four deoxy subunits at once.

6-Deoxy-D-mannose, called D-rhamnose, has been found exclusively in antigenic lipopolysaccharides (LPSs) associated with the cell walls of microorganisms.[1] Some examples include *Xanthomonas campestris*,[2] *Pseudomonas cepacia*,[1c] *P. syringae pv. Morspurunorum*,[3] *P. aeruginosa* IID 1008,[4] *P. maltophilia* 555,[5] *Myxobacterium* 402,[6] and *Escherichia hermanii*.[7] To date, this novel subunit has not been encountered in humans, plants, or animals. Given the rise in antimicrobial resistance, enzymes involved in the biosynthesis of the D-rhamnopyranosides would make promising targets for potentially xenobiotic antiinfectives.

E. hermanii is a member of the family of enterobacteriaceae, related to *E. coli*. It has been isolated from human wounds and sputum and has demonstrated pathogenicity against humans in vivo.[8] *E. hermanii* produces a β-lactamase and exhibits a distinctive antibiotic resistance penicillin, ampicillin, and carbenicillin.[9] Degradation studies have resulted in characterization of a repeat (1→3)-β-D-rhamnan from the cell walls of *E. hermanii* strain ATCC 33651.[10] The high content of the difficult β-D-rhamnosyl linkage combined with its potential medicinal relevance make this LPS O-chain constituent an appropriate candidate for development of methods aimed at synthesis of the β-D-rhamnopyranosides.

The stereoselective synthesis of the 1,2-*cis*-equatorial glycosidic bond as is found in both the β-mannosides and the β-rhamnosides is of perennial difficulty in carbohydrate chemistry.[11] Without the possibility of invoking neighboring group participation, the synthesis of such a linkage is

(1) (a) Hirooka, M.; Yoshimura, A.; Saito, I.; Ikawa, F.; Uemoto, Y.; Koto, S.; Takabatake, A.; Taniguchi, A.; Shinoda, Y.; Morinaga, A. *Bull. Chem. Soc. Jpn.* **2003**, *76*, 1409. (b) Spitali, M.; Smith, A. R. W. *J. Phytopath.* **2000**, *148*, 563. (c) Knirel, Y. A.; Shashkov, A. S.; Senchenkova, S. Y. N.; Ajiki, Y.; Fukuoka, S. *Carbohydr. Res.* **2002**, *337*, 1589. (d) Cerantola, S.; Montrozier, H. *Eur. J. Biochem.* **1997**, *246*, 360. (e) Winn, A. M.; Wilkinson, S. G. *Carbohydr. Res.* **1996**, *294*, 109. (f) Senchenkova, S. N.; Shashkov, A. S.; Kecskes, M. L.; Ahohuendo, B. C.; Knirel, Y. A.; Rudolph, K. *Carbohydr. Res.* **2000**, *329*, 831. (g) Vinogradov, E. V.; Campos-Portuguez, S.; Yokota, A.; Mayer, H. *Carbohydr. Res.* **1994**, *261*, 103. (h) Vinogradov, E. V.; Shashkov, A. S.; Knirel, Y. A.; Zdorovenko, G. M.; Solyanik, L. P.; Gubanova, N. Y.; Yakovleva, L. M. *Carbohydr. Res.* **1991**, *212*, 307. (i) Molinaro, A.; Silipo, A.; Lanzetta, R.; Newman, M.-A.; Dow, J. M.; Parrilli, M. *Carbohydr. Res.* **2003**, *338*, 277. (j) Beynon, L. M.; Bundle, D. R.; Perry, M. B. *Can. J. Chem.* **1990**, *68*, 1456.

(2) Hickman, J.; Ashwell, G. *J. Biol. Chem.* **1966**, *241*, 1424.

(3) Smith, A. R. W.; Zamze, S. E.; Munro, S. M.; Carter, K. J.; Hignett, R. C. *Eur. J. Biochem.* **1985**, *149*, 73.

(4) Yokota, S.; Kaya, S.; Sawada, S.; Kawamura, T.; Araki, Y.; Ito, E. *Eur. J. Biochem.* **1987**, *167*, 203.

