# ChemInform Abstract: Fluorous Mixture Synthesis (FMS) of Drug-Like Molecules and Enantiomers, Stereoisomers, and Analogues of Natural Products

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# 13

# Fluorous Mixture Synthesis (FMS) of Drug-like Molecules and Enantiomers, Stereoisomers, and Analogues of Natural Products

*Wei Zhang*

## 13.1 Introduction

### 13.1.1 Natural Products and Synthetic Drug-like Compounds

Mother nature is a good resource for new molecules; over 10 000 natural products are isolated each year [1]. Historically, natural products have provided a good number of leads for the development of new drugs [2]. However, since natural products are commonly screened as an extraction mixture, deconvolution of an active component and structural characterization are difficult tasks. In addition, isolation of natural products has a long cycle time and is considered expensive [3]. These limitations have prompted efforts to synthesize natural product analogues and natural-product-like compounds for biological screening and quantitative structure–activity relationship (QSAR) studies.

Besides natural products, synthetic small molecules are another source of drug candidates. Drug-like synthetic molecules usually have privileged structures such as indole, benzopyran, and benzodiazepinone rings [4]. Compared with natural products, drug-like synthetic small compounds tend to have more aromatic rings, planar molecular structures, and few stereogenic centers. They usually have appropriate size and hydrogen-bonding capacity to bind in the active-site pockets of biological targets. Chemical stability and

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physical properties such as solubility and log *P* are always considered in the design of synthetic compounds. The drug-likeness of synthetic compounds can be estimated by the "Lipinski rule-of-five." [5].

Natural product analogues and drug-like compounds are commonly prepared by solution-phase synthesis. Solution-phase chemistry has favorable reaction kinetics and is easy for intermediate analysis. However, compounds are produced "one at a time," followed by a tedious purification process such as chromatography. This is not productive in the preparation of compound libraries for QSAR studies. The advance of solid-phase synthesis and solid-supported solution-phase synthesis has significantly increased the throughput of compound purification processes. Nevertheless, disadvantages such as unfavorable heterogeneous reaction kinetics and difficulty in monitoring the reaction process have limited the capacity of solid-phase synthetic technology to access many natural product analogues and complex drug-like molecules [6]. The recent development of fluorous technologies has provided a solution-phase approach for the synthesis of complicated molecules and their analogues.

### 13.1.2 The Concept of Fluorous Mixture Synthesis

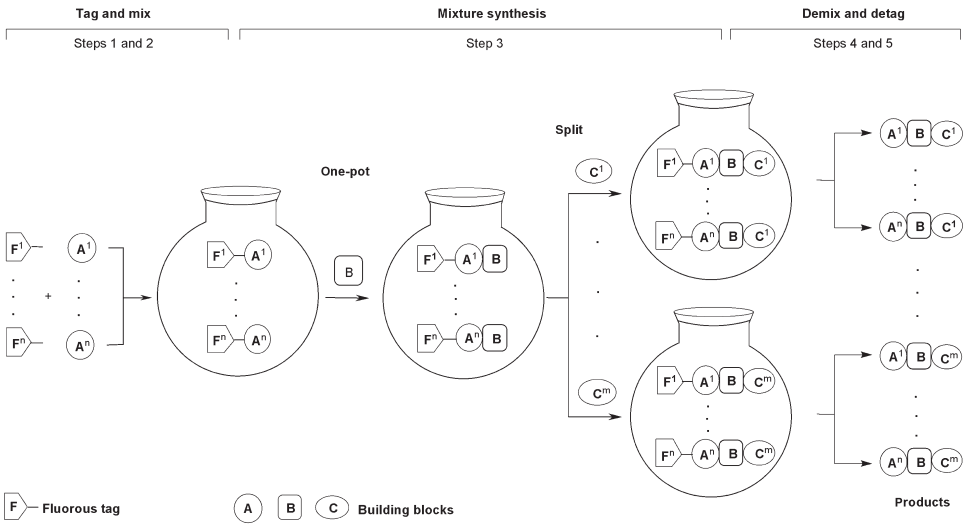
Fluorous chemistry integrates the characteristics of solution-phase reactions and the phase tag strategy developed for solid-phase chemistry [7–15]. Perfluoroalkyl chains instead of polymer beads are used as the phase tags to facilitate the separation process. In 2001 the Curran group first reported the concept of fluorous mixture synthesis (FMS) for solution-phase library synthesis [16]. FMS is able to produce individual pure compounds without the effort of deconvolution. It adapts literature procedures to synthesize complex natural products, their enantiomers and diastereomers. FMS can also be used for the development of new synthetic protocols and to make novel drug-like molecules [17, 18].

Fluorous mixture synthesis consists of the following general steps (see Scheme 13.1).

1. Individually attach a set of substrates to a corresponding set of homologous fluorous tags with increasing fluorine content.
2. Mix the tagged substrates in one pot.
3. Conduct multistep mixture synthesis in one-pot or in split-parallel fashion.
4. Separate the mixtures (demixing) of tagged products by high-performance chromatography (HPLC) on a fluorous stationary phase (F-HPLC).
5. Detag to release final products.

The efficiency of FMS is directly proportional to the number of components mixed (step 2), the length of mixture synthesis (step 3), and the number of splits (step 3).

A fluorous stationary phase such as Fluoro*Flash*®, $Si(Me)_2CH_2CH_2C_8F_{17}$ is hydrophobic and lipophobic, but has strong affinity for fluorous compounds [19]. F-HPLC has the capability to separate mixtures of fluorous compounds according to their fluorine content [20–24]. A molecule with a long perfluoroalkyl ($R^f$) chain gives a longer retention time. A typical mobile phase for fluorous F-HPLC is a gradient of MeOH–$H_2O$ with MeOH increasing up to 100%, which is similar to that in reversed-phase HPLC. Other solvents




***Scheme 13.1*** *Schematic diagram of FMS.*

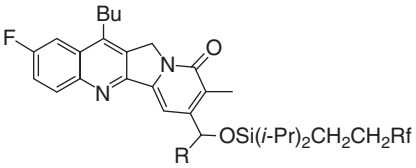
such as MeCN or THF can be used to replace MeOH for gradient elution. An F-HPLC trace shown in Figure 13.1 demonstrates a semipreparative-scale (~5 mg) separation of a mixture of seven fluorous-tagged mappicine analogues bearing different $R^1$ and $R^f$ groups [25].

### 13.1.3 Quasi-racemic FMS

Enantiopure or enantioenriched compounds can be obtained by asymmetric synthesis or by separation of a racemic reaction mixture. Quasi-racemic FMS provides a new approach to enantiomeric compounds (see Scheme 13.2) [26, 27]. Quasi-racemic synthesis starts with two individual *R*- and *S*-enantiomers attached to two different fluorous tags. After steps of mixture synthesis followed by F-HPLC demixing and detagging, two individual products as enantiomers are obtained (see Sections 13.2.1 and 13.2.2). The separation and identification of the final quasi-enantiomers are ensured by the phase-tag-based F-HPLC. In a more complicated quasi-racemic FMS, additional enantiomerically pure building blocks and fluorous tags can be used to generate more chiral centers and more than two products as stereoisomers (see Sections 13.2.3 to 13.2.8).

### 13.1.4 Tags for FMS

Each fluorous tag (also called a protecting group) has two attachment points. One is permanently bound to a perfluoroalkyl ($R^f$) chain and the other is temporarily attached to a reaction substrate so that it can be cleaved to release the product from the support at the




| Peak | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|
| R = | Me, | Pr, | Et, | *s*-Bu, | *i*-Pr, | *c*-$C_6H_{11}$, | $CH_2CH_2$-*c*-$C_6H_{11}$, |
| Rf = | $C_3F_7$, | $C_4F_9$, | $C_6F_{13}$, | $C_7F_{15}$, | $C_8F_{17}$, | $C_9F_{19}$, | $C_{10}F_{21}$, |

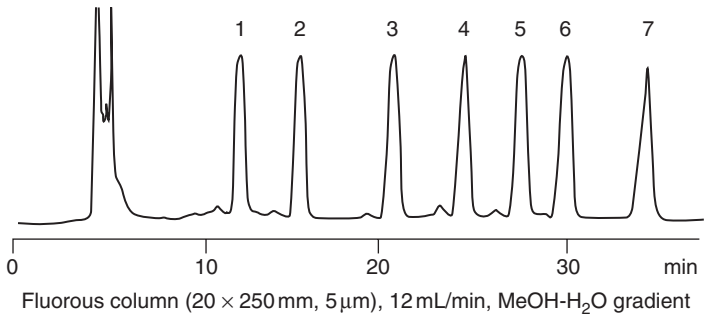
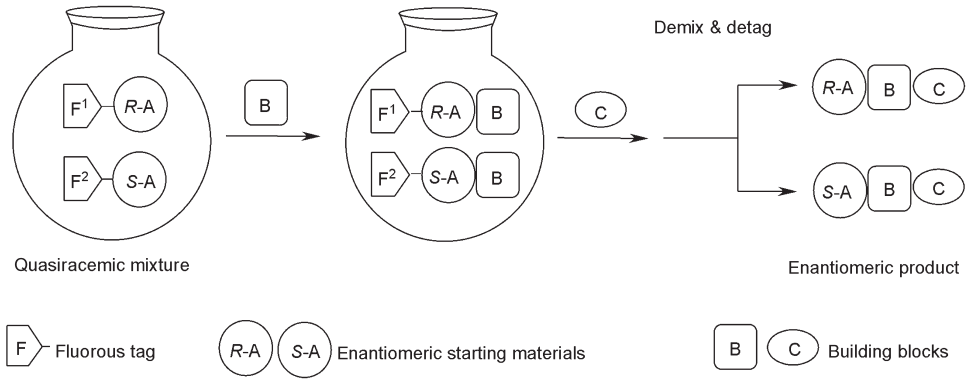
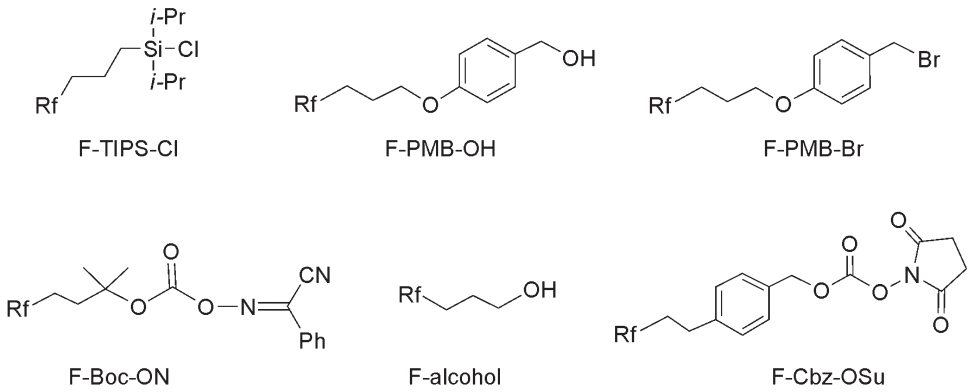



