JOC*Note*

Use of a Boroxazolidone Complex of 3-Iodo-L-tyrosine for Palladium-Catalyzed Cross-Coupling

William H. Walker, IV and Steven E. Rokita*

Department of Chemistry and Biochemistry, University of Maryland, College Park, Maryland 20742

rokita@umd.edu

Received November 18, 2002

Abstract: Complexation of 3-iodo-L-tyrosine with 9-borabicyclo[3.3.1]nonane (9-BBN) provides a convenient substrate for a palladium-catalyzed coupling reaction. The complex is stable to silica gel chromatography (hexanes/ethyl acetate), dilute triethylamine in THF, and potassium fluoride in DMF. The desired product, 3-ethynyl-L-tyrosine, was released from the complex by simply diluting its solution in methanol with chloroform. Interestingly, the complex remains stable in solutions of either methanol or chloroform individually.

As part of our laboratory's continuing interest in iodotyrosine deiodinase,[1] 3-ethynyl-L-tyrosine (**2**) was designed as a probe for its mechanism as well as a potential covalent label for its active site.[2] The Sonogashira reaction,[3,4] a palladium-catalyzed cross-coupling, was expected to provide a rapid and high-yielding synthesis of this unnatural amino acid. The mild conditions associated with the Sonogashira reaction were particularly attractive when compared to possible alternatives such as the Castro−Stephens coupling[5,6] that requires refluxing pyridine, or the classical synthesis of alkynes by dehydrohalogenation of vicinal or geminal dihalides that requires strong base. The harsh conditions of these latter procedures would likely lead to decomposition of the reactive ethynylphenol moiety.

Although a simple coupling might be envisioned for synthesis of **2** with ethynyltrimethylsilane and commercial 3-iodo-L-tyrosine (**1**) (see Scheme 1), the differing polarity of these reagents and lability of **2** necessitated use of an organic-soluble complex of the amino acid derived from 9-borabicyclo[3.3.1]nonane (9-BBN). Such complexes may offer a general and expedient method for solubilizing amino acids in organic solvents[7] and protecting the α-amino acid moiety from undesired reactions. The complex of 3-iodo-L-tyrosine in particular should provide a convenient entry into novel tyrosine analogues that serve as the structural core of various pharmaceuticals, neurotransmitters, and other biologically active molecules. Attempts at a one-step conversion of commercial 3-iodo-L-tyrosine (**1**) to an alkynyl derivative (**2**) failed when using suspensions of iodotyrosine in the solvents typically used for Sonogashira coupling, such as 1,4-dioxane, triethylamine, or a mixture of the two. No reaction occurred at ambient temperature, and at elevated temperatures, these mixtures underwent extensive decomposition. Protecting group strategies such as those used to generate ethynyltyrosine terpyridine ligands[8] were consequently unavoidable. Still, minimal protection was pursued to limit possible decomposition of the desired product during final deprotection.

The amino acid **1** was initially converted to its methyl ester, but cross-coupling of this derivative was also unsuccessful in the presence of $Cl_2(Ph_3P)_2Pd$ and CuI in triethylamine. Alternative protection of the α-amino group as the *N*-*tert*-butyl carbamate (BOC) similarly did not support the cross-coupling under equivalent conditions. In contrast, the fully protected *N*-BOC-3-iodo-L-tyrosine methyl ester efficiently cross-coupled with ethynyltrimethylsilane in yields of 96%. The resulting product was easily desilylated with KF in DMF to yield the terminal alkyne and hydrolysis of the methyl ester was conveniently promoted by α-chymotrypsin. In contrast, removal of the BOC protecting group proved to be troublesome since strongly acidic conditions (neat TFA, 10% TFA in CH_2Cl_2, or 1 M HCl in EtOAc) and even dilute aqueous acid (0.1 M HCl) resulted in hydration of **3** to form the acetyl derivative **4** (see Scheme 2).

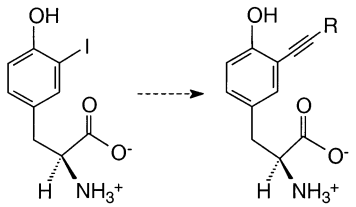
Other methods of protection, including those based on chelation, were then sought to protect both the amino and carboxylic acid groups simultaneously under mild conditions. Boron−amino acid complexes are generally formed under mild conditions and at ambient temperature.[7,9−12] The resulting products, boroxazolidones, are stable to both water and atmospheric oxygen, and yet are readily hydrolyzed in the presence of either aqueous acid or base depending on the nature of the substituents on boron.[11,12] Addition of excess ethylenediamine also releases the amino acid from its boroxazolidone complex,[7] but we found the mildest condition for decomplexation to be dilution of its solution in methanol with $CHCl_3$ (vide infra).