(5) Di Fabio, J. L.; Perry, M. B.; Bundle, D. R. *Biochem. Cell Biol.* **1987**, *65*, 968.

(6) Morrison, I. M.; Young, R.; Perry, M. B.; Adams, G. A. *Can. J. Chem.* **1967**, *45*, 1987.

(7) Perry, M. B.; Bundle, D. R. *Infect. Immun.* **1990**, *58*, 1391.

(8) Dahl, K. M.; Barry, J.; DeBiasi, R. L. *Clin. Infect. Dis.* **2002**, *35*, 96.

(9) (a) Fitoussi, F.; Arlet, G.; Grimont, P. A. D. *J. Antimicrob. Chemother.* **1995**, *36*, 537. (b) Chaudhury, A.; Nath, G.; Tikoo, A.; Sanyal, S. C. *J. Diarr. Dis. Res.* **1999**, *17*, 85.

(10) Perry, M. B.; Richards, J. C. *Carbohydr. Res.* **1990**, *205*, 371.

10.1021/ol061706m CCC: $33.50
Published on Web 08/17/2006

rendered somewhat more sensitive than that of the *trans*-glycosidic bond. Captivated by this challenge, our group has found considerable success in employing the torsionally and electronically disarming 4,6-*O*-benzylidene protecting group.[12] In our researches, this group has been used to synthesize a variety of β-D-mannopyranosides, including the (1→2)-, (1→4)-, and alternating (1→3)-, (1→4)-mannans.[13] However, even with this technology in hand, the challenge of the *cis*-glycosidic bond is considerably magnified in the biologically important rhamnopyranosides, which lack the functional arm for incorporation of a benzylidene-type directing effect.

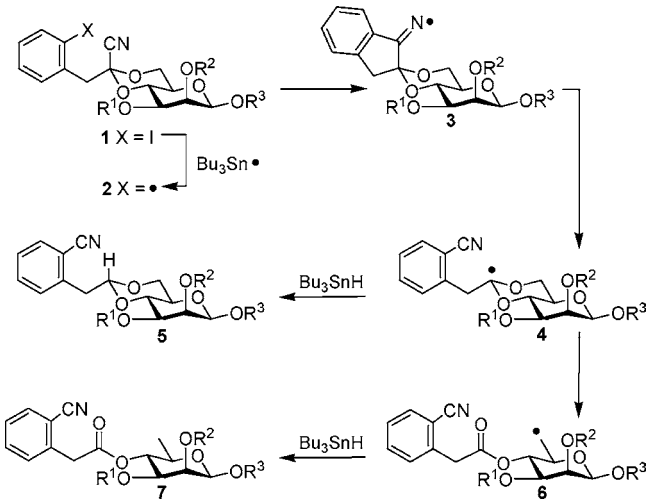
In general, strategies for synthesis of polysaccharides containing deoxy-sugars proceed via prior synthesis of an appropriately protected deoxy subunit, followed by extensive optimization of conditions for stereoselective glycosidation; recent efforts in this vein have been frustrated by low selectivities.[14] However, the reliability of the benzylidene-mediated mannosylations combined with their close structural relationship to the β-rhamnosides suggest inverting such a paradigm: synthesis of β-mannosyl linkages followed by a deoxygenation to provide the otherwise demanding subunits. This strategy is particularly attractive in the case of the β-D-rhamnopyranosides, where the starting material, D-mannose, is easily available in bulk. Thus, we have recently developed a protecting group that readily combines the stereoselectivity of a benzylidene acetal with a latent radical fragmentation pathway, providing a high-yielding deoxygenation in the last stage of oligosaccharide synthesis.[15] Herein, we demonstrate the broadest capabilities of this method to date, with a concise total synthesis of a tetrasaccharide fragment from the (1→3)-rhamnan of *E. hermanii* (ATCC 33651) via a one-pot quadruple radical fragmentation. To the best of our knowledge, this is the first synthesis of a β-rhamnan (of either the D- or L-modification) and an unique example of such a multiple radical deoxygenation.[16]