***Figure 13.1*** *Semi-preparative F-HPLC demixing of a seven-component mixture of mappicine analogues.*
*(*Source: *Reproduced with permission from Zhang, W., Luo, Z., Chen, C. H.-T. Curran, D. P., Solution-Phase Preparation of A 560-compound library of individually pure mappicine analogs by fluorous mixture synthesis, J. Am. Chem. Soc. (2002)* ***124****, 10443–10450. Copyright (2002) American Chemical Society.)*




***Scheme 13.2*** *Schematic diagram of quasi-racemic FMS.*

end of the synthesis. An ethylene or propylene spacer is used to minimize the electronic effect generated from the strong electron-withdrawing $R^f$ group, which affects the reactivity of the functional group. Compared with solid-supported linkers, fluorous tags have the following unique features: (i) good solubility in many organic solvents at room or elevated



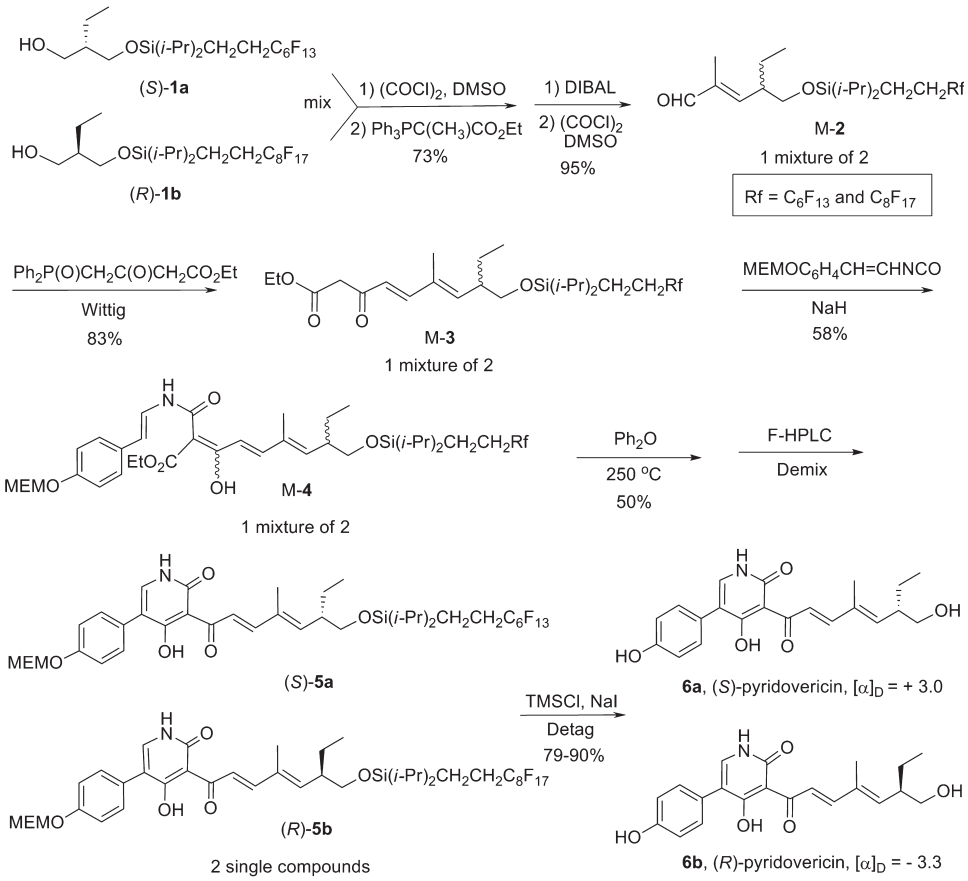

***Scheme 13.3*** *Tags for FMS.*

temperature; (ii) favorable homogeneous solution-phase reaction kinetics; (iii) easy intermediate analysis by conventional tools such as thin-layer chromatography (TLC), nuclear magnetic resonance (NMR), and liquid chromatography–mass spectrometry (LC-MS); and (iv) easy adoption of literature procedures for protecting group attachment and cleavage. Scheme 13.3 lists the fluorous protecting groups that have been developed for FMS [14]. They are the derivatives of common protecting groups such as TIPS, PMB, Boc, and Cbz, which are used to protect hydroxyl, amino, and carboxyl groups. These fluorous tags, with variation of the $R^f$ groups, are commercially available from Fluorous Technologies, Inc. [19].

## 13.2 FMS of Natural Products and Drug-like Compounds and Libraries

### 13.2.1 Synthesis of Enantiomers of Pyridovericin

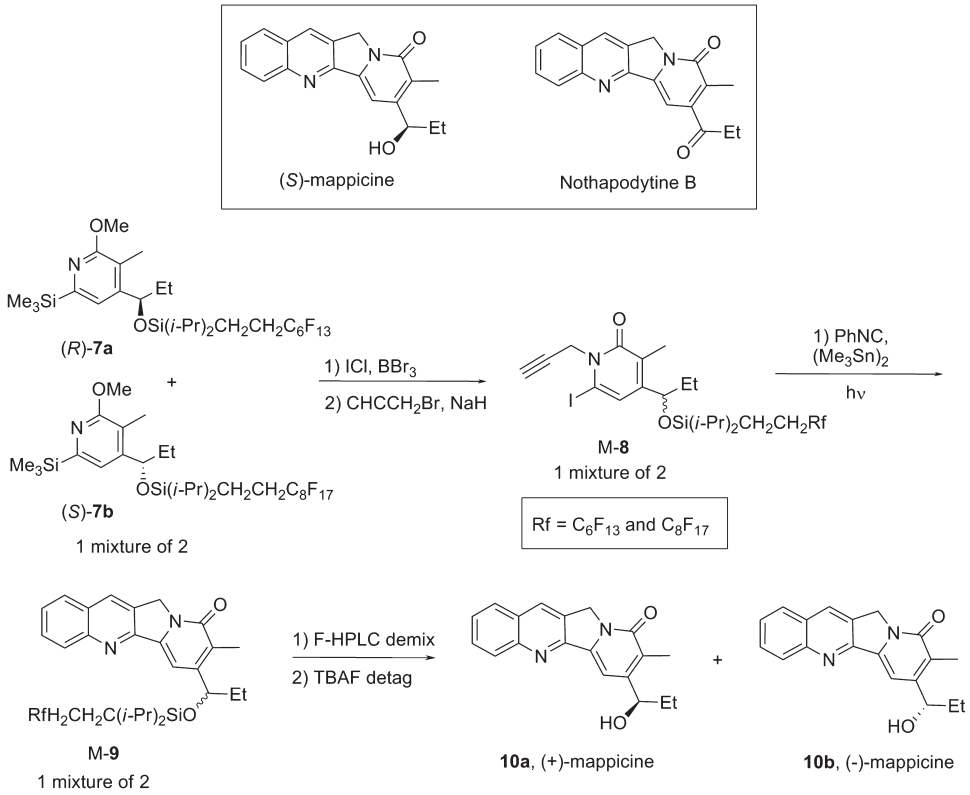
Pyridovericin was isolated from the entomopathogenic fungus *Beauveria bassiana* EPF-5 [28]. It is an inhibitor of the protein tyrosine kinase. The Curran group has demonstrated that enantiomers of pyridovericin can be prepared by quasi-racemic FMS (see Scheme 13.4) [29]. The (*S*)-**1a** and (*R*)-**1b** alcohols as the starting materials were attached to two fluorous silanes with $C_6F_{13}$ and $C_8F_{17}$ groups, respectively. They were then combined to form a quasi-enantiomeric mixture. The mixture was taken through a multistep synthesis through aldehydes M-**2** ("M" stands for mixture), β-ketoesters M-**3**, and then M-**4**. M-**4** was oxidized and then demixed by F-HPLC to give two quasi-enantiomers (*S*)-**5a** and (*R*)-**5b**. The fluorous tags were then removed to release the (*S*)-**6a** and (*R*)-**6b** enantiomers of pyridovericin. Quasi-racemic synthesis is the simplest version of FMS, with only two components in the mixture. Only one-pot, no split-parallel reactions are conducted in this quasi-racemic FMS.