At first, the *B*,*B*-diphenyl boroxazolidone of 3-iodo-L-tyrosine was considered since this derivative was expected to be more stable than a corresponding alkyl derivative[13] and a number of such complexes of amino acids had already been described in the literature.[11,12,14−18] For synthetic ease, the air-stable reagent

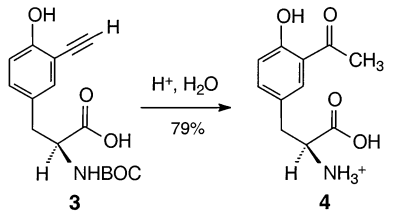
(1) Kunishima, M.; Friedman, J. E.; Rokita, S. E. *J. Am. Chem. Soc.* **1999**, *121*, 4722−4723.
(2) Walker, W. H.; Friedman, J. E.; Rokita, S. E. *Synthesis of Ethynyltyrosine as a Potential Mechanism-Based Inactivator*; Poster presentation at the National Meeting of the American Chemical Society, August 21, 2000, Washington, DC.
(3) Sonogashira, K.; Tohda, Y.; Hagihara, N. *Tetrahedron Lett.* **1975**, *50*, 4467−4470.
(4) Takahashi, S.; Kuroyama, Y.; Sonogashira, K.; Hagihara, N. *Synth. Commun.* **1980**, 627−630.
(5) Castro, C. E.; Stephens, R. D. *J. Org. Chem.* **1963**, *28*, 2163.
(6) Castro, C. E.; Havlin, R.; Honwad, V. K.; Malte, A.; Moje, S. *J. Am. Chem. Soc.* **1969**, *91*, 6464−6470.
(7) Dent, W. H.; Erickson, W. R.; Fields, S. C.; Parker, M. H.; Tromiczak, E. G. *Org. Lett.* **2002**, *4*, 1249−1251.
(8) Khatyr, A.; Ziessel, R. *Org. Lett.* **2001**, *3*, 1857−1860.
(9) Brown, H. C.; Gupta, A. K. *J. Organomet. Chem.* **1988**, *341*, 73−81.
(10) Miller, N. E. *Inorg. Chem.* **1974**, *13*, 1459−1466.
(11) Nefkens, G. H. L.; Zwanenburg, B. *Tetrahedron* **1983**, *39*, 2995−2998.
(12) Strang, C. J.; Henson, E.; Okamoto, Y.; Paz, M. A.; Gallop, P. M. *Anal. Biochem.* **1989**, *178*, 276−286.
(13) Baum, G. *J. Organomet. Chem.* **1970**, *22*, 269−271.

10.1021/jo0207022 CCC: $25.00
Published on Web 01/17/2003

SCHEME 1



SCHEME 2

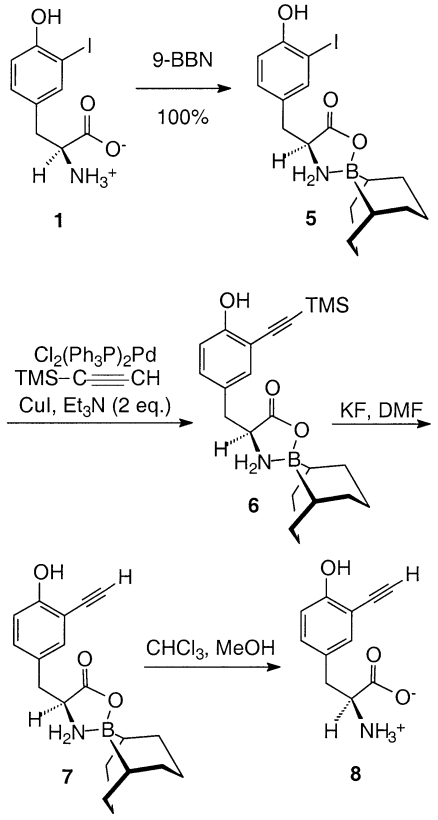


$NaBPh_4$ was selected over Ph_3B to generate the boroxazolidone. Although the desired product was obtained in high yield (94%), an 18 h reflux in aq HCl was required (Supporting Information). An alternative boron reagent was consequently sought that would complex the amino acid under more gentle conditions. Only a few bicyclononyl derivatives were known[9,11,18] but were nevertheless intriguing due to their convenient handling and the commercial availability of 9-BBN as a solid or solution in THF. Concurrent to the investigation described here, others were also exploring protection of amino acids with 9-BBN.[2,7] Use of 9-BBN in THF readily converted **1** to its boroxazolidone derivative **5** at ambient temperature in a quantitative yield (Scheme 3). Simple removal of solvent provided suitable material for immediate use in the Sonogashira cross-coupling reaction.