Benzylidene-protected hexopyranosides are known to undergo deoxygenation at C-6 via the NBS-mediated Hanessian−Hullar reaction.[17] However, the initiation step of this reaction, radical abstraction of the benzylidene proton, has proven indiscriminate in consort with the standard host of nonparticipating protecting groups necessary for oligosaccharide synthesis.[18] Similar incompatibilities are observed with Roberts' thiol-catalyzed benzylidene fragmentation.[19] Whereas the Hanessian−Hullar reaction likely occurs via a radical/polar crossover mechanism, Roberts' sequence proceeds via a purely radical mechanism.[20] This mechanism favors fragmentation to a primary radical at C-6 due to a conformationally less-strained transition state arising from planarization at the incipient C-6 radical.[21]

To avoid the problematic hydrogen atom abstraction step, we introduced the 4,6-*O*-[α-(2-(2-iodophenyl)-ethylthiocarbonyl)-benzylidene] group.[22] This group enabled the synthesis of the tetrasaccharide subunit from *E. hermanii* (ATCC 33650 and 33652).[23] However, the limited functional group compatibility of a key transesterification required to introduce the group minimized the overall scope. In subsequent work, we have identified a second-generation 4,6-*O*-[1-cyano-2-(2-iodophenyl)-ethylidene] acetal as a surrogate for the benzylidene fragmentation that is easily prepared, is easily installed, and is orthogonal to many protecting group manipulations.

The mechanism for the cyano group transfer/fragmentation (Scheme 1) is based upon chemistry first articulated by

Scheme 1. Radical Fragmentation Mechanism



Beckwith and later expanded by Rychnovsky.[24] As is frequently the case with radical reactions propagated by tin

(11) (a) Barresi, F.; Hindsgaul, O. In *Modern Methods in Carbohydrate Synthesis*; Khan, S. H., O'Neill, R. A., Eds.; Harwood Academic Publishers: Amsterdam, 1996; pp 251. (b) Demchenko, A. V. *Synlett* **2003**, 1225. (c) Pozsgay, V. In *Carbohydrates in Chemistry and Biology*; Ernst, B., Hart, G. W., Sinay, P., Eds.; Wiley-VCH: Weinheim, Germany, 2000; Vol. 1, pp 319. (d) Gridley, J. J.; Osborn, H. M. I. *J. Chem. Soc., Perkin Trans. 1* **2000**, 1471.

(12) (a) Crich, D.; Li, H. *J. Org. Chem.* **2002**, *67*, 4640. (b) Crich, D.; Dai, Z. *Tetrahedron* **1999**, *55*, 1569. (c) Crich, D.; Barba, G. R. *Tetrahedron Lett.* **1998**, *39*, 9339. (d) Crich, D.; de la Mora, M. A.; Cruz, R. *Tetrahedron* **2002**, *58*, 35. (e) Crich, D.; Banerjee, A. *Org. Lett.* **2005**, *7*, 1395. (f) Crich, D.; Dudkin, V. *J. Am. Chem. Soc.* **2002**, *124*, 2263. (g) Dudkin, V. Y.; Crich, D. *Tetrahedron Lett.* **2003**, *44*, 1787. (h) Dudkin, V. Y.; Miller, J. S.; Danishefsky, S. J. *J. Am. Chem. Soc.* **2004**, *126*, 736. (i) Miller, J. S.; Dudkin, V. Y.; Lyon, G. J.; Muir, T. W.; Danishefsky, S. J. *Angew. Chem., Int. Ed.* **2003**, *42*, 431. (j) Nicolaou, K. C.; Mitchell, H. J.; Rodriguez, R. M.; Fylaktakidou, K. C.; Suzuki, H.; Conley, S. R. *Chem.−Eur. J.* **2000**, *6*, 3149. (k) Kim, K. S.; Kang, S. S.; Seo, Y. S.; Kim, H. J.; Lee, Y. J.; Jeong, K.-S. *Synlett* **2003**, 1311. (l) Wu, X.; Schmidt, R. R. *J. Org. Chem.* **2004**, *69*, 1853. (m) Kwon, Y. T.; Lee, Y. J.; Lee, K.; Kim, K. S. *Org. Lett.* **2004**, *6*, 3901. (n) Tanaka, S.-I.; Takashina, M.; Tokimoto, H.; Fujimoto, Y.; Tanaka, K.; Fukase, K. *Synlett* **2005**, 2325.

