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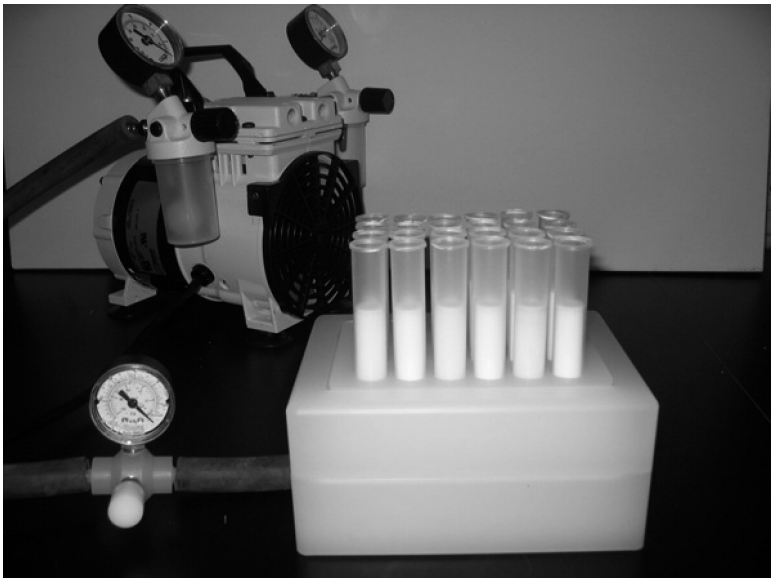

Plate-to-Plate Fluorous Solid-Phase Extraction for Solution-Phase Parallel Synthesis

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A commercially available Argonaut VacMaster-96 plate-to-plate solid-phase extraction (SPE) station equipped with 24 Fluoro*Flash* cartridges is employed for parallel purification of fluorous reaction mixtures. Each cartridge charged with 3 g of fluorous silica gel has the capability to produce up to 100 mg of purified small molecules. The 24-well receiving plate has a standard footprint that can be directly concentrated in a Genevac vacuum centrifuge. Important issues such as sample loading, product cross-contamination, cartridge reuse, and reproducibility are investigated. The SPE system has been demonstrated in the purification of three small libraries that were produced involving amine scavenging reactions with fluorous isatoic anhydride, amide coupling reactions with 2-chloro-4,6-bis[(perfluorohexyl)propyloxy]-1,3,5-triazine (fluorous CDMT), and amide coupling reactions with a newly developed fluorous Mukaiyama condensation reagent.

Introduction

Fluoro*Flash* silica gel has a perfluorooctylethylsilyl (−Si-$(CH_3)_2CH_2CH_2C_8F_{17}$) stationary phase. Solid-phase extraction (SPE) cartridges and HPLC columns packed with fluorous silica gel can be used for separations of highly fluorinated (fluorous) molecules from nonfluorous molecules.[1−3] When compared to liquid−liquid extraction with fluorous solvents, such as FC-72 (perfluorohexanes),[4] fluorous silica gel has the capability to separate compounds with significantly lower fluorine content. A small molecule attached to a C_6F_{17} or C_8F_{17} tag usually has sufficient retention on a Fluoro*Flash* SPE cartridge for convenient separation from nontagged molecules. Since the Curran group introduced fluorous SPE (F-SPE) for "light fluorous" synthesis in 1997,[5] this technique has been widely used in the purification of reaction mixtures containing fluorous catalysts;[6] scavengers;[7] reagents;[8] protecting groups;[9,10] and even biopolymers, such as proteins and DNA fragments.[11] However, F-SPE separations described in the literature were conducted with a single cartridge or on a 2 × 12 SPE vacuum manifold;[12] none of them was in a plate format. Since plate-to-plate SPE for sample preparation is a well-established technology,[13] we decided to incorporate it into fluorous synthesis to improve the efficiency of F-SPE.