Excellent yields (99%) of the alkyne product **6** were obtained from the boroxazolidone **5** after cross-coupling with ethynyltrimethylsilane, $Cl_2(Ph_3P)_2Pd$, CuI, and 1.1−2.2 equiv of Et_3N in THF. The product complex **6** was stable to chromatography on silica gel (hexanes: EtOAc 50:50), and chelation was unperturbed by desilylation of the alkyne using KF in DMF to form **7** (78%). A proton source is apparently necessary for successful desilylation, as no loss of TMS was observed after **6** was treated with KF in anhydrous DMF.

The product complex **7** was significantly more soluble than the corresponding complex of tyrosine in chlorinated solvents,[7] and this difference in solubility may have contributed to the extremely mild conditions used for release of the desired amino acid, 3-ethynyl-L-tyrosine (**8**). Previously, decomplexation of amino acids from their bicyclononyl derivatives required either treatment with strong acid (HCl) or greater than 4 equiv of ethylenediamine.[7] Either condition had the potential to decompose

SCHEME 3



our labile product. Deprotection in this case was found to require merely diluting its solution in methanol with $CHCl_3$ (50-fold excess) and collecting the solid product **8** by filtration. In contrast, **7** is stable in methanol and $CHCl_3$ solutions individually. Decomplexation does not appear to be effected by the presence of trace quantities of HCl in the $CHCl_3$, since decomplexation still proceeds in $CHCl_3$ that has been freshly washed with saturated aqueous $NaHCO_3$. Perhaps reversible decomplexation of **7** and precipitation of the free amino acid **8** drive the overall deprotection process. A final purification with anion-exchange chromatography yielded the desired 3-ethynyl-L-tyrosine (**8**) (62%). None of the synthetic procedures for preparation of **8** caused any detectable racemization of the α-carbon as detected by the complete consumption of **8** by L-amino acid oxidase (>99.7%, Supporting Information).

Structural transformations of water-soluble compounds continue to challenge organic chemists due in part to the lack of compatible reagents and the high density of functional groups typically present. While numerous protecting groups are available to address these problems, conditions necessary for protection and deprotection can often add new limitations to synthetic approaches. Construction of 3-ethynyl-L-tyrosine (**8**) illustrates the advantage of 9-BBN as a selective protecting group during cross-coupling of aromatic amino acids. On the basis of the generality of forming boroxalidone complexes,[7] the synthesis of many other unnatural aromatic amino acids may be envisioned simply by selecting the appropriate amino acid precursor, coupling partner, and cross-coupling protocol.

(14) Staatz, I.; Granzer, U. H.; Blume, A.; Roth, H. J. *Liebigs Ann. Chem.* **1989**, 127−131.
(15) Skoog, I. H. *J. Org. Chem.* **1964**, *29*, 492−493.
(16) Rettig, S. J.; Trotter, J. *Can. J. Chem.* **1977**, *55*, 958−965.
(17) Farfan, N.; Silva, D.; Santillan, R. *Heteroatom Chem.* **1993**, *4*, 533−536.
(18) Gonzalez, A.; Granell, J.; Piniella, J. F.; Alvarez-Larena, A. *Tetrahedron* **1998**, *54*, 13313−13322.

