1.9–1.4 (m, 8 H, CH_2); ^{13}C NMR δ 120 (t, J = 243 Hz, CF_2H), 43 (t, J = 20 Hz), 26.0, 25.9.

(Difluoromethyl)cyclohexane (5f). Dibromide **2f** was treated with 8 equiv of $NaBH_4$ at 70 °C for 13 h. The volatile material was vaccum transferred at 20–30 Torr to a −80 °C cold trap. The trap contents, which consisted of **5f**, dimethyl sulfide, and a few minor components, were distilled, yielding pure **5f**: bp 117–120 °C (lit.[2c] bp 125 °C): ^1H NMR δ 5.50 (td, J_{HF} = 56 Hz, J_{HH} = 4 Hz, 1 H, CF_2H), 1.85–1.6 (m), 1.4–0.95 (m).

***exo*-2-(Difluoromethyl)bicyclo[2.2.1]heptane (5g).** This compound was particularly difficult to prepare, due to the low reactivity of adduct **2g**. Regardless of the solvent (i.e. DMSO, sulfolane, DMPU) this reaction required at least 10 days at 80–85 °C for the conversion to exceed 70% (the choice of solvent had no effect on the product distribution). The following run illustrates the use of DMPU: A 150-mL tube was charged with $NaBH_4$ (6.05 g, 0.16 mol, 8 equiv), **2g** (6.08 g, 0.02 mol), and DMPU (50 mL). The mixture was stirred at 80–85 °C for 12 days. The usual workup yielded 1.66 g of crude containing about 15% unreacted monobromide **4g**. The pure compound had the following physical properties: bp 64–65 °C (50 Torr); ^1H NMR δ 5.48 (td, J_{HF} = 57.6 Hz, J_{HH} = 6.0 Hz, CF_2H), 2.42–2.18 (m, 2 H, H-1, H-4), 1.96–1.70 (m, 1 H, H-2), 1.64–1.06 (m, 8 H, CH_2); ^{13}C NMR δ 119.2 (t, J = 241 Hz, CF_2H), 46.0 (t, J = 19.5 Hz, C-2), 37.3 (dd, J = 6.6 Hz, J' = 2.9 Hz, C-1), 36.32, 36.25, 31.3 (dd, J = 5.3 Hz, J' = 2.5 Hz, C-3), 29.9, 28.6; IR 1179, 1126, 1067, 1014 cm^{-1}. Anal. Calcd for $C_8H_{12}F_2$: C, 65.73; H, 8.27. Found: C, 65.19; H, 8.41.

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Improved Metalation of 2,4,6-Tribromoanisole: Synthesis of 2-Methoxyresorcinol

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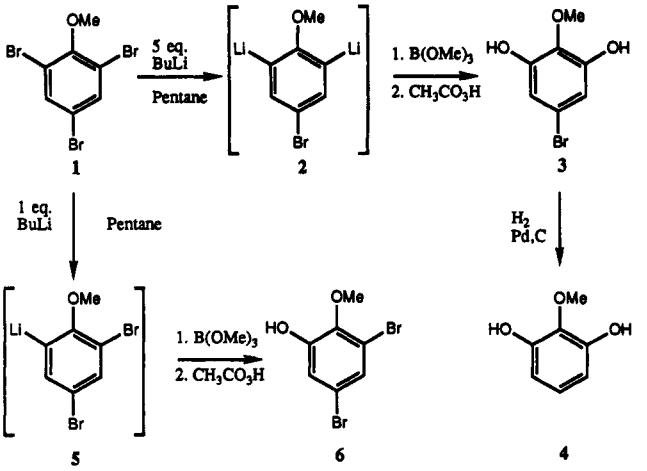
The only reported syntheses of 2-methoxyresorcinol (4) involve methylation of pyrogallol and separation of all possible methylated products, which produces the compound in about 1% yield.[1] We required large quantities of this material and envisioned a process based on bis-ortho metalation of anisole. Direct metalation of anisole requires addition of a chelating agent (e.g. TMEDA) to achieve a reasonable conversion to the anion.[2] For toxicological reasons this approach was not considered feasible. Of the readily available halogenated anisole derivatives, the chloro compounds do not lend themselves to metalation nearly as well as the corresponding bromo analogues; and in addition, in terms of cost and availability, the 2,4,6-tribromo derivative was considered to be the more practical substrate.