Results and Discussion

Evaluation of Plate-To-Plate SPE Separation System. Commercially available plate-to-plate SPE vacuum manifolds from United Chemical Technologies, Inc. (UTC); Supelco; Waters; and Argonaut have similar designs. We selected the Argonaut VacMaster-96 system because it has a modest price and good vacuum seal (Figure 1).[14] The vacuum is applied by connecting the outlet from the receiving plate to a vacuum pump.[15] Whatman receiving plates were used in this project.[16] These plates have a standard footprint and are configured in 24-, 48-, and 96-well formats. We selected the 24-well plate because it has the capability to purify a set of 24 compounds in 10−100-mg quantity, which is suitable for analogue synthesis in common medicinal chemistry programs. This plate system has the following technical features: (1) each 6-mL cartridge can be charged with 3 g of fluorous silica gel,[17] leaving enough space to fill with elution solvents; (2) each well in the receiving plate has a 10-mL volume to collect fractions; and (3) the receiving plate can be directly concentrated in a Genevac vacuum centrifuge.

Figure 1. Plate-to-plate SPE system: (A, left) a 4 × 6 VacMaster system connecting to a Wob-L piston vacuum pump and (B, right) 10-mL receiving well containing blue dye for cross-contamination test.

To evaluate the VacMaster-96 system for F-SPE, we first tested the possibility of cross-contamination that could be caused by a sample splashing from the cartridge tip to the adjacent wells in the receiving plate. Every other cartridge on the plate was charged with a nonfluorous blue dye[18] solution and eluted with 2 × 3 mL of 80:20 MeOH−H_2O. Visual inspection of the receiving plate indicated no cross-contamination (Figure 1B).

We next evaluated the sample loading level by using a mixture generated from an amine scavenging reaction with fluorous isatoic anhydride **3** (Scheme 1).[7d] The mixture contained urea **1**, scavenged amine **2**, and excess scavenger **3** in a molar ratio of 5:1:1. The cartridges were preconditioned with 80:20 MeOH/H_2O (2 × 3 mL). After the sample

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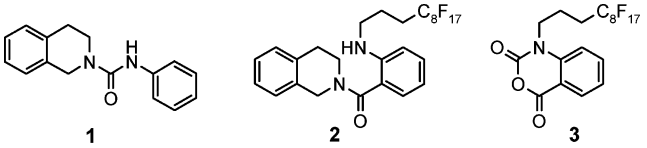
Scheme 1. A Mixture Sample for SPE Test



was loaded, each cartridge was eluted with 80:20 MeOH/H_2O (2 × 3 mL). In the mass loading test, the loading solvent (THF) and volume (0.3 mL) were fixed, and different amounts of the sample were tested at 12, 24, 36, 48, 90, and 150 mg. In all cases, no fluorous sample breakthrough was observed in the 80:20 MeOH−H_2O fractions. This result indicated a 3-g cartridge can be loaded with up to 150 mg (5% mass loading) of a mixture sample.

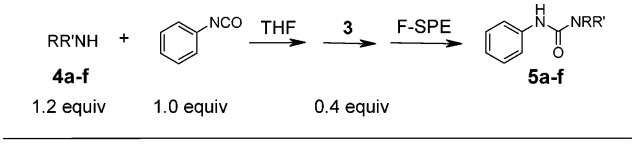
At 1% mass loading, different loading solvents, including dichloromethane (DCM), tetrahydrofuran (THF), and *N*,*N*-dimethylformamide (DMF), were also tested at four different volumes: 0.2, 0.4, 0.6, and 0.8 mL. Small amounts (<5%) of fluorous samples **2** and **3** were leached to the MeOH/H_2O fraction when 0.4 mL of DCM was used as the loading solvent. More fluorous sample (~10%) was leached when 0.4 mL of THF was used as the loading solvent. In contrast, no fluorous sample leaching was observed, even up to 0.8 mL of DMF. These results are consistent with the solvent fluorophilicity order THF > DCM ≫ DMF. F-SPE is a fluorine−fluorine affinity, rather than a mass-controlled separation process. It is sensitive to the volume of the loading solvent, especially with fluorophillic solvents such as THF and DCM. The selection of a loading solvent and use of an appropriate volume is critical to the success of F-SPE. For a 3-g Fluoro*Flash* cartridge and mass loading of <5%, we suggest the following loading solvents and volumes: THF, <0.30 mL; DCM, <0.35 mL; and DMF, <0.8 mL.