Experimental Section

***N-tert*-Butyloxycarbonyl-3-trimethylsilylethynyl-L-tyrosine Methyl Ester.** *N-tert*-Butyloxycarbonyl-3-iodo-L-tyrosine methyl ester (1.0 g, 2.4 mmol), $Cl_2(Ph_3P)_2Pd$ (170 mg, 0.24 mmol), and CuI (20 mg, 0.11 mmol) were added to an oven-dried flask containing a magnetic stir bar and fitted with a rubber septum and nitrogen inlet. The solids were dried under high vacuum for 30 min with constant stirring under ambient temperature. Freshly distilled anhyd Et_3N (50 mL) was added via an oven-dried nitrogen-flushed syringe and the resulting yellow solution was stirred for 5 min. Ethynyltrimethylsilane (490 mg, 5.0 mmol, 710 μL) was added via an oven-dried nitrogen-flushed syringe causing an immediate precipitation of a fine white solid from a pale green solution. Within 10 min, the reaction mixture became a thick slurry and was vigorously stirred for 4 h at ambient temperature. The mixture was filtered and the solids were washed with ether. The filtrate and ether wash were combined and evaporated under reduced pressure. The resulting residue was purified by silica gel chromatography (hexanes:ethyl acetate, 70:30 v/v) to yield *N-tert*-butyloxycarbonyl-3-trimethylsilylethynyl-L-tyrosine methyl ester (310 mg, 96%) as a yellow oil. IR (thin film) 3500, 3400, 2150, 1750, 1700, 1250 cm^{-1}; ^1H NMR (400 MHz, CD_3OD) δ 0.25 (s, 9H), 1.37 (s, 9H), 2.74 (dd, J = 14.0, 8.8 Hz, 1H), 2.95 (dd, J = 14.0, 5.4 Hz, 1H), 3.67 (s, 3H), 4.25 (dd, J = 8.8, 5.4 Hz, 1H), 6.72 (d, J = 8.3 Hz, 1H), 6.99 (dd, J = 8.3, 2.0 Hz, 1H), 7.14 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ 0.01, 28.4, 32.1, 37.3, 51.8, 54.3, 100.2, 100.9, 109.7, 115.0, 127.7, 131.6, 132.8, 154.6, 156.8, 171.9; MS (EI) m/z 391.2 (M^+), 335.1, 318.1, 274.1, 259.0, 201.7.

***N-tert*-Butyloxycarbonyl-3-ethynyl-L-tyrosine Methyl Ester.** Solid KF (45 mg, 0.77 mmol) was added to a solution of *N-tert*-butyloxycarbonyl-3-trimethylsilylethynyl-L-tyrosine methyl ester (280 mg, 0.71 mmol) in DMF (reagent grade, 15 mL) and stirred at ambient temperature for 4 h. The solution was diluted with water (100 mL) and extracted with diethyl ether (3 × 50 mL). The combined ether layers were washed with water (3 × 50 mL) and saturated aqueous NaCl (50 mL), dried over anh $MgSO_4$, filtered, and evaporated under reduced pressure to yield *N-tert*-butyloxycarbonyl-3-ethynyl-L-tyrosine methyl ester (180 mg, 79%) as a pale yellow oil IR (thin film) 3287, 2231, 2106, 1750, 1700 cm^{-1}; ^1H NMR (400 MHz, CD_3OD) δ 0.20 (s, 9H), 1.37 (s, 9H), 2.75 (dd, J = 13.6, 8.8 Hz, 1H), 2.96 (dd, J = 13.6, 5.6 Hz, 1H), 4.25 (dd, J = 8.8, 5.6 Hz, 1H), 6.72 (d, J = 8.4 Hz, 1H), 6.99 (dd, J = 8.4, 2.0 Hz, 1H), 7.12 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ 28.4, 32.1, 37.3, 51.8, 79.2, 100.2, 109.7, 115.0, 127.7, 131.6, 132.6, 154.6, 156.8, 171.9.

***N-tert*-Butyloxycarbonyl-3-ethynyl-L-tyrosine (3).** A solution of α-chymotrypsin (8.0 mg, 410 units, bovine pancreas) in aq NH_4OAc (2 mL, 0.5 M, pH 6.0) was added to a solution of *N-tert*-butyloxycarbonyl-3-ethynyl-L-tyrosine methyl ester (180 mg, 0.56 mmol) in DMF (2 mL) with stirring at ambient temperature for 12 h.[19] The solution was diluted with water (10 mL) and extracted with EtOAc (3 × 10 mL). The combined EtOAc layers were washed with water (3 × 10 mL) and saturated aqueous NaCl (10 mL), dried over anhyd $MgSO_4$, filtered, and evaporated under reduced pressure to yield *N-tert*-butyloxycarbonyl-3-ethynyltyrosine (**3**) (100 mg, 60%) as an amorphous solid. IR (thin film) 3285, 2977, 2932, 2150, 1670, 1595, 1508, 1418 cm^{-1};^1H NMR (400 MHz, CD_3OD) δ 1.39 (s, 9H), 2.80 (m, 1H), 3.03 (m, 1H), 3.52 (s, 1H), 4.10 (m, 1H), 6.70 (d, J = 8.4 Hz, 1H), 7.06 (dd, J = 8.4, 2.0 Hz, 1H), 7.19 (d, J = 2.0 Hz, 1H).