***Scheme 13.4*** *Quasi-racemic FMS of (S)- and (R)-pyridovericins.*

### 13.2.2 Synthesis of Enantiomers of Mappicine

The natural product (*S*)-mappicine was isolated from *Mappia foetida* [30]. Its analogue mappicine ketone, also known as nothapodytine B, was isolated from *nothapodytes foetida* and is active against herpes viruses (HSV) and human cytomegalovirus (HCMV) at a range of 3–13 μM [31]. One-pot total synthesis of both (*R*)- and (*S*)-mappicines has been accomplished by quasi-racemic FMS (see Scheme 13.5) [29]. Enantiomeric (*R*)- and (*S*)-alcohols tagged with silanes containing $C_6F_{13}$ and $C_8F_{17}$, respectively, were converted to quasi-enantiomers (*R*)-**7a** and (*S*)-**7b**. The mixture of these two compounds in 1 : 1 molar ratio was subjected to TMS group exchange with ICl followed by demethylation with $BBr_3$ to form pyridine M-**8**. *N*-Propargylation and subsequent radical cyclization with phenyl isonitrile provided quasi-racemic mixture M-**9**. The separation of this mixture by F-HPLC yielded two quasi-enantiomers. Both (+)-**10a** and (–)-**10b** mappicine were obtained in enantiopure forms after deprotection with TBAF in THF. Similar chemistry for the synthesis of a substituted mappicine library containing 560 analogues is described in Section 13.2.1.



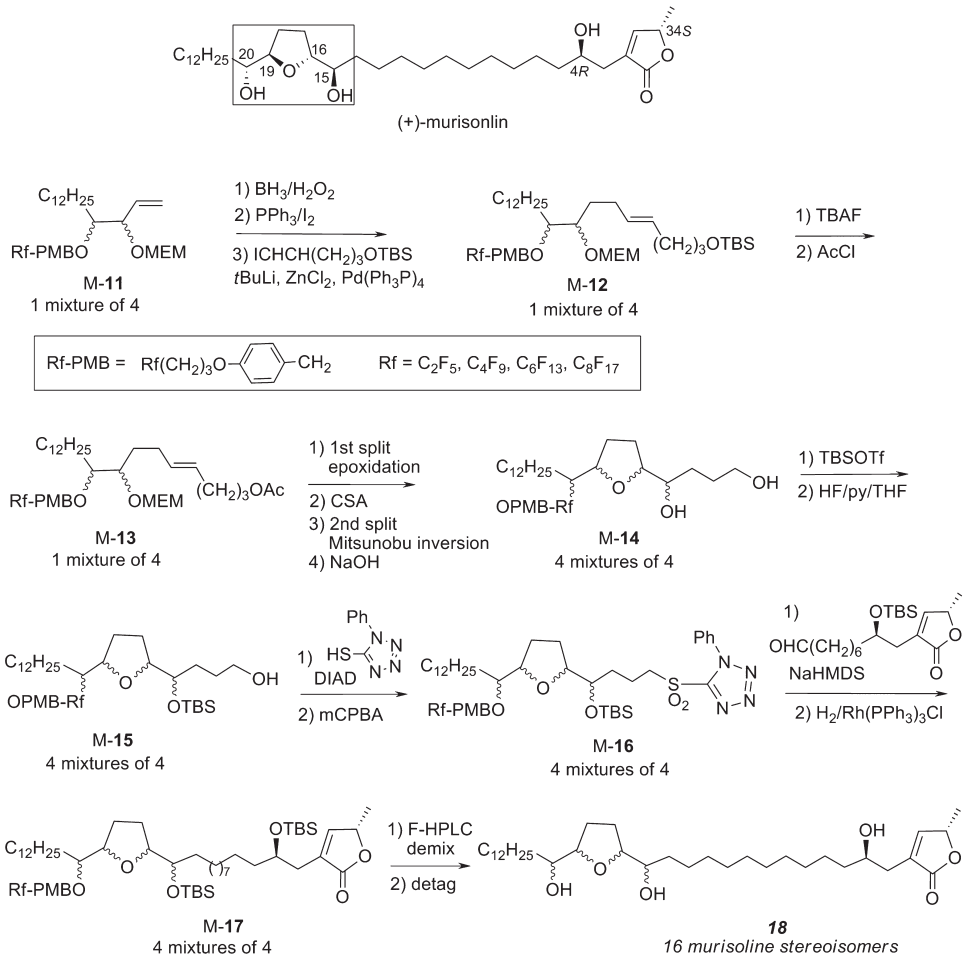

***Scheme 13.5*** *Quasiracemic FMS of (+)- and (–)-mappicines.*

### 13.2.3 Synthesis of Stereoisomers of Murisolin

The murisolin class of mono-tetrahydrofuran acetogenins has six stereocenters. Among the several known murisolin diastereomers, the most active one exhibits extremely high cytotoxicity ($IC_{50}$) at 1 femtomolar (fM) range, whereas the potency of other diastereomers may differ by factors of up to 1 billion [32].

The Curran group reported the FMS of 16 diastereomers of murisolin (see Scheme 13.6) [33]. Sixteen stereoisomers are derivatives of four stereocenters at C-15, C-16, C-19, and C-20 positions of dihydroxytetrahydrofuran fragment (shown in the small box of Scheme 13.6) with the 4(*R*) and 34(*S*) centers fixed. The FMS was started with M-**11,** a mixture of four enantiomerically pure compounds each tagged by a PMB with different $R^f$ groups ($C_2F_5$, $C_4F_9$, $C_6F_{13}$, and $C_8F_{17}$) (see Scheme 13.3). M-**11** was then taken through a sequence of organic reactions to form M-**17**. Two split and parallel syntheses were conducted from M-**13** to M-**14**. Fluorous HPLC demixing of four mixtures of M-**17** followed by detagging provided 16 desired diastereomers of murisolin **18**.

The synthetic scope for making murisolin stereoisomers has been extended through the development of the first double tagging strategy [34, 35] A mixture of four stereoisomers of dihydroxytetrahydofuran encoded with four fluorous tags at C-19 and C-20 was

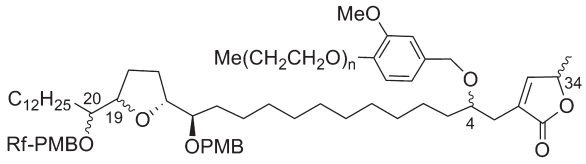



***Scheme 13.6*** *FMS of 16 stereoisomers of murisolin.*

coupled to a mixture of four stereoisomers of hydroxybutenolide fragments encoded with four oligoethylene glycol (OEG, $[OCH_2CH_2O]_n$) tags at C-4 and C-34 (see Scheme 13.7). The mixture containing 16 double-tagged murisolin diastereomers was first demixed by flash chromatography on normal silica gel to give four fractions according to the polarity (length) of the OEG tags. The second demixing with fluorous HPLC gave 16 individual double-tagged compounds. Both fluorous and OEG tags were cleaved by treatment with DDQ. Sixteen murisolin diastereomers were thus produced in a single solution-phase synthesis without splitting.

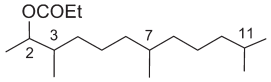
### 13.2.4 Synthesis of Stereoisomers of Pinesaw Fly Sex Pheromone

The propionate ester of 3,7,11-trimethyl-2-tridecanol is a female sex pheromone of the minor sawfly *Microdiprion pallipes* [36]. Pinesaw fly sex pheromones can be used as a

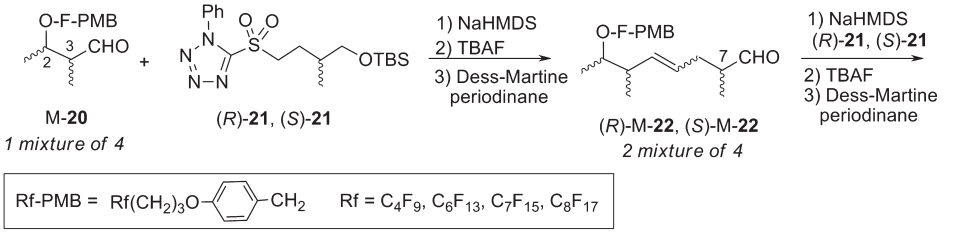



Double-tagged murisoline analogs

***Scheme 13.7*** *Double-tagged murisoline analogues.*




3,7,11-trimethyl-2-tridecanol ethyl ester
(pinesaw fly sex pheromone)




***Scheme 13.8*** *FMS of 16 stereoisomers of pinesaw fly sex pheromone.*

trap for pest control in infested pine tree areas. This molecule has four chiral centers and 16 possible stereoisomers. The Curran group accomplished the synthesis of all 16 stereoisomers of pinesaw fly sex pheromones by a four-component FMS (see Scheme 13.8) [37, 38] A mixture (M-**20**) of four enantiomerically pure aldehydes attached to fluorous PMB with different $R^f$ groups was split into two portions to react with sulfone (*R*)-**21** and (*S*)-**21**. This led to the formation of aldehyde (*R*)-M-**22** and (*S*)-M-**22**, respectively. Each of these two mixtures was subjected to reduction, coupling with (*R*)-**21** and (*S*)-**21**, TBS deprotection, oxidation, and Wittig reaction to give four mixtures of trienes M-**24**. The hydrogenation of alkene followed by F-HPLC demixing of M-**24** afforded 16 individual

F-PMB-attached tridecanols **25**. They were then converted to 16 pinesaw fly sex pheromone diastereomers **26** by parallel tag cleavage and acylation reactions.