Reuse of fluorous silica gel cartridges is important to reduce the cost of consumables and to minimize waste disposal. We prepared six reaction mixtures, **5a**−**f**, from fluorous isatoic anhydride scavenging reactions (Table 1).[7d] Six samples were each divided to 10 portions and subjected to 10 rounds of SPE on different cartridges. In each round, six samples were loaded onto six cartridges and eluted with 80:20 MeOH/H_2O (2 × 3 mL) to collect the product fraction. The plate was concentrated, and each product was analyzed by LC/MS. A new receiving plate was used to collect MeOH (2 × 3 mL) fractions containing fluorous components. The cartridge plate was then washed with acetone (2 × 3 mL) and preconditioned with 80:20 MeOH/H_2O (2 × 3 mL) for the next round of SPE. The results of 10 rounds of SPE using reconditioned cartridges are listed in Table 1. Product purities determined by LC/MS (UV_{254}) were >90%. We noticed compounds **5d**, **5e**, and **5f** had relatively large deviations (~7 mg, 30−35% of the expected yield). That was due in part to the solubility of these mixtures in concentrated loading solvent (THF), which generated a small amount of precipitation in the sample stock solution and, thus, affected the reproducibility of sample loading.

Plate-To-Plate SPE Separation for Parallel Synthesis. After conducting sample loading, cross-contamination, and cartridge reuse studies, we next evaluated the separation of three demonstration libraries. The first library was the preparation of ureas **6** by reaction of an array of four aryl isocyanates with six amines.[7d] Amines were used in slight excess (1.2 equiv). Unreacted amines were scavenged with fluorous isatoic anhydride **3** (0.4 equiv). The 24 reaction mixtures were directly loaded onto a preconditioned SPE cartridge plate. The cartridges were first eluted with 80:20 MeOH/H_2O (2 × 5 mL) to provide the target ureas, followed by MeOH or acetone (2 × 5 mL) to clean the cartridges. The results listed in Table 2 show that product yields are >79% with purities >90%, with the exception of only one product.

In a recent publication, Dembinski and co-workers reported the preparation of 2-chloro-4,6-bis[(perfluorohexyl)methoxy]-1,3,5-triazine (fluorous CDMT) and its use as a coupling reagent in the peptide synthesis.[19] We employed a similar reagent **7** for amide coupling reactions. Four representative

Table 1. SPE with Reconditioned Cartridges[a]



run	5a	5b	5c	5d	5e	5f
1	11mg	11mg	15mg	15mg	14mg	13mg
2	12	11	16	15	16	19
3	11	11	15	14	16	18
4	12	9	16	14	16	19
5	13	13	15	16	17	19
6	12	11	15	15	15	17
7	15	11	15	15	16	18
8	11	10	16	10	16	13
9	12	9	16	16	16	19
10	15	12	18	17	11	20

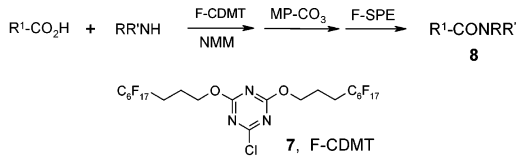
[a] 1% sample loading in THF (0.25 mL); samples were collected in 80:20 MeOH/H_2O (2 × 3 mL) fractions; cartridges were washed with MeOH (2 × 3 mL) and acetone (2 × 3 mL), then reconditioned with 80:20 MeOH/H_2O (2 × 3 mL) for the next runs.

Table 2. Ureas Generated from Amine Scavenging Reactions with Fluorous Isatoic Anhydride **3**[a]

	H_2NBu					
	95(92)*	83(96)	95(93)	86(96)	92(91)	100(96)
Cl	83(100)	81(99)	79(100)	81(100)	83(100)	90(100)
Me	100(80)	100(96)	100(97)	100(94)	100(96)	100(99)
MeO	100(94)	100(99)	100(100)	100(100)	100(100)	100(100)

[a] The asterisk indicates yield % (purity %). Purities were determined by LC/MS with UV_{254} detection.