3-Acetyl-L-tyrosine (4). A solution of *N-tert*-butyloxycarbonyl-3-ethynyl-L-tyrosine (5.0 mg, 16 mmol) in 10% TFA in CH_2Cl_2 (1.0 mL) was stirred at ambient temperature for 1 h. The mixture was diluted with water (5 mL) and extracted with CH_2Cl_2 (3 × 5 mL), and the aqueous layer was evaporated to yield crude 3-acetyl-L-tyrosine (**4**) (2.9 mg, 79%). ^1H NMR (400 MHz, CD_3OD) δ 2.55 (s, 3H), 2.96 (dd, J = 14.8, 8.0 Hz, 1H), 3.14 (dd, J = 14.8, 4.8 Hz, 1H), 3.72 (dd, J = 8.0, 4.8 Hz, 1H), 6.82 (d, J = 8.4 Hz, 1H), 7.35 (dd, J = 8.4, 2.0 Hz, 1H), 7.71 (d, J = 2.0 Hz, 1H); MS (FAB) m/z 246.0 (Na salt), 223.9 [M + H]$^+$, 207.0, 178.0, 158.9, 115.0. These data correspond to those previously reported for this compound.[20,21]

3-Iodo-L-tyrosinato-bicyclononylboron (5). 3-Iodo-L-tyrosine (**1**) (1.00 g, 3.26 mmol) was suspended in anhyd THF (5 mL) and vigorously stirred for 5 min. A solution of 9-BBN in THF (0.5 M, 9.0 mL) was added via syringe and the resulting suspension was stirred under N_2 at ambient temperature until all materials had dissolved (12 h).[11] The product was precipitated by addition of cyclohexane, filtered, and recrystallized from CH_3CN−water to yield 3-iodo-L-tyrosinato-bicyclononylboron (**5**) (1.2 g, 86%) as a white powder; mp (browned from 235 to 240 °C) 241 °C dec; IR (thin film) 3420, 2917, 2840, 2530, 2362, 1684, 1419, 1363, 1288, 1213 cm^{-1}; ^1H NMR (400 MHz, CD_3OD) δ 0.23 (s, 1H), 0.54 (s, 1H), 1.79 (m, 12H), 2.95 (dd, J = 14.7, 7.8 Hz, 1H), 3.15 (dd, J = 14.7, 5.0 Hz, 1H), 3.90 (dd, J = 7.8, 5.0 Hz, 1H), 4.62 (s, 1H), 6.80 (d, J = 8.3 Hz, 1H), 7.15 (dd, J = 8.3, 2.0 Hz, 1H), 7.70 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ 23.1, 23.7, 25.1, 25.6, 32.3, 32.5, 32.6, 32.7, 36.7, 37.0, 57.5, 116.0, 129.8, 131.5, 141.0, 157.4, 173.7, 176.4; MS (FAB) m/z 428 [M + H]$^+$, 262, 85; HRMS (FAB) calcd for $C_{17}H_{24}NO_3BI$ [M + H]$^+$ 428.0894, found 428.0883.

3-Trimethylsilylethynyl-L-tyrosinato-bicyclononylboron (6). 3-Iodo-L-tyrosinato-bicyclononylboron (**5**) (210 mg, 490 μmol), $Cl_2(Ph_3P)_2Pd$ (8.4 mg, 12 μmol), and CuI (3.2 mg, 17 μmol) were added to an oven-dried flask containing a magnetic stir bar and fitted with a rubber septum and nitrogen inlet. The solids were dried under high vacuum for 30 min with constant stirring. Freshly distilled anhyd THF (45 mL) and Et_3N (58 mg, 570 μmol, 80 μL) were added via an oven-dried nitrogen-flushed syringe and the resulting yellow solution was stirred for 5 min. Ethynyltrimethylsilane (98 mg, 1.0 mmol, 150 mL) was added via an oven-dried nitrogen-flushed syringe, and the reaction mixture was vigorously stirred for 4 h under ambient conditions. The mixture was filtered and the solids were washed with THF. The filtrate was purified by silica gel chromatography (hexanes: ethyl acetate 50:50 v/v) to yield 3-trimethylsilylethynyl-L-tyrosinato-bicyclononylboron (**6**) (194 mg, 99%); mp 108−122 °C. IR (thin film) 3504, 2843, 2690, 2658, 2148, 1708, 1492 cm^{-1}; ^1H NMR (400 MHz, CD_3OD) δ 0.14 (s, 9H), 0.23 (s, 1H), 0.48 (s, 1H), 1.79 (m, 12H), 2.98 (dd, J = 14.7, 8.0 Hz, 1H), 3.12 (dd, J = 14.7, 4.9 Hz, 1H), 3.86 (dd, J = 8.0, 4.9 Hz, 1H), 5.12 (br s, 1H), 6.33 (br s, 1H), 6.75 (d, J = 8.4 Hz, 1H), 7.8 (dd, J = 8.4, 2.1 Hz, 1H), 7.25 (d, J = 2.1 Hz, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ −1.2, 35.0, 56.3, 97.4, 101.1, 110.6, 115.4, 126.8, 130.8, 134.0, 157.5, 171.5; MS (FAB) m/z 398.3 [M + H]$^+$, 232.2, 203.2.

