Total syntheses of O^4,O^9-dimethylstealthins A and C [1]

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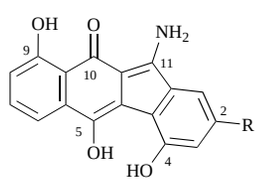
O^{12}-Acetyl-O^4,O^9-dimethylstealthin A (2-acetoxymethyl-11-amino-5-hydroxy-4,9-dimethoxybenzo[*b*]-fluoren-10-one and O^4,O^9-dimethylstealthin C (11-amino-5-hydroxy-4,9-dimethoxy-2-methylbenzo-[*b*]fluoren-10-one), methylated derivatives of potent radical scavengers produced by *Streptomyces viridochromogenes*, have been synthesized using Suzuki coupling as a key step.

Free radicals such as ROO$^{\bullet}$ can react at random by hydrogen abstraction and a variety of addition reactions to damage proteins, lipids and vitamins. In biological tissues, uncontrolled lipid peroxidation causing membrane destruction is increasingly regarded as an important event in the control or development of diseases.[2] Many diseases, such as atherosclerosis, inflammation and Parkinson's disease have been proven to be caused by oxygen-derived free radicals.[3] Thus these diseases may be treatable by prescribing radical scavengers.

Recently, a growing number of radical scavengers have been isolated from microorganisms. In 1992, the Seto group reported the isolation of stealthin A and B **1** and **2** as potent radical scavengers from *Streptomyces viridochromogenes*,[4] and showed that their radical-scavenging activities were 20–30 times as potent as that of vitamin E. The Gould group synthesized stealthin C **3** and demonstrated its existence in kinamycin biosynthesis.[5]



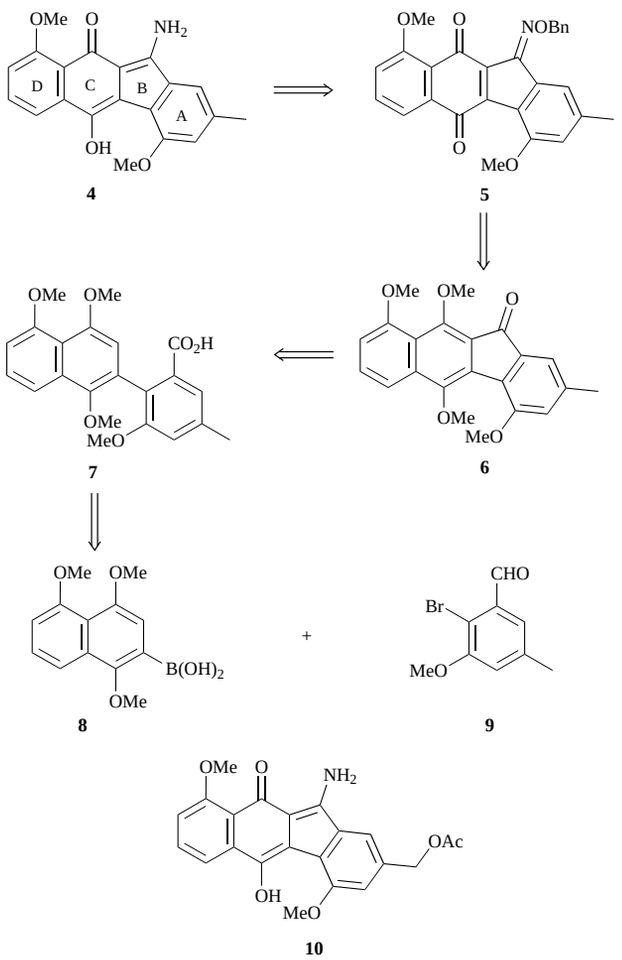
1 R = CH_2OH
2 R = CHO
3 R = CH_3

Structures of stealthins A, B and C

In the present paper, we describe the efficient synthesis of O^4,O^9-dimethylstealthin C and O^4,O^9-dimethylstealthin A. A retrosynthetic analysis for O^4,O^9-dimethylstealthin C **4** is outlined in Scheme 1. Our synthetic plan for constructing the benzo[*b*]fluoren-11-one skeleton **6** was by using Friedel–Crafts cyclization of biaryl carboxylic acid **7**, obtained by the Suzuki coupling reaction of the naphthylboronic acid **8** and aryl bromide **9**. We envisaged that the introduction of the nitrogen function to ketone **6** would be accomplished by the following sequence of reactions: 1, oximation; 2, oxidation of ring C; 3, partial reduction of the oxime. Furthermore, O^4,O^9-dimethylstealthin A acetate **10** would be derived from the fluorenone intermediate **6** by side-chain oxidation.

Results and discussion

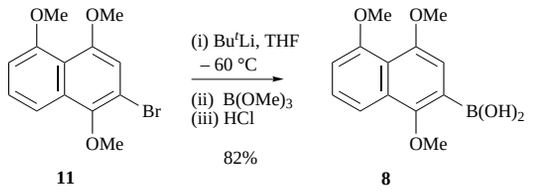
In order to obtain the required biaryl carboxylic acid **7**, we initially used Meyers coupling[6] of the Grignard reagent derived from 2-bromo-1,4,5-trimethoxynaphthalene[7] **11** with 2-(2,3-dimethoxy-5-methylphenyl)-4,4-dimethyl-4,5-dihydrooxazole.[8] However, the yields were poor and the preparation of the oxazole was difficult.