Table 3. Amide Coupling Products Generated with F-CDMT **7** as a Coupling Agent[a]



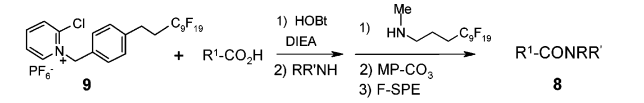
Boc-N(CO2H)	81(100)*	50(83)	57(91)	80(90)	100(60)	67(100)
Ph, CO2H	68(99)	45(88)	53(95)	70(100)	84(95)	62(93)
CO2H, N-Me	36(99)	37(92)	34(100)	67(97)	76(92)	47(95)
CbzN(H), CO2H	58(81)	58(81)	58(92)	45(90)	63(77)	37(99)

[a] The asterisk indicates yield % (purity %). Purities were determined by LC/MS with UV_{254} detection.

carboxylic acids and six amines were selected for 24 parallel reactions. The coupling reactions were conducted in the presence of *N*-methylmorpholine (NMM) using DCM as the reaction solvent. After the reactions were completed, a macroporous polystyrene anion-exchange resin (MP-CO_3)[20] was added to free the NMM salt. Reaction mixtures were filtered, and the filtrates were concentrated to dryness and then loaded to SPE cartridge plate with 0.3 mL of DMF. Following general procedures for plate SPE purification, 24 amides were collected in a single receiving plate for concentration and analysis. Results listed in Table 3 show product yields are in the range of 34−100% and purities are 60−100%.

In another amide coupling reaction, we employed the newly developed pyridinium salt **9** as a fluorous version of the Mukaiyama reagent.[21] The coupling reactions were conducted using the same set of substrates described in Table 3. Mixtures of fluorous pyridinium salt (1.3 equiv), carboxylic acid (1.2 equiv), 1-hydroxybenzotriazole (HOBt, 1.3 equiv), and DIEA (4.0 equiv) were reacted in THF for 10 min to form OBt esters. Amines (1.0 equiv) were added, and the mixtures were stirred for 1 h. *N*-Methyl-[3-(perfluorononyl)propyl]amine (0.4 equiv) in THF was then added to scavenge excess OBt esters. After 12 h, MP-CO_3 (10 equiv) was added, and the mixture was stirred for 3 h. After filtration, the filtrates were concentrated and loaded onto the SPE cartridge plate with 0.3 mL of DCM. Following general procedures for plate SPE purification, 24 amides were collected in a single receiving plate for concentration and analysis. The results listed in Table 4 show that most products have good to excellent yields and >95% purities. Even though the procedures for reactions with F-CDMT **7** are simple because no scavenging step is needed, a direct comparison of the results reported in Tables 3 and 4 clearly demonstrates that coupling reactions with fluorous pyridinium salt **9** are obtained with both higher yields and purities.[22]

Table 4. Amide Coupling Products Generated with **9** as a Coupling Agent[a]



Boc-N(CO2H)	98(99)*	82(97)	89(99)	98(99)	91(97)	93(99)
Ph, CO2H	95(99)	84(98)	96(99)	93(99)	96(98)	99(99)
CO2H, N-Me	40(99)	66(99)	20(98)	95(99)	95(99)	93(99)
CbzN(H), CO2H	88(86)	87(75)	87(75)	80(99)	80(99)	46(99)

[a] The asterisk indicates yield % (purity %). Purities were determined by LC/MS with UV_{210} detection.

In summary, a plate-to-plate SPE system consisting of a VacMaster-96 SPE station, 24-channel plates, and Fluoro*Flash* cartridges has been developed for parallel separation of fluorous reaction mixtures. This system is suitable for purification of 24 reactions producing up to 100 mg of product. Important issues, such as sample loading capability and cartridge reuse, have been investigated. Loading samples to fluorous silica gel packed in deep-well SPE plates instead of parallel-plugged cartridges and conducting SPE with 48- and even 96-well plates are currently under active investigation and will be reported in due course.

Experimental Section

General Methods. All fluorous reagents and cartridges are available from Fluorous Technologies, Inc.[17] Other reagents and solvents were obtained from commercial sources. SPE purifications were conducted on an Argonaut VacMaster SPE manifold with an Isolute Array-24 cartridge plate.[15] Whatman 24-well plates were used as the receiving plate.[17] LC/MS spectra were obtained on an Agilent 1100 system. A Genevac EZ-2 vacuum centrifuge was used for solvent evaporation. The purities of products were determined by LC/MS with a C18 column.

General Procedures for Plate-to-Plate SPE. Fluorous SPE cartridges were washed with acetone or MeOH (3 mL), followed by 80:20 MeOH/H_2O (2 × 3 mL). Sample mixtures in minimum amount of solvent were loaded onto the cartridges with a 6-channel pipet. The cartridges were first eluted with 80:20 MeOH/H_2O (2 × 5 mL) for the desired product, followed by a MeOH or acetone (2 × 5 mL) wash to clean the cartridges. The elution speed was controlled by adjusting the vacuum (5−10 mmHg) to allow ∼1 in./min movement of the solvent. The MeOH/H_2O fraction plates were concentrated in a Genevac EZ-2 vacuum centrifuge. The products were transferred to preweighed 1-dram vials with DCM and concentrated on the Genevac EZ-2 vacuum centrifuge.