(13) (a) Crich, D.; Banerjee, A.; Yao, Q. *J. Am. Chem. Soc.* **2004**, *126*, 14930. (b) Crich, D.; Li, W.; Li, H. *J. Am. Chem. Soc.* **2004**, *126*, 15081. (c) Crich, D.; Li, H.; Yao, Q.; Wink, D. J.; Sommer, R. D.; Rheingold, A. L. *J. Am. Chem. Soc.* **2001**, *123*, 5826.

(14) Bedini, E.; Carabellese, A.; Barone, G.; Parrilli, M. *J. Org. Chem.* **2005**, *70*, 8064.

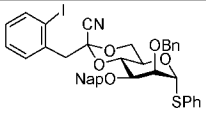
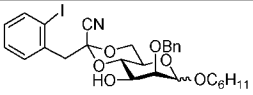
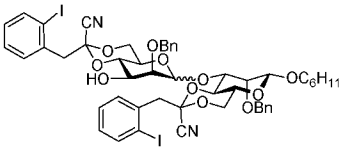
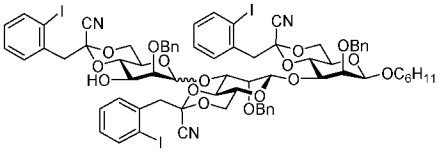
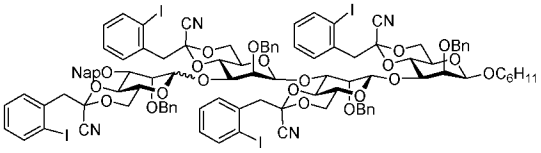
(15) Crich, D.; Bowers, A. A. *J. Org. Chem.* **2006**, *71*, 3452.

(16) For a previous example of a multiple radical fragmentation in oligosaccharide synthesis, see: Crich, D.; Hermann, F. *Tetrahedron Lett.* **1993**, *34*, 3385.

(17) (a) Hanessian, S.; Plessas, N. R. *J. Org. Chem.* **1969**, *34*, 1035. (b) Hanessian, S.; Plessas, N. R. *J. Org. Chem.* **1969**, *34*, 1045. (c) Hanessian, S.; Plessas, N. R. *J. Org. Chem.* **1969**, *34*, 1053. (d) Chana, J. S.; Collins, P. M.; Farnia, F.; Peacock, D. J. *J. Chem. Soc., Chem. Commun.* **1988**, 94. (e) Binkley, R. W.; Goewey, G. S.; Johnston, J. C. *J. Org. Chem.* **1984**, *49*, 992. (f) Hanessian, S. *Org. Synth.* **1987**, *65*, 243. (g) Hullar, T. L.; Siskin, S. B. *J. Org. Chem.* **1970**, *35*, 225.

(18) Liotta, L. J.; Dombi, K. L.; Kelley, S. A.; Targontsidis, S.; Morin, A. M. *Tetrahedron Lett.* **1997**, *38*, 7833.

Table 1. Preparation of Fragmentation Precursor **18** by Iterative Glycosidation[a]

donor	acceptor	product	yield (selectivity)
 **11**	$C_6H_{11}OH$		**12**: β; **13**: α 94% (β:α = 6:1)
11	**12**		**14**: β; **15**: α 78% (β:α = 10:1)
11	**14**		**16**: β; **17**: α 79% (β:α = 8:1)
11	**16**		**18**: β; **19**: α 86% (β:α = 10:1)

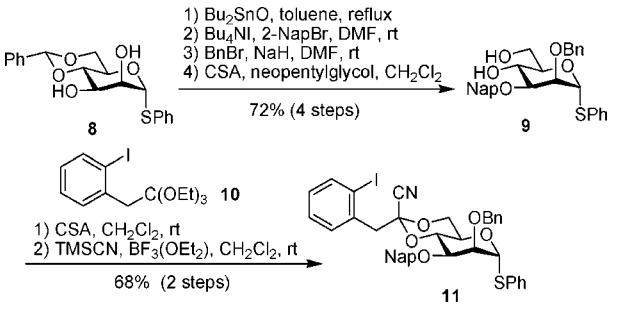
[a] Reagents: (1) Ph_2SO (1.5 equiv), TTBP (3.0 equiv), Tf_2O (1.7 equiv), CH_2Cl_2, −70/−20/−70 °C. (2) DDQ, CH_2Cl_2/H_2O (17:1).