Scheme 1 Retrosynthetic analysis for O^4,O^9-dimethylstealthin C **4**

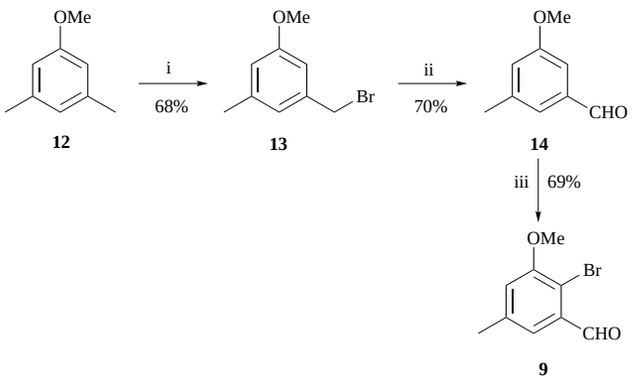
We then explored coupling reactions using a Suzuki protocol.[9] The required naphthylboronic acid **8** was available in a few steps from bromide **11** (Scheme 2). The crucial step for the synthesis of the boronic acid **8** was the halogen–metal exchange. Bromide **11** was treated with 3 mole equivalents of *tert*-butyllithium at −60 °C, and after 6 min the reaction was quenched with trimethyl borate, followed by acid hydrolysis to give the required boron compound **8** in 82% yield.

2-Bromo-3-methoxy-5-methylbenzaldehyde **9**, the partner in the biaryl synthesis, was prepared by the following reaction sequence using *ortho*-directive metallation as a key step (Scheme 3). Monobromination of 3,5-dimethylanisole **12** with *N*-bromosuccinimide (NBS) yielded the benzyl bromide **13** which, when oxidized by the Hass procedure[10] using 2-nitropropane and sodium ethoxide, afforded aldehyde **14** in



11 **8**

Scheme 2



12 **13** **14** **9**

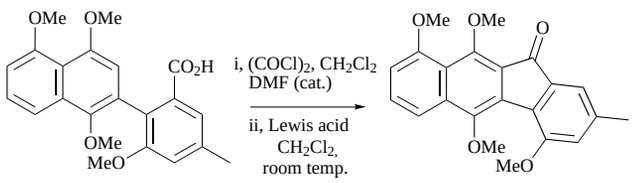
Scheme 3 *Reagents and conditions:* i, NBS, $(PhCO_2)_2$, CCl_4, reflux; ii, Me_2CHNO_2, EtONa, 90 °C; iii, *N*,*N*,*N*′-trimethylethylenediamine, Bu^{n}Li, THF, −65 °C; Bu^{n}Li, −65 to −25 °C; $BrCF_2CF_2Br$, −78 °C

33% overall yield from starting material **12**. Aldehyde **14** was then subjected to *ortho*-directive metallation using the Comins protocol[11] with lithium *N*,*N*,*N*′-trimethylethylenediamide. Treatment of the resulting lithium α-amino alkoxide with 1,2-dibromotetrafluoroethane afforded bromide **9** in 69% yield. When 1,1,2,2-tetrabromoethane was used as a brominating agent, the yield of bromide **9** was only 11%.

With compounds **8** and **9** in hand, attention was then turned to the Suzuki coupling[9] of these components. Thus treatment of substrates **8** and **9** with $Pd(PPh_3)_4$ as the catalyst in freshly distilled 1,2-dimethoxyethane (DME) gave biaryl aldehyde **15** in quantitative yield. Surprisingly, difficulties arose in the oxidation of aldehyde **15** to acid **7**. The conversion could not be realized with silver oxide (starting material was mainly recovered). Attempted oxidation using Jones' reagent gave α,β-unsaturated ketone **16**, which seemed to be formed by oxidation of the naphthalene nucleus followed by aldol condensation of the side-chain aldehyde with acetone. Oxidation with potassium permanganate, pyridinium dichromate (PDC) and sodium hypochlorite each gave an inseparable mixture.

A search of the literature revealed a possible alternative for the oxidation of aldehyde **15**. Kametani and Kano reported[12] conversion of aryl aldehydes into the corresponding acids with 10% hydrogen peroxide and 10% aq. sodium hydroxide at 80 °C. We repeated their procedure and obtained a mixture of the desired acid **7** and the alcohol **17** depending on the conditions used. These products had been formed by way of Cannizzaro disproportionation rather than oxidation if the aldehyde **15** was allowed to come into contact with base before the oxidant was added. Finally, acid **7** was obtained in high yield (91%) by treatment of aldehyde **15** in methanol at 60 °C with simultaneous addition of 15% aq. sodium hydroxide and 35% hydrogen peroxide and heating of the mixture at 90 °C for 1 h (Scheme 4).

We next investigated the synthesis of the required benzo[*b*]-fluoren-11-one **6**. Upon treatment of acid **7** with polyphosphoric acid,[13] no Friedel–Crafts cyclization occurred, but 1,10,12-trimethoxy-8-methyldibenzo[*c*,*h*]chromen-6-one was obtained. Therefore, the Friedel–Crafts cyclization of the corresponding acid chloride was investigated. Treatment of acid **7** with oxalyl dichloride gave 3-methoxy-5-methyl-2-(1,4,5-trimethoxynaphthalen-2-yl)benzoyl chloride. This was then subjected to Friedel–Crafts cyclization with Lewis acids (Scheme 5). Of the Lewis acids tested, titanium tetrachloride



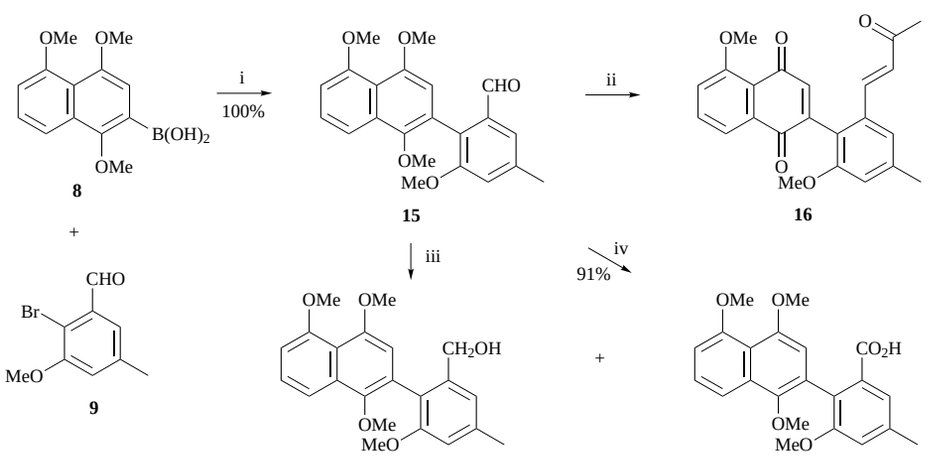
7 **6**

Entry	Lewis acid	Mol equiv.	Yield (%)
1	$AlCl_3$	2.0	no reaction
2	$TiCl_4$	2.0	79
3	$TiCl_4$	1.5	30
4	$SnCl_4$	4.3	23
5	$SnCl_4$	1.2	41