General Procedures for Parallel Urea Formation Reactions (Table 2). Four isocyanates, each in THF (0.6 mL),

were distributed into a row of 6 vials (0.1 mmol in 0.1 mL of THF for each vial) sitting in a 24-well plate. Six amines, each in THF (0.4 mL), were distributed into a column of 4 vials (0.12 mmol in 0.1 mL of THF for each vial). The reaction mixtures were shaken at 600 rpm for 1 h, and then fluorous isatoic anhydride **3** (0.04 mmol in 0.1 mL of THF for each vial) was added to each vial. The reaction mixtures were shaken at 600 rpm for 2 h at room temperature. Each mixture in 0.3 mL of THF was warmed to 40 °C and loaded onto SPE cartridges preconditioned with 80:20 MeOH/H_2O (2×3 mL). Standard SPE procedures were performed, and the MeOH/H_2O fraction plate was concentrated in a Genevac EZ-2 vacuum centrifuge. The products were transferred to preweighed 1-dram vials with DCM, concentrated on Genevac EZ-2 vacuum centrifuge, and weighed. All the products were analyzed by LC/MS.

General Procedures for Parallel Amide Coupling Reactions Using F-CDMT 7 (Table 3). Four carboxylic acids, each in DCM (1.8 mL), were distributed into a row of six vials (0.12 mmol in 0.3 mL of DCM for each vial). NMM (0.2 mmol in 0.3 mL of DCM) and fluorous CDMT **7** (0.13 mmol in 0.2 mL of DCM) were added to each vial. The plate of reaction mixtures was shaken at 600 rpm for 20 min. Six amines, each in DCM (0.8 mL), were distributed into a column of four vials (0.1 mmol in 0.2 mL of DCM for each vial). The plate was shaken at 600 rpm overnight before MP-CO_3 (0.6 mmol for each vial) was added. The reaction mixtures were shaken at 600 rpm for 2 h at room temperature and transferred into a 24 well filtration plate. The beads were washed with DCM (3×1 mL), and the filtrates in the receiving plate were concentrated on the Genevac EZ-2 vacuum centrifuge. The residues were dissolved in DMF (0.4 mL) and loaded onto fluorous SPE cartridges, which were preconditioned with 80:20 MeOH/H_2O (2×3 mL). Standard SPE procedures were performed, and the plate that had the MeOH/H_2O fraction was concentrated on the Genevac EZ-2 vacuum centrifuge. The products were transferred to preweighed 1-dram vials with DCM, concentrated on the Genevac EZ-2 vacuum centrifuge, and weighed. All products were analyzed by LC/MS.

General Procedures for Amide Coupling Reactions (0.1-mmol Scale) Using Fluorous Pyridinium Salt (Table 4). To each vial containing a mixture of carboxylic acid (0.72 mmol), HOBt (0.78 mmol), and pyridinium salt **9** (0.78 mmol) was added THF (5 mL) and *N*,*N*-diisopropylethylamine (0.41 mL, 2.3 mmol). The vials were shaken at 600 rpm for 10 min. The total volume of the solution was adjusted to 6.0 mL by adding THF. The solution was split into 6 (1 mL each) portions, and each solution was mixed with a solution of an amine (0.1 mmol) in THF (1 mL). The reactions were monitored by LC/MS. Upon disappearance of the amine, a solution of *N*-methyl-[3-(perfluorononyl)-propyl]amine in THF (0.08 M, 0.5 mL, 0.04 mmol) was added. Upon disappearance of the active ester (HOBt ester) intermediate (LC/MS analysis), MP-carbonate (loading = 3.4 mmol/g, 0.45 g, 1.5 mmol) was added. The mixture was shaken at 600 rpm for 3 h, and the resin was then filtered off and rinsed with THF (3×2 mL). The filtrates were combined and concentrated on a Genevac EZ-2 vacuum centrifuge. Each sample was dissolved in DCM (0.3 mL) (in the case for which product did not fully dissolve in DCM, a small amount of DMF was added to improve solubility). Each solution was loaded onto a SPE cartridge preconditioned with 80:20 MeOH/H_2O (2×3 mL). The standard SPE procedure was performed as described above, and the MeOH/H_2O fraction plates were concentrated on the Genevac EZ-2 centrifuge. Each product was then dissolved, transferred to a preweighed vial, concentrated, and weighed. All of the products were analyzed by LC/MS for purity assessment.