hydrides, several competing reactions are possible, including premature reduction of radicals **2**−**4**, making the rapid rate of cyano group migration essential for our synthesis. The challenge of synthesizing a polymeric rhamnan by this methodology can be seen as one of minimizing a possible three different byproducts per monomer subunit or, for a tetrasaccharide, promoting one product in 3^4. With this in mind, we commenced our synthesis.

The synthesis of the (1→3)-tetrasaccharide required preparation of only one suitably protected monomer, **11**. This was achieved from diol **9**, which was prepared from 4,6-*O*-benzylidene-protected thiomannoside **8**[25] in 72% yield over four steps using standard reactions (Scheme 2). The 4,6-*O*-[1-cyano-2-(2-iodophenyl)-ethylidene] acetal was cleanly introduced as a single diastereomer via Lewis acid promoted transcyanation chemistry developed by Utimoto and coworkers.[26] Monomer **11** was then employed in the sequential, linear synthesis of a β-mannotetraose. After each coupling, deprotection was achieved prior to chromatographic purification of the newly synthesized oligomer. Thus, a single purification provided the acceptor for each subsequent step in the elaboration of the growing chain. All couplings resulted in high yields and high β-selectivities, as is consistent with a benzylidene-type directing effect (Table 1). The β-anomers could be readily assigned by their characteristic H-5 multiplets in the ^{1}H NMR spectrum, with the reducing end H-5 resonance at δ ~3.3 and subsequent residues further upfield still at δ 2.6−2.9.[27]

The fully protected tetrasaccharide was subjected to conditions of radical fragmentation in refluxing xylenes. In the development of the protecting group, it was found that the higher boiling point of xylenes favored the fragmentation pathway over the reduction of the benzylidene radical **4**.

Scheme 2. Synthesis of Donor **11**



(19) (a) Roberts, B. P.; Smits, T. M. *Tetrahedron Lett.* **2001**, *42*, 3663. (b) Dang, H.-S.; Roberts, B. P.; Sekhon, J.; Smits, T. M. *Org. Biomol. Chem.* **2003**, *1*, 1330. (c) Fielding, A. J.; Franchi, P.; Roberts, B. P.; Smits, T. M. *J. Chem. Soc., Perkin Trans. 2* **2002**, 155. (d) Cai, Y.; Dang, H.-S.; Roberts, B. P. *J. Chem. Soc., Perkin Trans. 1* **2002**, 2449. (e) Jeppesen, L. M.; Lundt, I.; Pedersen, C. *Acta Chem. Scand.* **1973**, *27*, 3579.

(20) (a) McNulty, J.; Wilson, J.; Rochon, A. C. *J. Org. Chem.* **2004**, *69*, 563. (b) Crich, D.; Bowers, A. A.; Yao, Q. *Carbohydr. Res.* **2006**, *341*, 1748.

(21) Roberts, B. P.; Dang, H.-S.; Cai, Y. *J. Chem. Soc., Perkin Trans. 1* **2002**, 2449.

(22) Crich, D.; Yao, Q. *Org. Lett.* **2003**, *5*, 2189.

(23) Crich, D.; Yao, Q. *J. Am. Chem. Soc.* **2004**, *126*, 8232.