Scheme 5

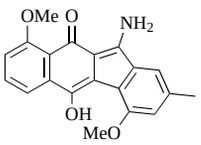
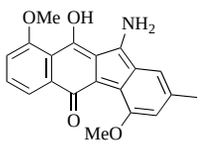
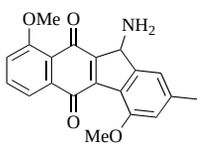
gave a satisfactory result to yield 4,5,9,10-tetramethoxy-2-methylbenzo[*b*]fluoren-11-one **6**. The physical data of product **6** were identical with those previously reported by the Gould group[14] except for the IR band at 1733 cm^{-1}. From the structure of product **6**, this band is not explicable and thus may have been an artifact.

As the attempted demethylation of compound **6** with boron tribromide gave a complex mixture of partially demethylated products, we proceeded to the functionalization of compound **6**



Scheme 4 *Reagents and conditions:* i, $Pd(PPh_3)_4$, DME, 2 M Na_2CO_3; ii, Jones' reagent, Me_2CO, room temp.; iii, 10% H_2O_2, 10% NaOH, MeOH, 80 °C; iv, 35% H_2O_2, 15% NaOH, MeOH, 90 °C

Table 1 Calculated ΔH_f-values for tautomers of O^4,O^9-dimethylstealthin C by the MOPAC[18] method

Tautomers	ΔH_f /kcal mol^{-1}
 **4**	– 75.44
 **26**	– 60.87
 **27**	– 69.36

without complete demethylation followed by reprotection with removable protecting groups. Furthermore, it seemed that the main radical scavenging action of stealthins stems from redox properties of the cross-conjugated 4-amino-3-formyl-1,3-dien-1-ol system in the B and C rings. Thus, the benzo[*b*]fluorenone **6** was subjected to functional-group interconversion without demethylation.

Treatment of compound **6** with *O*-benzylhydroxylamine gave *O*-benzyloxime **18** in 70% yield. Reduction of **18** with zinc in refluxing acetic acid afforded 4,5,9,10-tetramethoxy-2-methyl-11*H*-benzo[*b*]fluoren-11-ylamine. Protection of the amino group of the amine with *N*-(benzyloxycarbonyl)succinimide yielded the corresponding carbamic acid benzyl ester in 86% yield from the oxime **18**. This was then subjected to cerium(IV) ammonium nitrate (CAN) oxidative demethylation; however, the product obtained was not a quinone but the ketone **6**.

We therefore changed the order of the reaction sequence. Oxidative demethylation of compound **18** with CAN and pyridine-2,6-dicarboxylic acid *N*-oxide[15] gave quinone **5** in 77% yield. Reduction of oxime **5** with zinc in refluxing acetic acid gave 11-amino-5-hydroxy-4,9-dimethoxy-2-methylbenzo-[*b*]fluoren-10-one (O^4,O^9-dimethylstealthin C) **4** in 80% yield as a purple solid (Scheme 6).

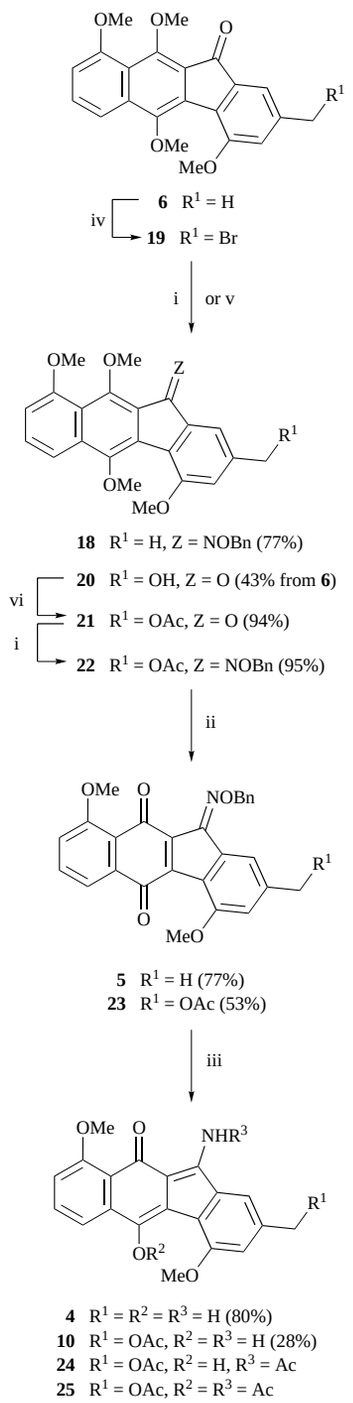
Stealthin C was reported[16] to be 'NMR silent'; however, compound **4** gave clear ^{1}H and ^{13}C NMR spectra. In the ^{1}H NMR spectrum of compound **4**, all signals are well separated and the assignments (see Experimental section) were confirmed by nuclear Overhauser effect (NOE) measurements. Although the ^{1}H and ^{13}C NMR, IR and UV spectra of compound **4** were consistent with the structure proposed, the fast-atom bombardment (FAB) mass spectrum showed the highest-mass peak at *m*/*z* 639 (2M − 31). The reason for this discrepancy remains unknown.[17]

There might be many tautomers of compound **4**. Comparison of the heats of formation (ΔH_f) obtained using the MOPAC method[18] suggested tautomer **4** to be the most stable (Table 1).