The metalation of 2,4,6-tribromoanisole (1) has been reported to occur in pentane at room temperature with *n*-butyllithium. The resultant dianion reacted with carbon dioxide to give 2-methoxy-5-bromo 1,3-dicarboxylic acid in 75% yield.[3] Adaption of this protocol using trimethyl borate[4] in place of carbon dioxide provided only very complex product mixtures. Use of tetrahydrofuran as solvent to solubilize the tribromide at lower temperatures led to no improvement in the complexity of the resultant product mixture. Only after unsuccessfully examining various approaches to effect metalation on *solutions* of the tribromide at low temperature was the metalation attempted on a pentane *suspension* of this compound at −10 to −20 °C.[5] As indicated in Scheme I, this proved successful. Cooling a *room temperature pentane solution* of 1 to −20 °C produces a *suspension*, which, upon addition of *n*-butyllithium, gave an even thicker suspension. Depending on the stoichiometry of the metalation step, the dianion (2) or the monoanion (5) could be quenched with trimethyl borate and the dimethyl borate derivative(s) subsequently oxidized to give 3 or 6 in 91% and 87% yields, respectively. Additionally, 3 was subjected to a standard catalytic hydrogenation to produce 2-methoxyresorcinol 4 quantitatively.

Scheme I



Presumably, during this heterogeneous lithiation procedure, insoluble, unreacted 1 is protected from reactions with the metalated species being generated, thereby reducing side reactions. Reaction of the lithiated intermediates with other electrophiles, especially in a stepwise manner, presents a possible route to highly functionalized anisole derivatives. Moreover, this approach provides ready access to 2-methoxyresorcinol a compound which heretofore was essentially inaccessible.

Experimental Section

5-Bromo-2-methoxybenzene-1,3-diol (3). To a suspension of 1 (10 g, 30 mmol) in 200 mL of dry pentane at −20 °C under Ar was added a solution of *n*-butyllithium in hexanes (93.8 mL of a 1.6 M solution, 150 mmol) over 10 min with vigorous mechanical stirring. This suspension was allowed to warm to −10 °C over 15 min. Upon cooling to −30 °C, neat trimethyl borate (15.6 g, 150 mmol) was added all at once. The solution was warmed to 0 °C over 30 min and then cooled to −10 °C. A solution

(1) Geissman, T. A.; Moje, W. *J. Am. Chem. Soc.* **1951**, *73*, 5765. Spath, E.; Schmidt, H. *Ber.* **1941**, *74*, 193.
(2) Crowther, G. P.; Sundberg, A. M.; Sarpeshkar, A. M. *J. Org. Chem.* **1984**, *49*, 4657.
(3) Gilman, H.; Langham, W.; Moore, F. W. *J. Am. Chem. Soc.* **1940**, *62*, 2327.
(4) For leading references describing the use of this approach to synthesize phenolic compounds, see: Kidwell, R. L.; Murphy, M.; Darling, S. D. *Organic Syntheses*; Wiley: New York, 1973; Collect. Vol. V, p 918. Alabaster, R. J.; Cottrell, I. F.; Marley, H.; Wright, S. H. B. *Synthesis* **1988**, 950.
(5) In addition to using Gilman's exact conditions for this reaction, other room temperature experiments were performed in which the trimethyl borate stoichiometry was varied from 1:1 to 10:1 with respect to both the lithiated anisole and *n*-BuLi. During these experiments, the reaction components were all soluble and in all cases only complex reaction mixtures were obtained.

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of 40% peracetic acid/acetic acid (25 mL) was added over 30 min. Upon completion of the addition, the solution was warmed to 0 °C over 30 min and then cooled to -10 °C, whereupon 25 mL of saturated aqueous $NaHSO_3$ was added dropwise over 30 min. Upon warming to room temperature, equal volumes of water and diethyl ether were added. The organic layer was separated, dried over $MgSO_4$, and evaporated to provide a yellow liquid, which was dissolved in boiling CCl_4 and allowed to cool to room temperature to produce **3** as a white solid (6.0 g, 91%): mp 124-126 °C; ^1H NMR (300 MHz, $CDCl_3$, ppm) 6.68 (s, 2 H), 5.38 (br s, 2 H), 3.86 (s, 3 H); ^{13}C NMR (75.5 MHz, $CDCl_3$, ppm) 149.47, 138.52, 116.89, 111.76, 61.26. Anal. Calcd for $C_7H_7O_3Br$ (219.04): C, 38.39; H, 3.22; Br, 36.48. Found: C, 38.34; H, 3.27; Br, 36.66.