### 13.2.5 Synthesis of Stereoisomers of (–)-Dictyostatin

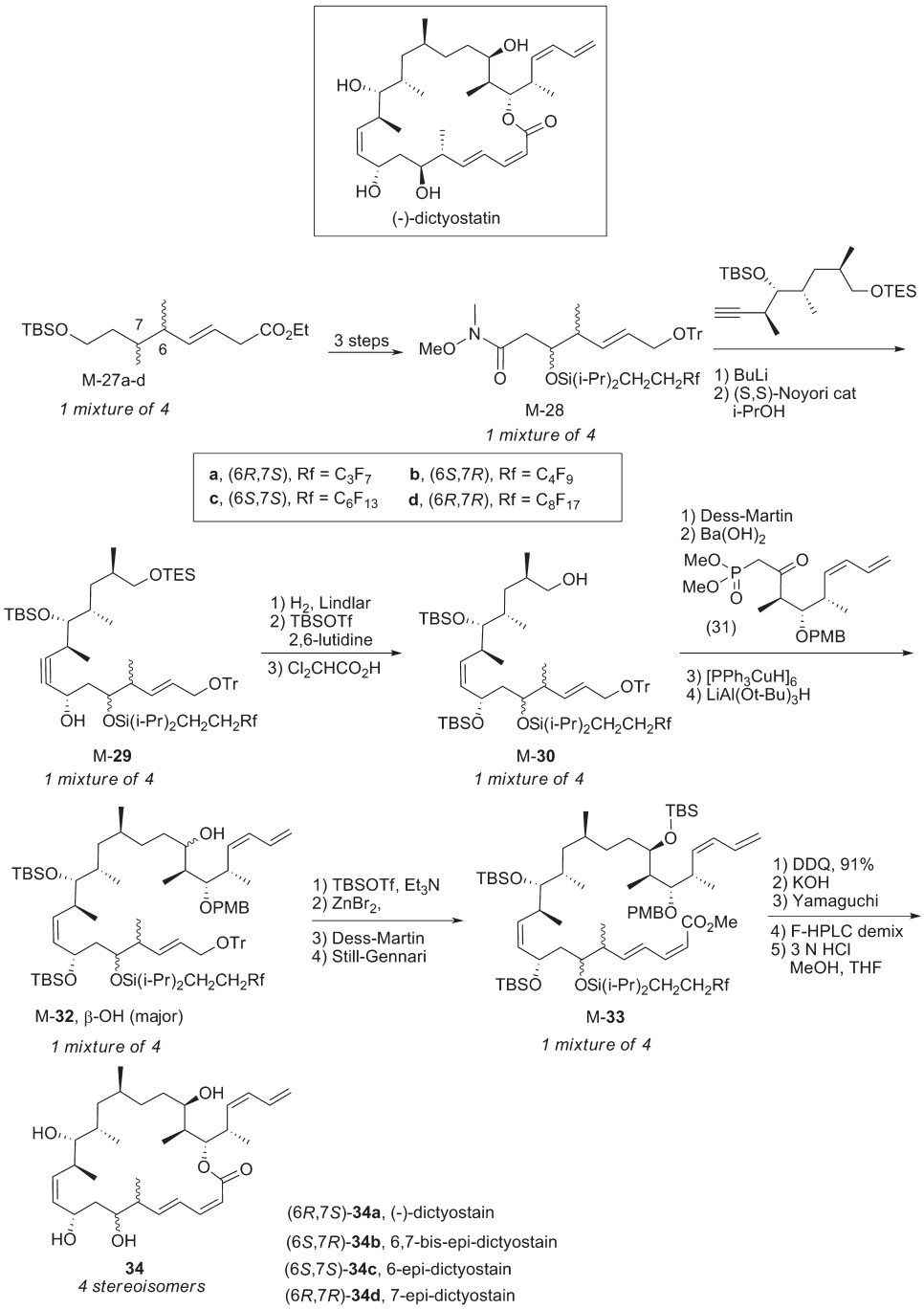
(–)-Dictyostatin is a marine macrolactone that has potent anticancer activity [39]. This compound had been known for over a decade before its stereostructure was confirmed through total synthesis by the Paterson [40] and Curran [41] groups in 2004. Further biological testing on the synthetic sample showed that dictyostatin has equal or better activity against paclitaxel-resistant cell lines than its open-chain analogue discodermolide, radiolabeled paclitaxel, and epothilone B [42]. The Curran group modified their total synthesis route (over 20 steps) for FMS of (–)-dictyostatin and three C-6 and C-7 diastereomers (see Scheme 13.9) [43]. Instead of making these diastereomers in four parallel multistep syntheses, the FMS enables them to be produced in a single set. This is the longest multistep FMS conducted to date.

At the premix stage, a set of four enantiopure alcohols with chiral centers at C-6 and C-7 were individually tagged with a set of four fluorous TIPS-type silanes containing $C_3F_7$, $C_4F_9$, $C_6F_{13}$, and $C_8F_{17}$ tags, respectively. The coded alcohols were then converted to fluorous esters **27a-d**. These four esters were blended in a ratio of 1.5 : 1 : 1 : 1.5 and then the resulting mixture M-**27a-d** was converted to M-**28** in three steps of FMS. M-**28** was coupled with an alkynyllithium and then reduced by (*S,S*)-Noyori's catalyst to give M-**29**. The alkyne group in M-**29** was reduced to the *cis*-alkene by Lindlar hydrogenation and the resulting secondary hydroxy group was protected with the TBS group. The cleavage of TES with dichloroacetic acid gave M-**30**. Dess–Martin oxidation of the primary alcohol followed by coupling with **31** gave the α,β-unsaturated ketone. The reduction of C-17–C-18 alkene with Stryker's reagent followed by reduction of the C-19 ketone with LiAl(O*t*-Bu)$_3$H gave β-alcohol M-**32** as the major product, which was isolated by silica gel chromatography. TBS protection of the C-19 hydroxy group, removal of the trityl group with $ZnBr_2$, oxidation of the allylic alcohol with the Dess–Martin reagent, then the Still–Gennari reaction provided (*E*),(*Z*)-diene M-**33**. The removal of PMB with DDQ, basic hydrolysis of the conjugated ester, followed by macrolactonization under Yamaguchi conditions gave a mixture of major (2*Z*),(4*E*) and minor (2*E*),(4*E*) macrolactones. F-HPLC demixing of the final mixture provided the four individual components. The desilylation with 3 N HCl in MeOH afforded dictyostatin (6*R*,7*S*)-**34a** and the other three C-6,C-7-*epi*-dictyostatin diastereomers after HPLC purification.

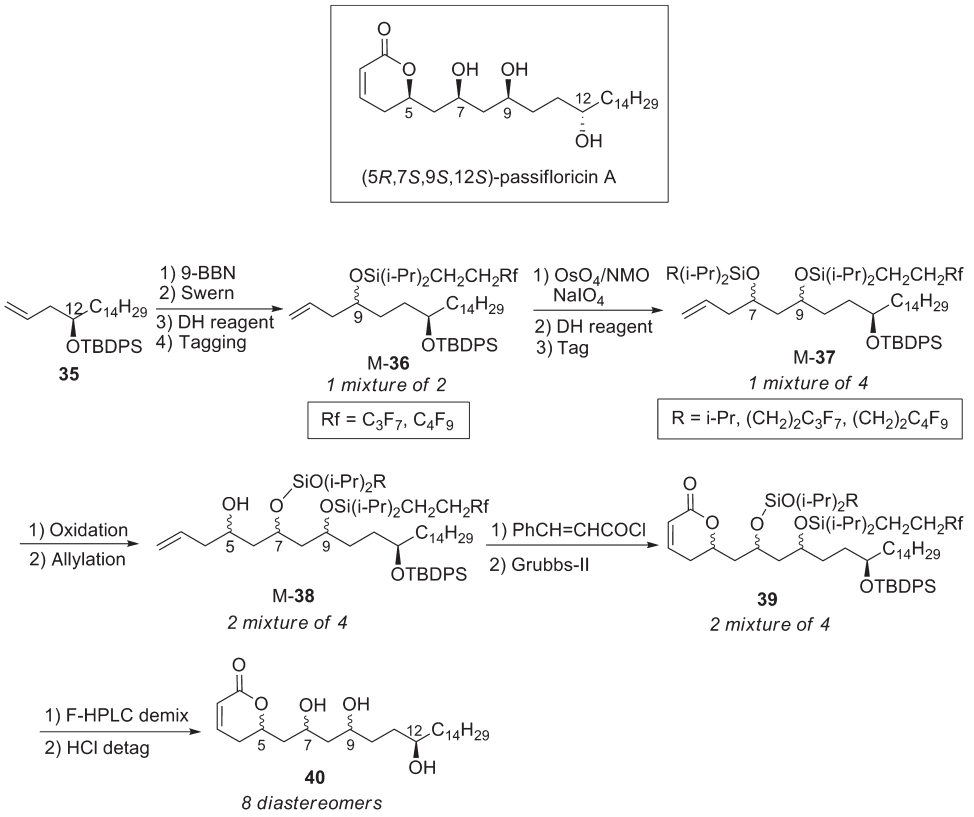
These four compounds were assayed against human ovarian carcinoma cells for their antiproliferative effects [43]. It was found that bis-*epi* diastereomer (6*S*,7*R*)-**34b** was less active than the other isomers, while monoepimer (6*R*,7*R*)-**34d** was equipotent to dictyostatin (6*R*,7*S*)-**34a**, and another monoepimer (6*S*,7*S*)-**34c** was four times more potent.