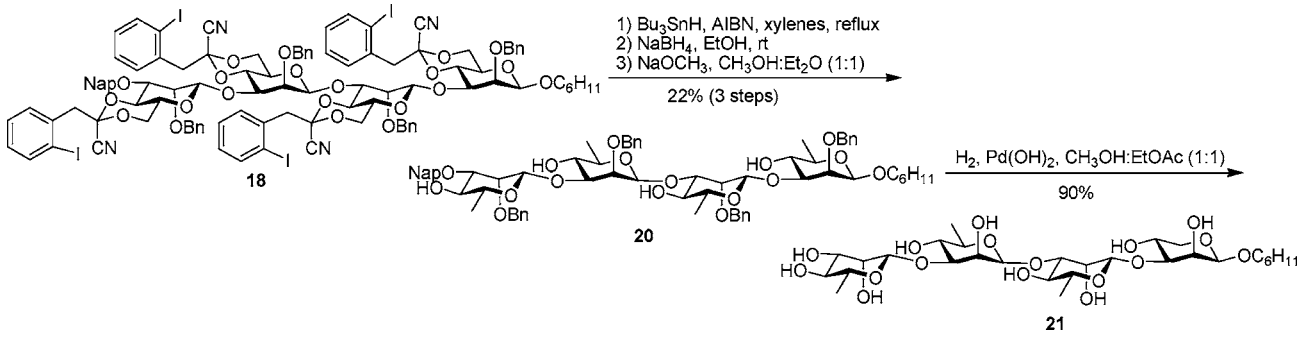
(24) (a) Beckwith, A. L. J.; Easton, C. J. *J. Am. Chem. Soc.* **1981**, *103*, 615. (b) Rychnovsky, S. D.; Swenson, S. S. *Tetrahedron* **1997**, *53*, 16489.

(25) Crich, D.; Li, W.; Li, H. *J. Am. Chem. Soc.* **2004**, *126*, 15081.

(26) Utimoto, K.; Wakabayashi, Y.; Horiie, T.; Inoue, M.; Shishiyama, Y.; Obayashi, M.; Nozaki, H. *Tetrahedron* **1983**, *39*, 967.

(27) That it was the residues of the nonreducing end with upfield resonances was confirmed by NOE experiments on the β-dimer **13**. In the NOE spectrum, a clear correlation was observed between the cyclohexyl proton and the reducing end anomeric proton, which was further correlated to the downfield H-5 signal. The upfield H-5 multiplet correlated with the remaining anomeric peak.

Scheme 3. Radical Fragmentation and Deprotection of Tetrasaccharide **18**



Adapting the conditions from the developmental work, a 4 h addition of tin hydride to a 0.0015 M solution of substrate in xylenes at reflux, followed by $NaBH_4$ reduction to facilitate removal of the tin residues, and then saponification allowed initial separation of the desired product cleanly on silica gel from traces of byproducts arising from incomplete fragmentation. The pure tetraol, **20**, was isolated in 22% yield from **18**. Subsequent global deprotection with palladium hydroxide and hydrogen proceeded to give the tetrasaccharide **21** in 90% yield (Scheme 3).

Table 2. Comparison of ^{1}H NMR Chemical Shifts and Coupling Constants of Internal Residues of **21** with Those of the Natural Rhamnan

	chemical shift (ppm)		coupling constant (Hz)		
H	isolated[a]	synthetic[b]	isolated[a]	synthetic[b]	$\Delta\delta$
H-1	4.81	4.66	*s*	*s*	−0.15
H-2	4.26	4.11	2.7	3.0	−0.15
H-3	3.87	3.72	9.7	9.5	−0.15
H-4	3.54	3.38	9.2	9.5	−0.16
H-5	3.46	complex	5.9	complex	
H-6	1.34	1.19	-	6.0	−0.15

[a] Data for the isolated polysaccharide from ref 10. [b] Values for the synthetic polysaccharide recorded in D_2O at room temperature at 500 MHz.

In both the ^{1}H and ^{13}C NMR spectra of the synthetic tetrasaccharide, resonances from the two end units can be distinguished from the compounded peaks of the internal subunits. Despite these differences, there is excellent agreement between shifts of the internal residues of the synthetic polymer and those of the natural tetrasaccharide, as is illustrated in Table 2.

In conclusion, we have developed a concise synthetic route to a β-(1→3)-D-rhamnotetraose, in which four challenging β-glycosidic linkages are installed with a high degree of stereoselectivity due to the disarming effect of the 4,6-*O*-[1-cyano-2-(2-iodophenyl)-ethylidene] protecting group. The key step is a late-stage radical deoxygenation, which occurs simultaneously on all four residues of the tetramer.

Acknowledgment. We thank the NIH (GM 57335) for financial support.

Supporting Information Available: Full experimental details and characterization data. This material is available free of charge via the Internet at http://pubs.acs.org.

OL061706M