The demethylation of compound **4** with boron tribromide in the dark gave a photo- and oxygen-sensitive product, which upon immediate acetylation gave a mixture of partially acetylated products.

Having established the construction of the stealthin C framework, we next carried out the synthesis of the stealthin A analogue. Intermediate **6** was converted into bromide **19** by radical bromination with NBS. Alkaline hydrolysis of bromide **19** with calcium carbonate gave the benzyl alcohol **20** in 43% yield from compound **6**. Acetylation of alcohol **20** under standard conditions afforded acetate **21** (94%), which was then converted into *O*-benzyl oxime **22** in 95% yield as a mixture of *syn* and *anti* isomers. Oxidative demethylation of compound **22** with CAN and pyridine-2,6-dicarboxylic acid *N*-oxide yielded quinone **23** in 53% yield. Reduction of quinone **23** with zinc in refluxing acetic acid afforded 2-acetoxymethyl-11-amino-5-hydroxy-4,9-dimethoxybenzo[*b*]fluoren-10-one (O^{12}-acetyl-O^4,O^9-dimethylstealthin A) **10** in 28% yield as a purple solid (Scheme 6).

The physical data (IR, UV, ^{1}H and ^{13}C NMR) of product **10** supported the structure proposed, but, again, the FAB-MS



Scheme 6 *Reagents and conditions:* i, $BnONH_2$·HCl, AcONa, aq. MeOH; ii, $(NH_4)_2Ce(NO_3)_6$, pyridine-2,6-dicarboxylic acid *N*-oxide, aq. MeCN, 0 °C ~room temp.; iii, Zn, AcOH, reflux; iv, NBS, $(PhCO_2)_2$, CCl_4, reflux; v, $CaCO_3$, 1,4-dioxane–water; vi, Ac_2O, py, DMAP (cat)

spectrum showed discrepancy (*m*/*z* 755, 2M − 31). However, the FAB-MS spectra of 11-acetamido-2-acetoxymethyl-5-hydroxy-4,9-dimethoxybenzo[*b*]fluoren-10-one **24** and 11-acetamido-5-acetoxy-2-acetoxymethyl-4,9-dimethoxybenzo[*b*]-fluoren-10-one **25** obtained by standard acetylation of compound **10** showed normal molecular peaks at *m*/*z* 437 (M + 2) and 479 (M + 2), respectively.

Attempted demethylation of compound **10** with boron tribromide failed due to partial decomposition and to the photolability of the demethylated products.

In summary, we have developed a new method for the construction of the benzo[*b*]fluorenone skeleton and subsequent functional interconversion into stealthins. The key features of the synthetic strategy include (1) Suzuki coupling reaction of the arylboronic acid **8** and the bromide **9**, (2) Friedel–Crafts intramolecular cyclization to the benzo[*b*]fluoren-11-one **6**, and (3) functional interconversion of compound **6** into O^4,O^9-dimethylstealthins by the reaction sequence of oximation, oxidative demethylation, and reduction. This present work offers new, rather stable and useful compounds for studying radical-scavenging actions.

Experimental

All non-aqueous reactions were carried out under argon in oven-dried glassware unless otherwise indicated. Tetrahydrofuran (THF), diethyl ether and DME were distilled from benzophenone ketyl prior to use. Acetonitrile, dichloromethane, dimethylformamide (DMF) and pyridine were dried over CaH_2 under nitrogen for 15 h, after which they were decanted and distilled.

Column chromatography was carried out using Merck Kieselgel 60 (230–400 mesh). TLC was carried out on commercially available pre-coated plates (Merck silica Kieselgel $60F_{254}$). ^1H and ^{13}C NMR spectra were recorded on a JEOL GSX270 (270 MHz) FT spectrometer or Hitachi R-600L (60 MHz) spectrometer using $SiMe_4$ as an internal standard. *J*-Values are given in Hz. Mass spectra were recorded on a JEOL JMS-HX100 spectrometer using a JMA-DA5000 data system for high-resolution analysis. IR spectra were recorded on a Shimadzu FTIR-8100 spectrometer. UV–visible spectra were recorded on a UV-2200 spectrophotometer.

Mps were determined with a Yanagimoto micro apparatus and are uncorrected. The purities of new compounds were checked by high-performance liquid chromatography (HPLC) using a Phenomenex PRODIGY 5ODS-2 (250 × 4.60 mm) column under the conditions described below.

1,4,5-Trimethoxynaphthalen-2-ylboronic acid 8

tert-Butyllithium (0.73 cm³ of 1.7 M solution in hexane; 1.27 mmol) was added dropwise to a stirred solution of 2-bromo-1,4,5-trimethoxynaphthalene **11** (0.307 g, 1.03 mmol) in THF (6.0 cm³) under argon at −60 °C. The resulting dark brown suspension was stirred at −60 °C for 10 min and then trimethyl borate (0.67 cm³, 6.13 mmol) was added dropwise. After 10 min the yellow mixture was allowed to warm to room temp. and was stirred for a further 1 h. The mixture was acidified with dil. hydrochloric acid and then extracted with CH_2Cl_2. The combined extracts were dried ($MgSO_4$), and evaporated under reduced pressure to leave a pale brown oil, which was purified by column chromatography on silica with $CHCl_3$–hexane (1 : 1) as eluent to give title compound **8** (0.222 g, 82%) as a brown solid, mp 101 °C; ν_{max}(KBr)/cm^{-1} 3387, 2943, 1596, 1463, 1380, 1350, 1261, 1078 and 785; δ_H(270 MHz; $CDCl_3$) 3.95 (3 H, s, CH_3O), 3.99 (3 H, s, CH_3O), 4.00 (3 H, s, CH_3O), 6.51 (2 H, br, 2 × OH), 6.95 (1 H, dd, *J* 8.1 and 1.4, 6-H), 7.17 (1 H, s, 3-H), 7.45 (1 H, t, *J* 8.1 and 1.4, 7-H) and 7.67 (1 H, dd, *J* 8.1 and 1.4, 8-H).