2-Methoxybenzene-1,3-diol (4). To a solution of **3** (5 g, 23 mmol) in ethyl acetate (30 mL) was added 5% Pd/C (500 mg), and this suspension was hydrogenated on a Paar shaker to produce **4** quantitatively (3.2 g) as a white solid after filtration of the suspension and evaporation of the filtrate: mp 85-87 °C (lit.[1] mp 84.5-85 °C); ^1H NMR (300 MHz, $CDCl_3$, ppm) 6.88 (t, 1 H, J = 8.3 Hz), 6.51 (d, 2 H, J = 8.2 Hz), 5.31 (br s, 2 H), 3.88 (s, 3 H); ^{13}C NMR (75.5 MHz, $CDCl_3$, ppm) 148.97, 134.63, 124.80, 108.19, 61.17. Anal. Calcd for $C_7H_8O_3$ (140.14): C, 60.00; H, 5.75. Found: C, 59.76; H, 5.72.

3,5-Dibromo-2-methoxyphenol (6). The procedure described above was followed with the exception that 20 mL (32 mmol) of 1.6 M *n*-butyllithium and 3.2 g (32 mmol) of trimethyl borate were used. The product was obtained as a yellow oil, which was distilled (bp 120 °C (0.24 Torr)) to give 7.4 g (87%) of **6** as a colorless oil which solidified upon standing: mp 67-68 °C; ^1H NMR (300 MHz, $CDCl_3$, ppm) 7.22 (d, 1 H, J = 2.3 Hz), 7.08 (d, 1 H, J = 2.3 Hz), 5.75 (br s, 1 H), 3.90 (s, 3 H); ^{13}C NMR (75.5 MHz, $CDCl_3$, ppm) 150.49, 143.91, 126.94, 118.43, 117.61, 116.33, 61.20. Anal. Calcd for $C_7H_6Br_2O_2$ (281.93): C, 29.82; H, 2.14; Br, 56.68. Found: C, 29.89; H, 2.18; Br, 56.93.

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The Synthesis of Heterobifunctional Linkers for the Conjugation of Ligands to Molecular Probes

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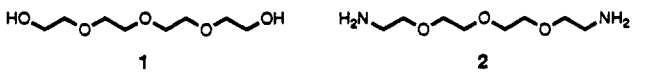
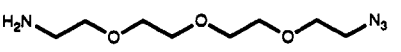
The availability of bifunctional water-soluble compounds with flexible dimensions is important for the conjugation of small molecules to proteins or molecular probes.[3,4] These bifunctional molecules can be used in antibody production, drug delivery, protein immobilization, and for the study of enzymes and receptors.[5-8] Polyethylene glycol (PEG) derivatives are ideal for these purposes because they are inexpensive, water soluble, and available in a variety of lengths. However, currently available symmetrical PEG derivatives such as diol **1** and diamine **2** are difficult to functionalize selectively.[9]



1 **2**

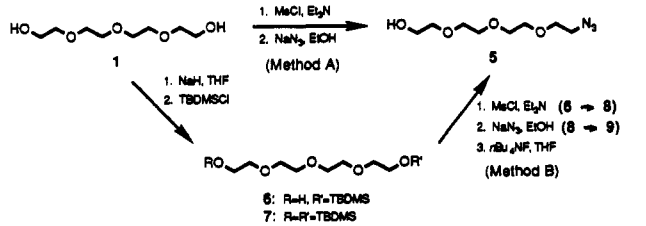
In this paper we describe the synthesis of a heterobifunctional PEG derivative **3**, which contains a free amine



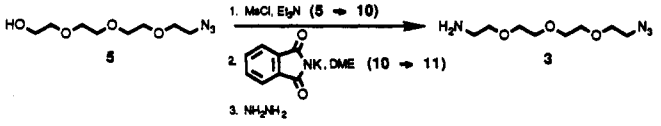
3

that can be conjugated to biological molecules directly by an amide linkage (or via the corresponding isothiocyanate) and an azide that can be reduced to an amine for conjugation to other molecules. The azide reduction can be accomplished by mild, biocompatible reagents such as 1,3-propanedithiol.[10,11] The byproducts of the reduction can be easily removed from the reaction by dialysis or lyophilization and in many cases their presence does not interfere with biological assays. Compound **3** can also be reacted with small organic soluble molecules for the synthesis of heterobifunctional compounds. We have used