3-Ethynyl-L-tyrosinato-bicyclononylboron (7). 3-Trimethylsilylethynyl-L-tyrosinato-bicyclononylboron (**6**) (194 mg, 0.52 mmol) was dissolved in DMF (10 mL), treated with KF (28 mg, 0.47 mmol), and stirred at ambient temperature for 2 h. The reaction was quenched with 1 M $KHSO_4$ (2 mL), which led to precipitation of the product (**7**) (120 mg, 78%) as a white solid. ^1H NMR (400 MHz, CD_3OD) δ 0.24 (s, 1H), 0.52 (s, 1H), 1.58 (m, 12H), 2.95 (dd, J = 14.8, 7.6 Hz, 1H), 3.14 (dd, J = 14.8, 5.2 Hz, 1H), 3.91 (dd, J = 7.6, 5.2 Hz, 1H), 5.17 (br s, 1H), 6.43 (br s, 1H), 6.80 (d, J = 8.4 Hz), 7.14 (dd, J = 8.4, 2.0 Hz, 1H), 7.31 (d, J = 2.0 Hz, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ 25.1, 25.6, 31.7, 32.2, 32.5, 32.6, 36.4, 37.0, 57.8, 80.7, 82.9, 111.2, 116.9, 128.3, 132.3, 135.8, 176.6.

3-Ethynyl-L-tyrosine (8). 3-Ethynyl-L-tyrosinato-bicyclononylboron (**7**) (120 mg, 0.40 mmol) was dissolved in a minimum amount of MeOH (100 μL), diluted with $CHCl_3$ (5 mL), and stirred at ambient temperature for 3 h. The precipitated product was filtered, dissolved in 0.1 M aq NH_3, and loaded onto Dowex-1 anion-exchange resin (1 mL). The resin was then washed with water and the product was eluted with 1.0 M AcOH and lyophilized to yield 3-ethynyl-L-tyrosine (**8**) (49 mg, 62%) as a

(19) Walton, E.; Rodin, J. O.; Stammer, C. H. *J. Org. Chem.* **1962**, *27*, 2255−2257.

(20) Boger, D. L.; Yohannes, D. *J. Org. Chem.* **1987**, *52*, 5283−5286.

(21) Ranganathan, S.; Tamilarasu, N. *Tetrahedron Lett.* **1994**, *35*, 447−450.

white solid. IR (thin film) 3362, 2958, 2925, 2150, 1635, 1616 cm^{-1}; ^1H NMR (400 MHz, CD_3OD) δ 2.74 (dd, J = 14.7, 8.2 Hz, 1H), 2.96 (dd, J = 14.7, 4.6 Hz, 1H), 3.44 (s, 1H), 3.57 (dd, J = 8.2, 4.6 Hz, 1H), 6.66 (d, J = 8.4 Hz, 1H), 6.97 (dd, J = 8.4, 2.2 Hz, 1H), 7.10 (d, J = 2.2 Hz, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ 36.9, 57.4, 80.7, 83.1, 111.1, 117.0, 128.0, 132.4, 135.4, 159.1, 173.8; MS (FAB) 206.0 $[M + H]^+$, 189.0, 161.0, 160.0, 133.0, 131.1, 115.1; HRMS (FAB) calcd for $C_{11}H_{11}NO_3$ 206.0817 $[M + H]^+$, found 206.0822.

Acknowledgment. The authors thank Drs. Simon Hinkley and Yiu-Fai Lam for assistance in collecting and interpreting NMR data.

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

JO0207022