### 13.2.6 Synthesis of Stereoisomers of Passifloricins

In the total synthesis of an eight-membered stereoisomer library containing the enantiomer of passifloricin A and seven other stereoisomers at C-5, C-7, and C-9, an "en route" protocol was developed by the Curran group to introduce stereocenters and fluorous tags




***Scheme 13.9*** *FMS of (–)-dictyostatin and three stereoisomers.*




***Scheme 13.10*** *FMS of eight stereoisomers of passifloricin.*

during the synthesis [44]. This protocol is different from other FMS in which building blocks with coded stereocenters were premade and pretagged. In addition, the total fluorine content of each tagged molecule is defined by two tags, which allows using two fluorous tags for a four-component FMS. The number of fluorous tags is less than that of components in the mixture.

Enantiopure allyl silyl ether (*R*)-**35** was subjected to a sequence of hydroboration and oxidation reactions (see Scheme 13.10). Half of the resulting aldehyde was treated with the (*R,R*)-Duthaler–Hafner (DH) reagent, and the resulting alcohol was tagged with fluorous triisopropylsilyl trifluoromethanesulfonate (F-TIPSOTf) bearing a $C_4F_9$ group. The other half was treated with the (*S,S*)-DH reagent, and the alcohol was tagged with the F-TIPS group bearing $C_3F_7$. The mixed quasi-racemic M-**36** was split to two portions and subjected to oxidation, allylation, and then tagging reactions. The product from the (*R,R*)-DH reagent got a new nonfluorous TIPS tag, and the product from the (*S,S*)-DH reagent got the repeat $C_3F_7$ tag. A pair of two-compound mixtures was mixed to make a four-compound mixture M-**37**. Repeated split, oxidation, and allylation of M-**37** gave two four-component mixtures M-**38.** The mixtures were acylated, followed by ring-closing metathesis to provide the full stereoisomer library of protected passifloricins as two mixtures of four compounds M-**39**. Each of these two mixtures has four components with

$C_3F_7$, $C_4F_9$, two $C_3F_7$, and two $C_4F_9$ tags. Because the number of fluorine atoms on each component in the mixture is different, they were easily demixed by F-HPLC. All eight isomers of **39** were deprotected individually by exposure to 3 N HCl in EtOH to provide eight compounds **40** including the enantiomer of passifloricin A (*5S*,7*R*,9*R*,12*R*) and its seven diastereomers with R configurations at C-12 and all the possible configurations at C-5, C-7, and C-9.

### 13.2.7 Synthesis of Stereoisomers of Lagunapyrone B

Lagunapyrones A, B, and C were isolated during an investigation of the secondary metabolites of estuarine actinomycetes [45]. These natural products feature a 24-carbon chain consisting of an α-pyrone ring with two adjacent stereocenters (C-6,7) separated by 11 carbon atoms from a second group of three stereocenters (C-19–21). All seven of the double bonds in the backbone of the lagunapyrones are trisubstituted, and lagunapyrones A, B, and C differ only in the nature of the group attached to C-2 (R = Me, Pr, Bu). Lagunapyrone B exhibits moderate activity ($ED_{50}$ = 3.5 μg/mL) against a human colon cancer cell line [45].

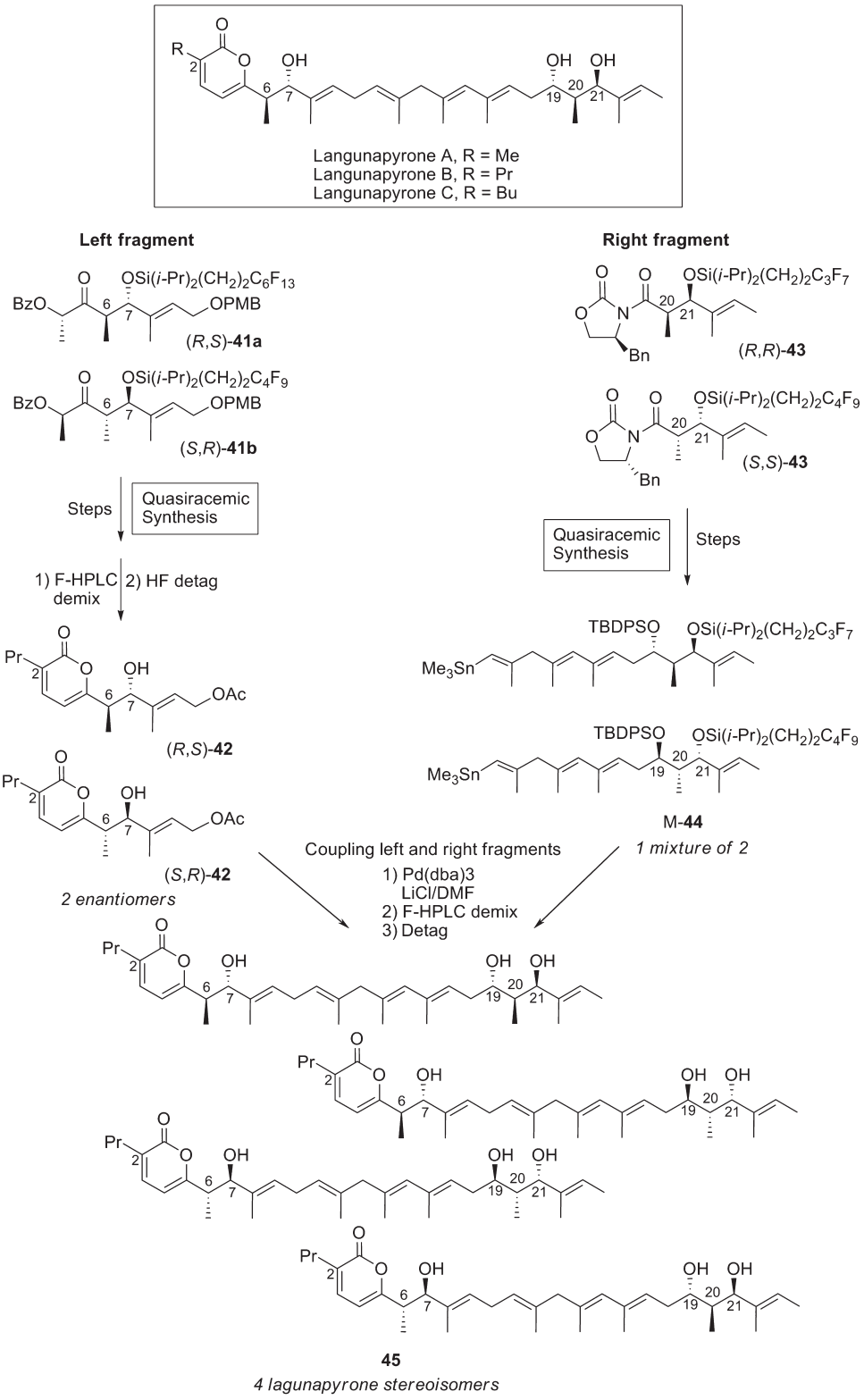
Despite the novel skeleton and interesting biological activity, no synthetic efforts toward the lagunapyrones had been published until the Curran group's work on FMS (see Scheme 13.11) [46]. Quasi-racemic FMS of M-**41** was conducted to construct the left fragment, which has two stereocenters at C-6 and C-7 and an α-pyrone ring. The quasi-racemic product mixture of the left fragment was demixed by F-HPLC and detagged with HF to give (*R,S*)-**42** and (*S,R*)-**42**. The right fragment M-**44** containing C-19–21 stereocenters was also prepared by quasi-racemic FMS. The quasi-racemic mixture of the right fragment M-**44** was reacted with enantiomers (*R,S*)-**42** and (*S,R*)-**42** through the Stille coupling reaction to give two quasi-racemic mixtures of tagged products. These two mixtures were separated by F-HPLC and detagged to afford four lagunapyrone B stereoisomers **45**.