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References and Notes

(1) For general reviews on fluorous silica gel-based separations, see: (a) Curran, D. P. *Synlett* **2001**, 1488. (b) Curran, D. P. In *Handbook of Fluorous Chemistry*; Gladysz, J. A., Curran, D. P., Horvath, I. T., Eds.; Wiley-VCH: Weinheim, 2004, pp 101−127.

(2) For recent reviews on fluorous synthesis, see: (a) Curran, D. P. In *Handbook of Fluorous Chemistry*; Gladysz, J. A., Curran, D. P., Horvath, I. T., Eds.; Wiley-VCH: Weinheim, 2004, pp 128−155. (b) Zhang, W. *Chem. Rev.* **2004**, *104*, 2531. (c) Fache, F. *New J. Chem.* **2004**, *28*, 1277. (d) Zhang, W. *Tetrahedron* **2003**, *59*, 4475. (e) Pozzi, G.; Shepperson, I. *Coord. Chem. Rev.* **2003**, *242*, 115. (f) Dobbs, A. P.; Kimberley, M. R. *J. Fluorine Chem.* **2002**, *118*, 3. (g) Tzschucke, C. C.; Markert, C.; Bannwarth, W.; Roller, S.; Hebel, A. *Angew. Chem., Int. Ed.* **2002**, *41*, 3964.

(3) For fluorous HPLC separations, see: (a) Curran, D. P.; Luo, Z. *J. Am. Chem. Soc.* **1999**, *121*, 9069. (b) Curran, D. P.; Oderaotoshi, Y. *Tetrahedron* **2001**, *57*, 5243. (c) Luo, Z.; Zhang, Q.; Oderaotoshi, Y.; Curran, D. P. *Science* **2001**, *291*, 1766. (d) Zhang, W.; Luo, Z.; Chen, C. H.-T.; Curran, D. P. *J. Am. Chem. Soc.* **2002**, *124*, 10443.

(4) (a) Gladysz, J. A.; Emnet, C. In *Handbook of Fluorous Chemistry*; Gladysz, J. A., Curran, D. P., Horvath, I. T., Eds.; Wiley-VCH: Weinheim, 2004, pp 11−23. (b) Gladysz, J. A. In *Handbook of Fluorous Chemistry*; Gladysz, J. A., Curran, D. P., Horvath, I. T., Eds.; Wiley-VCH: Weinheim, 2004, pp 41−55.

(5) Curran, D. P.; Hadida, S.; He, M. *J. Org. Chem.* **1997**, *62*, 6714.

(6) (a) Matsugi, M.; Curran, D. P. *J. Org. Chem.* **2005**, *70*, 1636. (b) Curran, D. P.; Fischer, K.; Moura-Letts, G. *Synlett* **2004**, 1379.

(7) (a) Zhang, W.; Curran, D. P.; Chen, C. H.-T. *Tetrahedron* **2002**, *58*, 3871. (b) Lindsley, C. W.; Zhao, Z.; Leister, W. H. *Tetrahedron Lett.* **2002**, *43*, 4225. (c) Lindsley, C. W.; Zhao, Z.; Leister, W. H.; Strauss, K. A. *Tetrahedron Lett.* **2002**, *43*, 6319. (d) Zhang, W.; Chen, C. H.-T.; Nagashima, T. *Tetrahedron Lett.* **2003**, *44*, 2065. (e) Werner, S.; Curran, D. P. *Org. Lett.* **2003**, *5*, 3293.

(8) (a) Dobbs, A. P.; McGregor-Johnson, C. *Tetrahedron Lett.* **2002**, *43*, 2807. (b) Dandapani, S.; Curran, D. P. *Tetrahedron* **2002**, *58*, 3855. (c) Lindsley, C. W.; Zhao, Z.; Newton, R. C.; Leister, W. H.; Strauss, K. A. *Tetrahedron Lett.* **2002**, *43*, 4467−4470. (d) Zhang, W.; Chen, C. H.-T.; Lu, Y.; Nagashima, T. *Org. Lett.* **2004**, *6*, 1473−1476.