3-Bromomethyl-5-methylanisole 13

A mixture of 3,5-dimethylanisole **12** (4.2 cm³, 0.032 mol), NBS (6.25 g, 0.035 mol) and a catalytic amount of benzoyl peroxide in tetrachloromethane (125 cm³) was heated under reflux for 4 h. The solvent was removed under reduced pressure and the residue was extracted with chloroform. The extract was washed with water, dried ($MgSO_4$) and evaporated, leaving a yellow oil **13** (8.46 g). The crude product was used for the next step without further purification.

3-Methoxy-5-methylbenzaldehyde 14

To a solution of sodium ethoxide [prepared from ethanol (220 cm³) and sodium (5.05 g, 0.216 mol)] was added 2-nitropropane (14.7 cm³, 0.194 mol) followed by a solution of bromide **13** (25.5 g, 0.118 mol) and ethanol (10 cm³). The reaction mixture was heated for 4 h at 90 °C. After the solvent had been removed under reduced pressure, the residue was extracted with chloroform (3 × 50 cm³). The combined extracts were dried ($MgSO_4$) and evaporated to leave a dark brown oil, which was purified by distillation through a Vigreux column under reduced pressure (bp 57 °C/0.23 mmHg) to give title aldehyde **14** as a pale yellow oil (8.81 g, 33% from **12**); ν_{max}(film)/cm^{-1} 2842, 2731, 1701, 1606, 1596, 1544, 1469, 1386, 1332, 1298, 1157, 1066, 848, 707 and 680; δ_H(60 MHz; $CDCl_3$) 2.39 (3 H, s, CH_3), 3.84 (3 H, s, OCH_3), 7.00 (1 H, s, 4-H), 7.19 (1 H, s, 2-H), 7.23 (1 H, s, 6-H) and 9.93 (1 H, s, CHO).

2-Bromo-3-methoxy-5-methylbenzaldehyde 9

To a solution of 3.6 cm³ (28.3 mmol) of *N*,*N*,*N*′-trimethylethylenediamine in 2 cm³ of THF at −65 °C were added 29.3 mmol of 1.56 M Bu^{n}Li in hexane solution dropwise. After 30 min, a solution of aldehyde **14** (4.02 g, 26.8 mmol) in THF (25 cm³) was added, the mixture was stirred for 30 min, and Bu^{n}Li (79.2 mmol) was added. After the reaction mixture had been placed in a freezer (−20 °C) for 14 h, 1,2-dibromo-1,1,2,2-tetrafluoroethane (15.5 cm³, 0.013 mol) was added at −60 °C, and the mixture was allowed to warm to room temp. and was quenched with water. The solvent was removed under reduced pressure, and the residue was acidified with dil. hydrochloric acid, extracted with dichloromethane, dried over $MgSO_4$, and concentrated to give a crude product. This was purified by column chromatography on silica with chloroform–hexane (1 : 1) as eluent to give *bromo aldehyde* **9** (4.22 g, 69%) as needles, mp 111–113 °C (Found: C, 47.22; H, 3.93. $C_9H_9BrO_2$ requires C, 47.19; H, 3.96%); ν_{max}(KBr)/cm^{-1} 2964, 2864, 1689, 1591, 1575, 1438, 1315, 1290, 1164, 1083 and 858; δ_H(270 MHz; $CDCl_3$) 2.38 (3 H, s, CH_3), 3.93 (3 H, s, OCH_3), 6.94 (1 H, m, 4-H), 7.34 (1 H, m, 6-H) and 10.40 (1 H, s, CHO).

3-Methoxy-5-methyl-2-(1′,4′,5′-trimethoxynaphthalen-2′-yl)-benzaldehyde 15

To a suspension of $Pd(PPh_3)_4$ (0.359 g, 0.310 mmol) in DME (5 cm³) was added a solution of aldehyde **9** (1.42 g, 6.21 mmol) in DME (22 cm³), and the mixture was stirred for 15 min under argon. A solution of boronic acid **8** (1.79 g, 6.83 mmol) in ethanol (4 cm³) was added, and the mixture was stirred for 10 min and was then treated with 2 M aq. sodium carbonate (7 cm³). The resulting mixture was heated under reflux for 24 h. After cooling, the solvent was removed under reduced pressure, and the residue was acidified with dil. hydrochloric acid and extracted with chloroform (3 × 40 cm³). The combined extracts were dried ($MgSO_4$) and evaporated. The crude product was purified by column chromatography on silica, using chloroform–hexane (1 : 1) as eluent, to give *biaryl aldehyde* **15** (1.77 g, 78%) as yellow needles, mp 171–174 °C (from CH_2Cl_2–hexane) (Found: C, 72.12; H, 6.05. $C_{22}H_{22}O_5$ requires C, 72.08; H, 6.02%); ν_{max}(KBr)/cm^{-1} 2933, 1693, 1600, 1581, 1458, 1386, 1377, 1309, 1284, 1269, 1126, 1080, 1043 and 763; δ_H(270 MHz; $CDCl_3$) 2.48 (3 H, s, CH_3), 3.45 (3 H, s, OCH_3), 3.79 (3 H, s, OCH_3), 3.91 (3 H, s, OCH_3), 3.98 (3 H, s, OCH_3), 6.71 (1 H, s, 3′-H), 6.92 (1 H, dd, *J* 8.1 and 1.1, 6′-H), 7.09 (1 H, d, *J* 1.1,

4-H), 7.44 (1 H, t, *J* 8.1, 7′-H), 7.50 (1 H, d, *J* 1.1, 6-H), 7.75 (1 H, dd, *J* 8.1 and 1.1, 8′-H) and 9.71 (1 H, s, CHO); *m*/*z* (EI) 366.1449 (M^+). $C_{22}H_{22}O_5$ requires *M*, 366.1467.