### 13.2.8 Synthesis of Truncated Analogues of (+)-Discodermolide

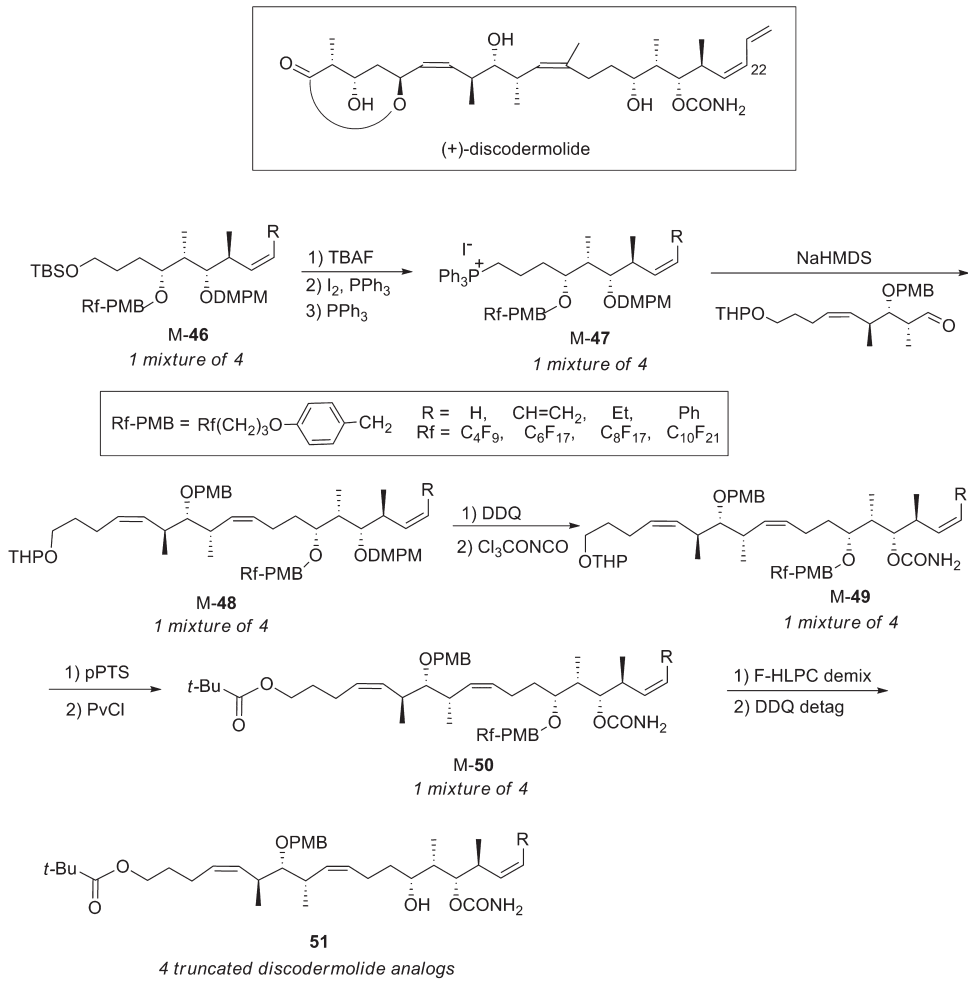
FMS of truncated analogues of the natural product (+)-discodermolide at the C-22 position has been accomplished by the Curran group [47]. Four starting materials with different R group (H, $CH{=}CH_2$, Et, Ph) were protected with the corresponding fluorous PMB ($R^f$ = $C_4F_9$, $C_6F_{13}$, $C_8F_{17}$, $C_{10}F_{21}$) (see Scheme 13.12) and mixed to form M-**46**. 4-Component FMS converted M-**46** sequentially to phosphonium salt M-**47**, the Wittig reaction product M-**48**, carbamate M-**49,** and then tagged product M-**50**. Four truncated discodermolide analogues **51** were produced after demixing and detagging of M-**50**.

### 13.2.9 Synthesis of a Mappicine Library

The power of FMS has been further demonstrated in the preparation of a 560-member library of mappicine analogues (see Scheme 13.13) [25]. In this case, the fluorous tags encoded substrates with different substitutions rather than substrates with different

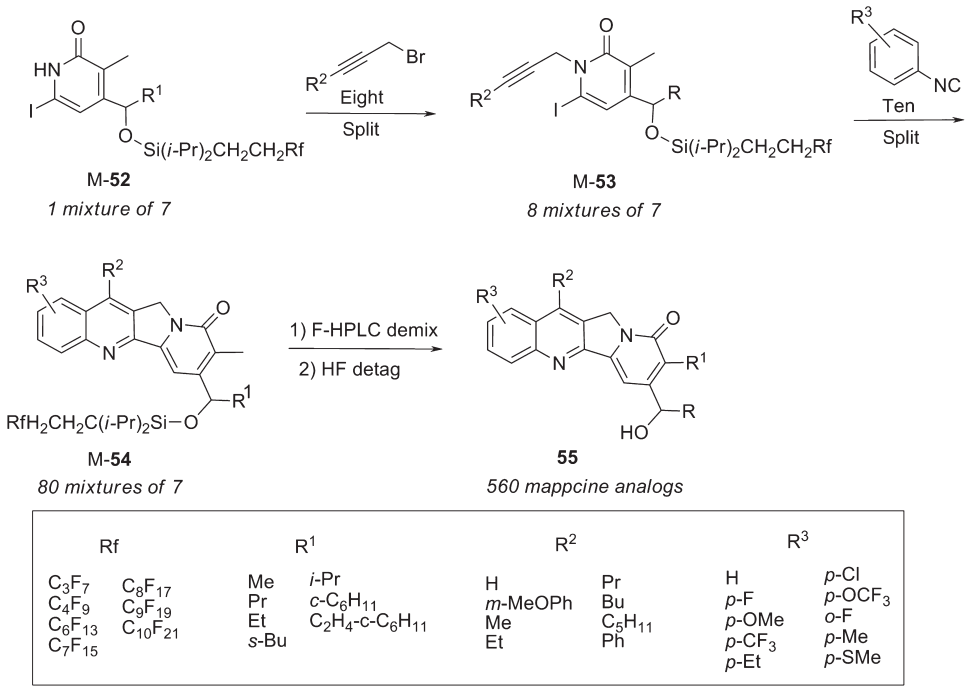



***Scheme 13.11*** *FMS of four stereoisomers of lagunapyrone B.*




***Scheme 13.12*** *FMS of four truncated discodermolide analogues.*

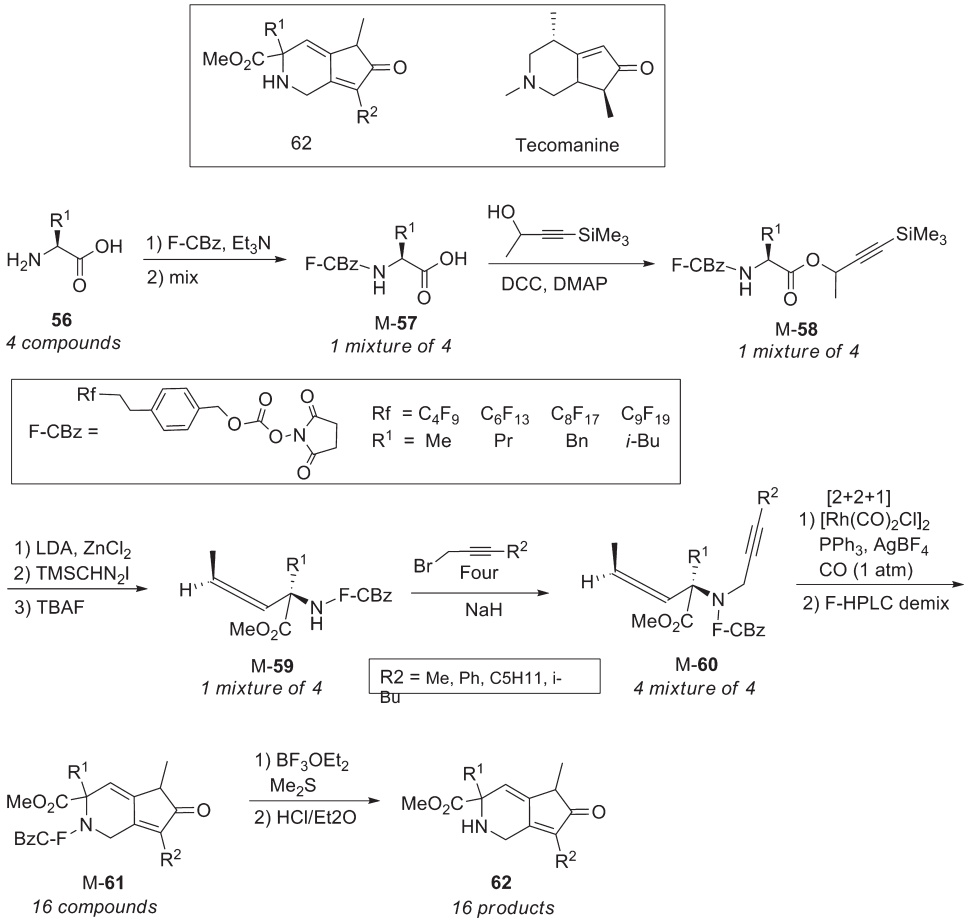
stereochemistry as was described in Section 13.2.2 [29]. A seven-component mixture M-**52** was prepared by iodo-exchange followed by demethylation using the same procedures described in Scheme 13.3. M-**52** was split into 8 portions and subjected to *N*-propargylations with 8 different bromides to give 8 mixtures of M-**53**. Each of the 8 mixtures of M-**53** was further split into 10 portions for radical annulation reaction with isonitriles. The resulting 80 mixtures of M-**54** were demixed by F-HPLC and then detagged by HF-pyridine to give a 560-member mappicine library **55** (see Figure 13.1). After each step, the reaction mixture could be analyzed by fluorous HPLC and the by-products or unreacted starting materials were removed by flash column chromatography with normal-phase silica gel. The synthesis of this 560-membered library is accomplished in 90 reactions (not including the parallel detagging reactions), while 630 steps are needed for a corresponding parallel synthesis. The overall separations required only 90 chromatography steps compared with 630 chromatography steps needed for a parallel synthesis.




***Scheme 13.13*** *Seven-component FMS of a 560-membered mappicine library.*

### 13.2.10 Synthesis of Tecomanine-like Compounds

Tecomanine belongs to a family of natural alkaloids. This compound has shown powerful hypoglycemic activity [48]. The 4-alkylidine cyclopentenone scaffold **62** has a ring skeleton similar to tecomanine. These types of cyclopentenones can be considered as novel bicyclic α-amino acid derivatives that can potentially be useful in the synthesis of peptidomimetics. The Brummond [49] and Curran [50] groups developed a four-component FMS protocol to produce a 16-compound library of the 4-alkylidine cyclopentenone via a rhodium-catalyzed [2 + 2 + 1] allenic Pauson–Khand cycloaddition (see Scheme 13.14). Four F-CBzs containing $C_4F_7$, $C_6F_{13}$, $C_8F_{17}$, and $C_9F_{19}$ groups were individually attached to a set of amino acids **56** with four different $R^1$. The resulting F-CBz-protected amino acids were mixed in equimolar amounts and then reacted in single pot with a propargyl alcohol to form a four-component mixture of esters M-**57**. The allenic amino acid mixture M-**59** was obtained through the Claisen rearrangement of propargyl esters M-**58**. The allene mixture M-**59** was split into four portions and reacted with one of four different propargyl bromides to afford four mixtures of alkynyl allenes M-**60**. The [2 + 2 + 1] cycloaddition of allenes M-**60** followed by F-HPLC demixing afforded 16 individual 4-alkylidene cyclopentenones **61**. The fluorous Cbz tag was removed by treatment with dimethyl sulfide in the presence of $BF_3$-$Et_2O$ to afford 16 final products **62**.