Scheme I



Scheme II



(1) Office of Naval Research predoctoral fellow; AT&T Bell Laboratories GRPW awardee.
(2) American Cancer Society Junior Faculty Awardee 1990-1993, Grant No. JFRA-261.
(3) Lee, R. T.; Wong, T.-C.; Lee, R.; Yue, L.; Lee, Y. C. *Biochemistry* **1989**, *28*, 1856.
(4) Fernandez-Santana, V.; Marino-Albernas, J. R.; Verez-Bencomo, V.; Perez-Martinez, C. S. *J. Carbohydr. Chem.* **1989**, *8*, 531.
(5) Antibody production: (a) Magnusson, G.; Ahlfors, S.; Dahmen, J.; Jansson, K.; Nilsson, U.; Noori, G.; Stenvall, K.; Tjornebo, A. *J. Org. Chem.* **1990**, *55*, 3932. (b) Peeters, J. M.; Hazendonk, T. G.; Beuvery, E. C.; Tesser, G. I. *J. Immunol. Methods* **1989**, *120*, 133. (c) Lemieux, R. U.; Bundle, D. R.; Baker, D. A. *J. Am. Chem. Soc.* **1975**, *97*, 4076.
(6) Drug delivery: (a) Park, B. K.; Kitteringham, N. R. *Drug Metab. Rev.* **1990**, *22*, 87. (b) Laguzza, B. C.; Nichols, C. L.; Briggs, S. L.; Cullinan, G. J.; Johnson, D. A.; Starling, J. J.; Baker, A. L.; Bumol, T. F.; Corvalan, J. R. F. *J. Med. Chem.* **1989**, *32*, 548. (c) Neville, D. M., Jr.; Srinivasachar, K.; Stone, R.; Scharff, J. *J. Biol. Chem.* **1989**, *264*, 14653.
(7) Protein immobilization: (a) Bhatia, S. K.; Shriver-Lake, L. C.; Prior, K. J.; Georger, J. H.; Calvert, J. M.; Bredehorst, R.; Ligler, F. S. *Anal. Biochem.* **1989**, *178*, 408. (b) Janda, K. D.; Ashley, J. A.; Jones, T. M.; McLeod, D. A.; Schloeder, D. M.; Weinhouse, M. I. *J. Am. Chem. Soc.* **1990**, *112*, 8886.
(8) Enzyme and receptor studies: (a) Kozikowski, A. P.; Tuckmantel, W. *Tetrahedron Lett.* **1989**, *30*, 4613. (b) Wang, G. T.; Matayoshi, E.; Huffaker, H. J.; Krafft, G. A. *Tetrahedron Lett.* **1990**, *31*, 6493. (c) Bertozzi, C. R.; Bednarski, M. D. *J. Am. Chem. Soc.* Submitted for publication.
(9) Jacobson, A. R.; Makris, A. N.; Sayre, L. M. *J. Org. Chem.* **1987**, *52*, 2592.
(10) Azide reduction with 1,3-propanedithiol can be accomplished in aqueous pyridine or DMF as well as in absolute methanol or ethanol: Bayley, H.; Sandring, D. N.; Knowles, J. R. *Tetrahedron Lett.* **1978**, 3633.
(11) Alternative methods of azide reduction that are compatible with most biological samples include the following. (a) Dithiothreitol (DTT) in physiological buffers: Staros, J. V.; Bayley, H.; Standring, D. N.; Knowles, J. R. *Biochem. Biophys. Res. Commun.* **1978**, *80*, 568. Cartwright, I. L.; Hutchinson, D. W.; Armstrong, V. W. *Nucleic Acids Res.* **1976**, *3*, 2331. (b) H_2S in aqueous pyridine: Adachi, T.; Yamada, Y.; Inoue, I. *Synthesis* **1977**, *45*. (c) Na_2S/triethylamine: Belinka, B. A., Jr.; Hassner, A. *J. Org. Chem.* **1979**, *44*, 4712. (d) Ph_3P/NH_4OH in pyridine: Mungall, W. S.; Greene, G. L.; Heavner, G. A.; Letsinger, R. L. *J. Org. Chem.* **1975**, *40*, 1659.

0022-3263/91/1956-4326$02.50/0