(9) For a review on fluorous protecting groups and tags, see: (a) Zhang, W. In *Handbook of Fluorous Chemistry*; Gladysz, J. A., Curran, D. P., Horvath, I. T., Eds.; Wiley-VCH: Weinheim, 2004, pp 222−236. (b) Zhang, W. *Curr. Opin. Drug Discovery Dev.* **2004**, *7*, 784.

(10) (a) Luo, Z. Y.; Williams, J.; Read, R. W.; Curran, D. P *J. Org. Chem.* **2001**, *66*, 4261. (b) Curran, D. P.; Amatore, M.; Campbell, M.; Go, E.; Guthrie, D.; Luo, Z. *J. Org. Chem.*

2003, *68*, 4643. (c) Cioffi, C. L.; Berlin, M. L.; Herr, R. J. *Synlett* **2003**, 841. (d) Read, R.; Zhang, C. *Tetrahedron Lett.* **2003**, *44*, 7045. (e) Zhang, W.; Lu, Y. *Org. Lett.* **2003**, *5*, 2555. (f) Zhang, W. *Org. Lett.* **2003**, *5*, 1011. (g) Chen, C. H.-T.; Zhang, W. *Org. Lett.* **2003**, *5*, 1015. (h) Nagashima, T.; Zhang, W. *J. Comb. Chem.* **2004**, *6*, 942. (i) Zhang, W.; Chen, C. H−T.; Lu, Y.; Nagashima, T. *Org. Lett.* **2004**, *6*, 1473. (j) Zhang, W.; Tempest, P. *Tetrahedron Lett.* **2004**, *45*, 6757. (k) Huang, Y.; Qing, F.-L. *Tetrahedron* **2004**, *65*, 8341. (l) Villard, A.-L.; Warrington, B. H.; Ladlow, M. *J. Comb. Chem.* **2004**, *6*, 611. (m) McAllister, L. A.; McCormick, R. A.; Brand, S.; Procter, D. J. *Angew. Chem., Int. Ed.* **2005**, *44*, 452.

(11) (a) de Visser, P. C.; van Helden, M.; Filtppov, D. V.; van der Marel, G. A.; Drijfhout, J. W.; van Boom, J. H.; Noort, D.; Overkleeft, H. S. *Tetrahedron Lett.* **2003**, *44*, 9013. (b) Beller, C.; Bannwarth, W. *Helv. Chim. Acta* **2005**, *88*, 171. (c) Brittain, S. M.; Ficarro, S. B.; Brock, A.; Peters, E. C. *Nat. Biotech.* **2005**, *23*, 463.

(12) A 2 × 12 SPE manifold is available from Supelco, Fisher, and Waters.

(13) For a special issue of SPE for sample preparations, see: Poole, C. F.; Wilson, I. D. *J. Chromatogr., A* **2000**, *885*, 1−471.

(14) Argonaut VacMaster-96 SPE manifold, Product no. 121-9600. Cartridges are placed on Isolute array-24 base plate, Product no. 121-9650 (www.argotech.com).

(15) Wob-L piston pump, model 2545 (www.welchvacuum.com).

(16) Whatman 24-well (10 mL) receiving plate, Product no. 7701-51022 (www.whatman.com).

(17) Fluoro*Flash* silica gel cartridges, 3 g, 6 mL (www.fluorous-.com).

(18) Sudan blue II (17354-14-2) from Aldrich.

(19) Markowicz, M. W.; Dembinski, R. *Synthesis* **2004**, 80.

(20) From Argonaut Technologies, Inc.

(21) Nagashima, T.; Petro, M.; Lu, Y.; Zhang, W. *Tetrahedron Lett.* **2005**, *46*, 6585.

(22) After conducting coupling reactions with F-CDMT, we found literature which indicated that in DCM, the complex of CDMT with NMM could cause NMM demethylation. The relatively low yields and purities listed in Table 3 may relate in part to using DCM as the reaction solvent. For NMM demethylation, see: Kunishima, M.; Kawachi, C.; Iwasaki, F.; Terao, K.; Tani, S. *Tetrahedron Lett.* **1999**, *40*, 5327.

CC050061Z