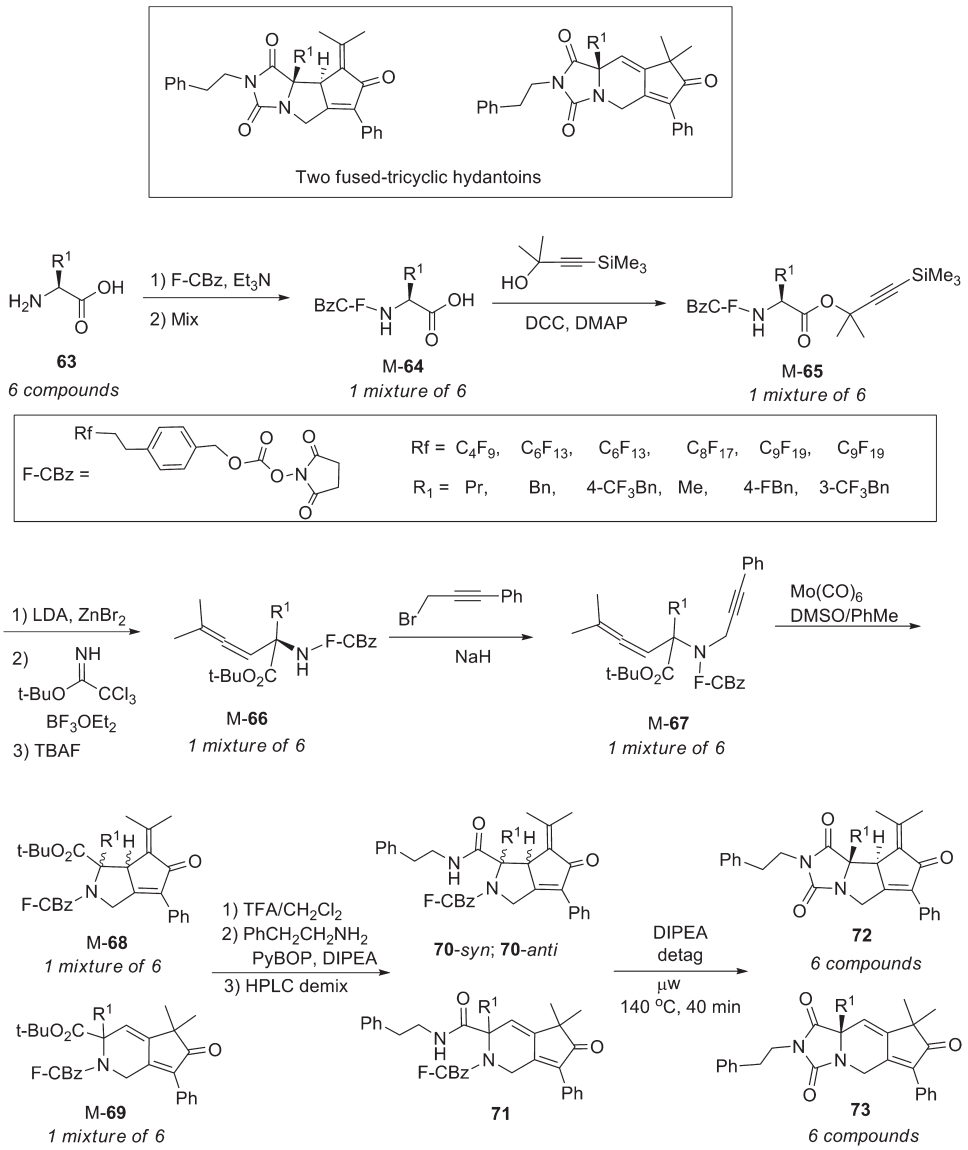



***Scheme 13.14*** *FMS of 16 tecomanine-like compounds.*

### 13.2.11 Synthesis of Fused-Tricyclic Hydantoins

In addition to the "en route" protocol described in Section 13.2.7 for the synthesis of pasifloricins, the use of "redundant tags" for the synthesis of fused-tricyclic hydantoins is another example of FMS in which the number of fluorous tags is less than that of components in the mixture. Since molecules for FMS have fluorine atoms both on the parent structure and on the tag, each component in the mixture has different total fluorine content – even the tag may be redundant.

The six amino acids **63** were prepared individually, and mixed to give acids M-**64** (see Scheme 13.15) [51]. Esterification of M-**64**, zinc-chelated ester enolate Claisen rearrangement of M-**65**, *tert*-butyl esterification, and removal of the $Me_3Si$ group yielded M-**66**. The alkynyl allenes M-**67** were obtained by *N*-propargylation. The allenic Pauson–Khand reaction of M-**67** afforded three products; (*R*)-alkylidenecyclopentenone



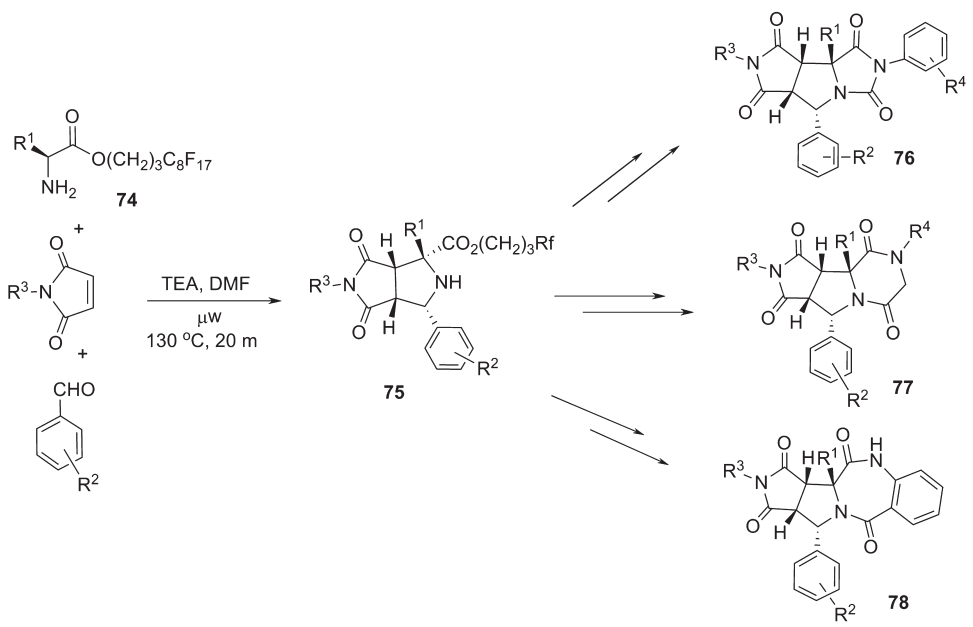

***Scheme 13.15*** *FMS of two fused-tricyclic hydantoin scaffolds.*

stereoisomers M-**68**-*syn* and M-**68**-*anti* along with 4-alkylidenecyclopentanone regioisomer M-**69**. This complex mixture showed only three main spots in a standard silica TLC analysis, and it was purified by flash chromatography over regular silica gel. The single mixture of M-**68**-*syn*, M-**68**-*anti*, and M-**69** was treated with TFA to cleave the *tert*-butyl group, and the resulting acid mixture was coupled with phenethylamine. The mixture was separated by normal silica gel flash chromatography to give three submixtures containing predominantly M-**70**-*syn*, M-**70**-*anti*, and M-**71**. These mixtures were then demixed by

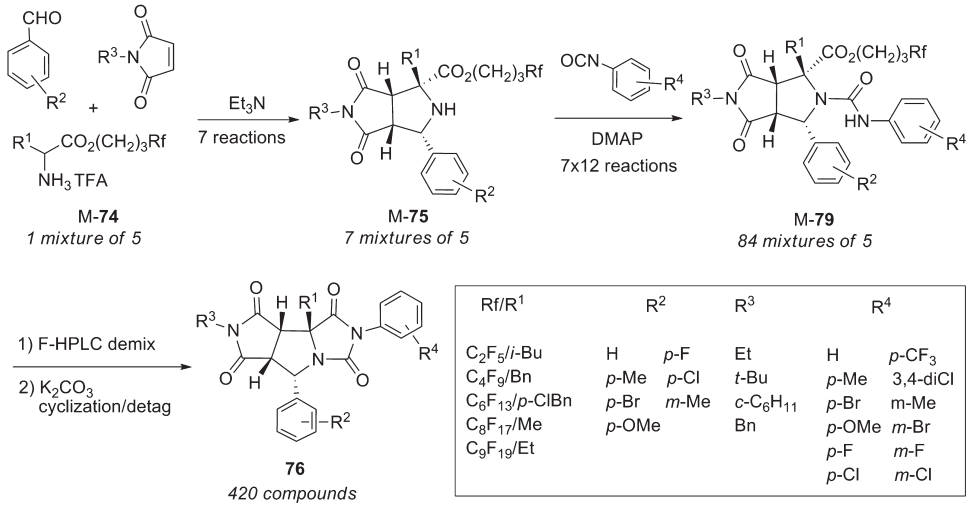
F-HPLC to give 17 crude individual products **70**-*syn/anti* and **71**. These crude products were not isomerically pure. Removal of the fluorous tag and hydantoin formation was achieved by treatment of the individual amides **70**-*syn/anti* and **71** with diisopropylethylamine (DIPEA) under microwave conditions. The cyclative cleavage reactions of **70**-*syn* and **70**-*anti* provided the same products **72**. Normal-phase HPLC purification gave 11 of 12 possible final products **72** and **73**.