Oxidation of aldehyde 15

To a solution of aldehyde **15** (0.0882 g) in 10 cm^3 of acetone was added a slight excess of Jones' reagent. After addition of propan-2-ol, the solvent was removed under reduced pressure and the residue was extracted with dichloromethane. The crude product was purified by column chromatography on silica, using AcOEt–hexane (1 : 1) as eluent, to give enone **16** as an oil; ν_{max}(film)/cm^{-1} 1657, 1587, 1283, 1259 and 1242; δ_H(270 MHz; $CDCl_3$) 2.22 (3 H, s, $COCH_3$), 2.42 (3 H, s, CH_3), 3.73 (3 H, s, OCH_3), 4.05 (3 H, s, OCH_3), 6.64 (1 H, d, *J* 16.2, CH=C*H*CO), 6.76 (1 H, s, 3-H), 6.82 (1 H, m, ArH), 7.15 (1 H, m, ArH), 7.35 (1 H, dd, *J* 6.5 and 2.2, 6-H), 7.37 (1 H, d, *J* 16.2, C*H*=CHCO), 7.71 (1 H, t, *J* 6.5, 7-H) and 7.79 (1 H, dd, *J* 6.5 and 2.2, 8-H).

3-Methoxy-5-methyl-2-(1′,4′,5′-trimethoxynaphthalen-2′-yl)-benzoic acid 7

To a warm solution of aldehyde **15** (0.253 g, 0.690 mmol) in methanol (20 cm^3) maintained at 60 °C was added a mixture of 15% aq. sodium hydroxide (10 cm^3) and 35% hydrogen peroxide (5 cm^3). After further addition of hydrogen peroxide (40 cm^3), the mixture was heated under reflux for 1 h. After cooling, the solvent was removed under reduced pressure, and the residue was extracted with diethyl ether. The aqueous phase was acidified with dil. hydrochloric acid and extracted with dichloromethane. The combined extracts were dried ($MgSO_4$) and evaporated to give *biaryl acid* **7** (0.241 g, 91%) as a yellow solid (Found: C, 65.77; H, 5.90. $C_{22}H_{22}O_6 \cdot H_2O$ requires C, 65.98; H, 6.04%); ν_{max}($CHCl_3$)/cm^{-1} 3300, 2929, 1685, 1600, 1581, 1463, 1384, 1377, 1263, 1234 and 1080; δ_H(270 MHz; $CDCl_3$) 2.44 (3 H, s, CH_3), 3.50 (3 H, s, OCH_3), 3.72 (3 H, s, OCH_3), 3.85 (3 H, s, OCH_3), 3.97 (3 H, s, OCH_3), 5.51 (1 H, br, CO_2H), 6.59 (1 H, s, 3′-H), 6.87 (1 H, dd, *J* 8.4 and 1.0, 6′-H), 6.98 (1 H, m, 4-H), 7.38 (1 H, m, 6-H), 7.38 (1 H, t, *J* 8.4, 7′-H) and 7.70 (1 H, dd, *J* 8.4 and 1.0, 8′-H); *m*/*z* (EI) 382.1394 (M^+). $C_{22}H_{22}O_6$ requires *M*, 382.1416.

Cyclization of acid 7 with polyphosphoric acid

A mixture of acid **7** (0.071 g, 0.19 mmol) and polyphosphoric acid (~1.5 g) was heated at 120 °C for 10 h. After the mixture had been treated with water, it was filtered through a pad of Celite. The filtrate was extracted with CH_2Cl_2, and the extract was dried ($MgSO_4$) and evaporated. The crude product was triturated with diethyl ether and the undissolved material was removed. Evaporation of the solvent gave 1,10,12-trimethoxy-8-methyldibenzo[*c*,*h*]chromen-6-one as a brown oil (25 mg); ν_{max}(film)/cm^{-1} 1719, 1589; δ_H(60 MHz; $CDCl_3$) 2.50 (3 H, s, CH_3), 4.01 (3 H, s, OCH_3), 4.03 (3 H, s, OCH_3), 4.08 (3 H, s, OCH_3), 7.00 (1 H, dd, *J* 8.4 and 1.2, 4-H), 7.15 (1 H, m, 9-H), 7.51 (1 H, t, *J* 8.4, 3-H), 7.96 (1 H, m, 7-H), 8.22 (1 H, dd, *J* 8.4 and 1.2, 2-H) and 8.41 (1 H, s, 11-H).

4,5,9,10-Tetramethoxy-2-methylbenzo[*b*]fluoren-11-one 6

To a stirred solution of acid **7** (0.172 g, 0.450 mmol) in CH_2Cl_2 (8 cm^3) were added oxalyl dichloride (0.3 cm^3, 3.44 mmol) and DMF (10 drops) under nitrogen. After 1.5 h, the solution was evaporated under reduced pressure, the residue was dissolved in CH_2Cl_2 (20 cm^3), and the solution was cooled to 0 °C. To this solution was added titanium tetrachloride (0.13 cm^3, 0.935 mmol) and the mixture was poured into ice–water after 2 h and extracted with CH_2Cl_2 (3 × 20 cm^3). The combined extracts were dried ($MgSO_4$) and evaporated to leave a solid, which was purified by column chromatography on silica, using AcOEt–hexane (1 : 3) as eluent, to give the *benzo*[b]*fluorenone* **6** (0.110 g, 67%) as orange needles, mp 181–182 °C. The physical data of product **6** were identical with those reported by Gould and co-workers [14] except for the IR band at 1733 cm^{-1}.