### 13.2.12 Synthesis of Novel Heterocyclic Compound Libraries

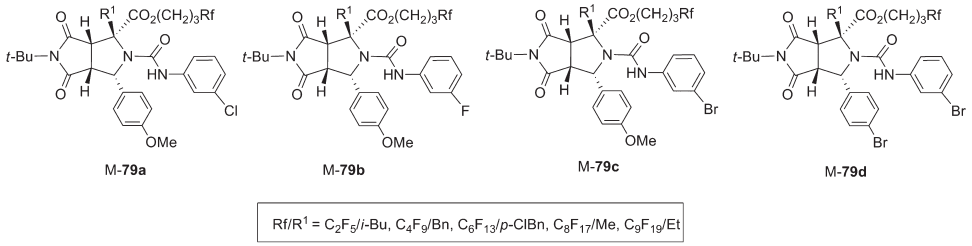
Zhang and co-workers recently developed a [3 + 2] cycloaddition/detag/cyclization protocol for diversity-oriented synthesis (DOS) of hydantoin-, piperazinedione-, and benzodiazepine-fused heterocyclic scaffolds **76, 77**, and **78** (see Scheme 13.16) [52]. Each of these three scaffolds has four stereocenters on the central pyrrolidine ring and up to four points of diversity ($R^1$ to $R^4$). The ring skeleton of these compounds resembles the structures of some known biologically active compounds; the structure of compound **76** is similar to tricyclic thrombin inhibitors [53], the structure of compound **77** is similar to diketopiperazine-based inhibitors of human hormone-sensitive lipase [54], and compound **78** contains a privileged benzodiazepine moiety which exists in numerous pharmaceutically interesting compounds [55]. The key intermediate **75** for DOS was made by one-pot [3 + 2] cycloaddition of fluorous amino ester **74** with slight excess of benzaldehydes and maleimides. The cycloaddition was highly stereoselective. The stereostructure of **75** was confirmed by an x-ray crystal analysis.




***Scheme 13.16*** *DOS of three novel heterocyclic scaffolds.*




***Scheme 13.17*** *Five-component FMS of a 420-member fused-hydantoin library.*




***Scheme 13.18*** *Four FMS samples for parallel demixing.*

A 420-membered hydantoin-fused tricyclic compound library was synthesized by a five-component FMS (see Scheme 13.17) [56]. Five α-amino acids bearing different $R^1$ groups were paired with five perfluoroalkyl alcohols in such combinations as $C_2F_5$/*i*-Bu, $C_4F_9$/Bn, $C_6F_{13}$/*p*-ClBn, $C_8F_{17}$/Me, $C_9F_{19}$/Et. An equimolar mixture of five fluorous amino esters M-**74** was split into seven portions for 1,3-dipolar cycloaddition reaction with one of the seven benzaldehydes and one of the four maleimides. The resulting seven mixtures of M-**75** were each split into 12 portions and reacted with one of the 12 phenylisocyanates to form 84 mixtures of M-**79**. F-HPLC demixing followed by parallel detagging produced a 420-member library of **76**.

The incorporation of four-column parallel F-HPLC coupled with a multichannel MS interface increased the speed both for sample analysis and sample demixing [56]. A 5 min analysis method for baseline separation of five components of M-**79a–d** was applied to four-channel parallel LC-MS analysis (see Scheme 13.18 and Figure 13.2). Four mixture samples containing a total of 20 compounds could be separated in 5 min, which dramatically improves the efficiency of FMS.

The synthesis of a 60-member benzodiazepine-fused tetracyclic compound library **78** has also been accomplished by the Zhang group [56]. A mixture of two fluorous amino

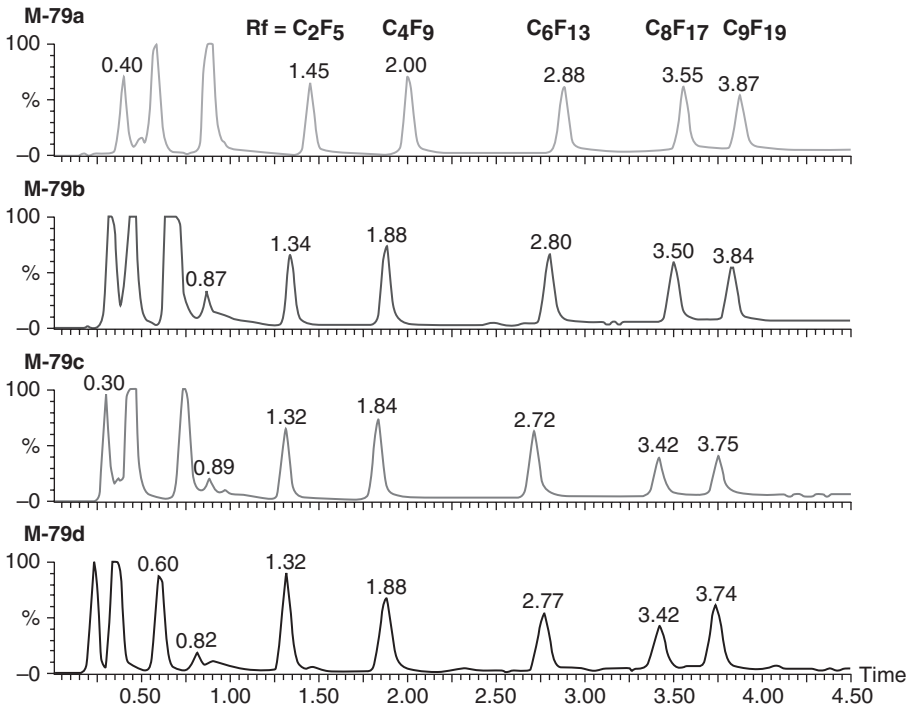
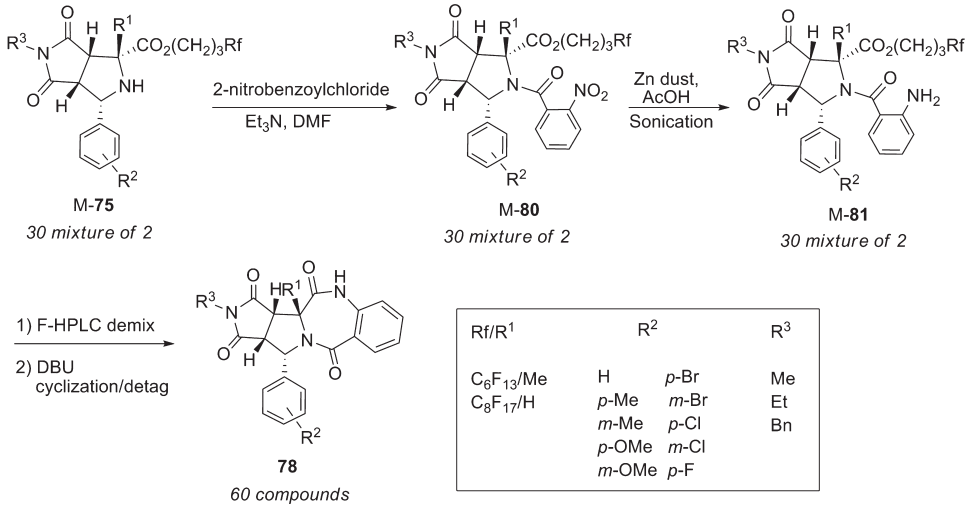



***Figure 13.2*** *Four-column parallel LC-MS of four FMS reaction mixtures (M-**79a–d**). (*Source: *Reproduced with permission from Zhang, W., Lu, Y., Chen, C. H.-T., Zeng, L., Kassel, D. B., Fluorous mixture synthesis of two libraries with hydantoin- and benzodiazepinedione-fused heterocyclic scaffolds,* J. Comb. Chem. *(2006)* **8**, *687–695. Copyright (2006) American Chemical Society.)*

acids ($R^1$ = H and Me) attached to $C_6F_{13}$ and $C_8F_{17}$ was reacted with 10 aldehydes and three maleimides to give 30 mixtures of M-**75** (see Scheme 13.19). *N*-Acylation followed by nitro group reduction with zinc dust in acetic acid under sonication gave 30 mixtures of M-**81**. F-HPLC demixing of M-**81** gave 60 individual compounds. These compounds then underwent cyclative tag cleavage with 1,8-diazabicyclo[4.3.0]non-5-ene (DBU) to form the corresponding benzodiazepine-fused tetracyclic compounds **78**.

## 13.3 Conclusions

FMS is a new and highly efficient solution-phase synthetic technology for making individually pure products. Fluorous tag-based HPLC ensures the identification and separation of reaction mixtures. FMS has advantages of homogeneous reaction environment, easy analysis and purification of reaction intermediates, easy adoption of traditional solution-phase reaction conditions, no requirement to use large excess of reagents for reaction completion, and good chemical and thermal stability of fluorous tags. It has been




***Scheme 13.19*** *Two-component FMS of a 60-member fused-benzodiazapindione library.*

demonstrated in the synthesis of drug-like molecules, complex natural products and their enantiomers, diastereomers, and analogues. As a promising new technology, FMS will play increasingly important roles in organic chemistry, medicinal chemistry, and compound library synthesis.

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