4,5,9,10-Tetramethoxy-2-methylbenzo[*b*]fluoren-11-one *O*-benzyloxime 18

A mixture of ketone **6** (0.160 g, 0.439 mmol), *O*-benzylhydroxylamine hydrochloride (0.712 g, 4.46 mmol) and sodium acetate (0.357 g, 4.35 mmol) in methanol (30 cm^3)–water (6 cm^3) was heated under reflux for 4 h. The methanol was evaporated off, the mixture was extracted with chloroform (3 × 30 cm^3), and the organic phase was then washed with 0.5 M hydrochloric acid, dried ($MgSO_4$) and concentrated to give a yellow solid. The crude product was purified by column chromatography on silica, using $CHCl_3$–hexane (7 : 1), to give a mixture of *syn*- and *anti*-*O*-benzyloximes **18** (0.144 g, 70%) as yellow needles; ν_{max}(KBr)/cm^{-1} 2922, 1616, 1606, 1583, 1569, 1461, 1365, 1346, 1272, 1057, 1029, 945 and 740; δ_H(270 MHz; $CDCl_3$) 2.42 (s, CH_3), 2.44 (s, CH_3), 3.62 (s, OCH_3), 3.77 (s, OCH_3), 3.86 (s, OCH_3), 3.96 (s, OCH_3), 3.98 (s, OCH_3), 4.00 (s, OCH_3), 4.01 (s, OCH_3), 5.47 (s, ArCH_2O), 5.50 (s, ArCH_2O), 6.77–6.88 (m, ArH), 7.3–7.6 (m, ArH), 7.83 (dd, *J* 8.4 and 1.1, 6-H), 7.84 (dd, *J* 8.4 and 1.1, 8-H) and 8.12 (m, 1-H); *m*/*z* (EI) 469.1917 (M^+). $C_{29}H_{27}NO_5$ requires *M*, 469.1889.

4,9-Dimethoxy-2-methylbenzo[*b*]fluorene-5,10,11-trione 11-*O*-benzyloxime 5

To a solution of compound **18** (0.0878 g, 0.187 mmol) in a mixture of acetonitrile (10 cm^3) and water (0.8 cm^3) containing suspended pyridine-2,6-dicarboxylic acid *N*-oxide (0.091 g, 0.49 mmol) was added a cooled solution of CAN (0.262 g, 0.477 mmol) in a mixture of acetonitrile (2 cm^3) and water (0.8 cm^3) during 20 min. The reaction mixture was stirred for 20 min in an ice-bath and then at room temp. for 1 h. After removal of the solvent under reduced pressure, the mixture was extracted with chloroform (3 × 10 cm^3). The crude quinone was purified by column chromatography on silica, using AcOEt–hexane (1 : 7) as eluent, to give *quinone* **5** (0.0637 g, 77%) as red needles, mp 214–221 °C (Found: C, 72.62; H, 4.90; N, 3.33. $C_{27}H_{21}NO_5 \cdot 1/2H_2O$ requires C, 72.31; H, 4.93; N, 3.12%); ν_{max}(KBr)/cm^{-1} 3440, 2937, 1670, 1652, 1585, 1467, 1269, 1039 and 1008; δ_H(270 MHz; $CDCl_3$) 2.38 (3 H, s, CH_3), 3.99 (3 H, s, OCH_3), 4.00 (3 H, s, OCH_3), 5.66 (2 H, s, ArCH_2O), 6.85 (1 H, m, 3-H), 7.27 (1 H, dd, *J* 8.1 and 1.1, 8-H), 7.31–7.58 (5 H, m, Ph), 7.63 (1 H, t, *J* 8.1, 7-H), 7.78 (1 H, dd, *J* 8.1 and 1.1, 6-H) and 7.81 (1 H, m, 1-H); *m*/*z* (EI) 439.1399 (M^+). $C_{27}H_{21}NO_5$ requires *M*, 439.1420.

11-Amino-5-hydroxy-4,9-dimethoxy-2-methylbenzo[*b*]fluoren-10-one 4

A solution of oxime **5** (0.0295 g, 0.0668 mmol) in 2 cm^3 of glacial acetic acid was treated with zinc powder (0.0292 g, 0.0668 mmol) and boiled for 150 min. After filtration of the mixture, the filtrate was diluted with water and extracted with chloroform (3 × 10 cm^3). The combined extracts were washed with water, dried ($MgSO_4$) and evaporated. The residue was partitioned in ethyl acetate, and the small amount of precipitated material was filtered off. The filtrate was evaporated to leave title compound **4** (0.0179 g, 80%) as a purple solid, mp >300 °C; HPLC column: PRODIGY ODS-2 250 × 4.60 mm; mobile phase: MeCN–water 80 : 20→90 : 10 during 20 min; flow rate 1.00 cm^3 min^{-1}; pressure: 1800 psi; temp.: 20 °C; 15.49 min; ν_{max}(KBr)/cm^{-1} 3197, 2922, 1642, 1625, 1581, 1463, 1313 and 1296; λ_{max}(EtOH)/nm (ε) ($c = 2.03 \times 10^{-4}$ mol l^{-1}) 206 (1.07×10^4), 280 (1.11×10^4), 472 (2.67×10^3) and 506 (2.61×10^3); λ_{max}(EtOH–HCl)/nm 220, 280, 473 and 509; λ_{max}(EtOH–NaOH)/nm 212, 286, 347, 444 and 651; δ_H(270 MHz; $CDCl_3$) 2.22 (2 H, br, NH_2), 2.23 (3 H, s, CH_3), 3.81 (3 H, s, 9-OCH_3), 4.10 (3 H, s, 4-OCH_3), 6.60 (1 H, m, 1-H), 6.66 (1 H, m, 3-H), 6.98 (1 H, dd, *J* 8.1 and 1.1, 6-H), 7.48 (1 H, t, *J* 8.1, 7-H), 7.61 (1 H, dd, *J* 8.1 and 1.1, 8-H) and 10.63 (1 H, s, OH); δ_C(67.94 MHz; $CDCl_3$) 21.5, 55.9, 56.4, 111.1, 114.7, 114.8, 117.3, 118.1, 120.0, 121.2, 131.0, 133.6, 136.5, 138.1, 140.6, 142.5, 150.1, 151.5, 161.0 and 180.3; *m*/*z* (FAB, positive

mode, matrix: 3-nitrobenzyl alcohol) 639 (2M − 31), 623 and 609.

2-Hydroxymethyl-4,5,9,10-tetramethoxybenzo[*b*]fluoren-11-one 20

A solution of compound **6** (0.210 g, 0.577 mmol), NBS (0.124 g, 0.694 mmol) and benzoyl peroxide (0.025 g) in CCl_4 (20 cm^3) was refluxed for 2 days under nitrogen. The solvent was removed under reduced pressure, and the residue was extracted with chloroform. The combined extracts were washed with water, dried ($MgSO_4$) and evaporated to leave bromide **19** (0.255 g) as a yellow oil.

To a solution of bromide **19** in 1,4-dioxane (15 cm^3) was added an aqueous mixture of calcium carbonate (0.288 g, 2.87 mmol in 20 cm^3), and the mixture was refluxed for 20 h. After removal of the solvent, the residue was extracted with chloroform. The combined extracts were washed successively with dil. hydrochloric acid and water, dried ($MgSO_4$) and evaporated. The crude product was purified by column chromatography on silica with chloroform as eluent to give *alcohol* **20** (0.0953 g, 43%) as a brown solid, mp 202–204 °C; ν_{max}(KBr)/cm^{-1} 3517, 2931, 1701, 1602, 1587, 1573, 1357, 1350, 1051 and 771; δ_H(270 MHz; $CDCl_3$) 2.33 (1 H, br, OH), 3.87 (3 H, s, OCH_3), 3.88–3.92 (3 H, m, OCH_3), 4.01 (3 H, s, OCH_3), 4.02 (3 H, s, OCH_3), 4.74 (2 H, br s, CH_2OH), 6.91 (1 H, dd, *J* 8.1 and 1.1, 6-H), 7.10 (1 H, m, 3-H), 7.31 (1 H, m, 1-H), 7.49 (1 H, t, *J* 8.1, 7-H) and 7.77 (1 H, dd, *J* 8.1 and 1.1, 8-H); *m*/*z* (EI) 380.1286 (M^+). $C_{22}H_{20}O_6$ requires *M*, 380.1260.

2-Acetoxymethyl-4,5,9,10-tetramethoxybenzo[*b*]fluoren-11-one 21

To a solution of compound **20** (0.0998 g, 0.262 mmol) and catalytic amount of 4-(dimethylamino)pyridine (DMAP) in pyridine (4 cm^3) was added acetic anhydride (0.05 cm^3, 0.525 mmol), and the mixture was stirred for 2 min. After azeotropic distillation of the mixture with toluene, the residue was extracted with chloroform. The combined extracts were washed with water, dried ($MgSO_4$) and evaporated to give acetate **21** (0.105 g, 94%) as a reddish brown solid, mp 172 °C; ν_{max}(KBr)/cm^{-1} 2935, 1745, 1699, 1604, 1587, 1573, 1361, 1350, 1249, 1055 and 771; δ_H(270 MHz; $CDCl_3$) 2.15 (3 H, s, $COCH_3$), 3.90 (3 H, s, OCH_3), 4.00 (3 H, s, OCH_3), 4.04 (3 H, s, OCH_3), 4.06 (3 H, s, OCH_3), 5.14 (2 H, s, CH_2OAc), 6.92 (1 H, dd, *J* 8.1 and 1.1, 6-H), 7.10 (1 H, d, *J* 1.6, 3-H), 7.42 (1 H, d, *J* 1.6, 1-H), 7.51 (1 H, t, *J* 8.1, 7-H) and 7.79 (1 H, dd, *J* 8.1 and 1.1, 8-H).

2-Acetoxymethyl-4,5,9,10-tetramethoxybenzo[*b*]fluorene-11-one 11-*O*-benzyloxime 22

A mixture of ketone **21** (0.245 g, 0.581 mmol), *O*-benzylhydroxylamine hydrochloride (0.932 g, 5.84 mmol) and sodium acetate (0.481 g, 5.86 mmol) in methanol (30 cm^3)–water (5 cm^3) was heated under reflux overnight. The solvent was evaporated off, the mixture was extracted with chloroform, and the organic phase was then washed with dil. hydrochloric acid, dried ($MgSO_4$) and concentrated to give a yellow oil. The crude product was purified by column chromatography on silica, with AcOEt–hexane (6:1) as eluent, to give the syn- and anti-O-*benzyloxime* **22** as an orange oil (0.292 g, 95%); ν_{max}(film)/cm^{-1} 2931, 1739, 1616, 1604, 1583, 1509, 1463, 1454, 1371, 1348, 1271, 1236, 1064, 1058 and 1016; δ_H(270 MHz; $CDCl_3$) 2.08 (3 H, s, OCH_3), 2.15 (3 H, s, $COCH_3$), 3.79 (3 H, s, OCH_3), 3.86 (3 H, s, OCH_3), 3.97 (3 H, s, OCH_3), 3.99 (3 H, s, OCH_3), 4.03 (3 H, s, OCH_3), 4.40 (3 H, s, OCH_3), 5.15 (2 H, s, OCH_2OAc), 5.16 (2 H, s, OCH_2OAc), 5.48 (2 H, s, OCH_2Ph), 5.50 (2 H, s, OCH_2Ph), 6.89 (1 H, dd, *J* 8.1 and 1.1, 6-H), 6.95 (1 H, d, *J* 1.1, 3-H), 7.05 (1 H, d, *J* 1.1, 3-H), 7.27–7.60 (m), 7.85 (1 H, dd, *J* 8.1 and 1.1, 8-H) and 8.26 (1 H, d, *J* 1.1, 1-H); *m*/*z* (EI) 527.1944 (M^+). $C_{21}H_{29}NO_7$ requires *M*, 527.1744.

2-Acetoxymethyl-4,9-dimethoxybenzo[*b*]fluorene-5,10,11-trione 11-*O*-benzyloxime 23

To an ice-cold solution of compound **22** (0.0918 g, 0.174 mmol) and pyridine-2,6-dicarboxylic acid *N*-oxide (0.0907 g, 0.495 mmol) in acetonitrile (3 cm^3) was added cooled aq. CAN (0.240 g, 0.437 mmol in 3 cm^3) during 10 min. The reaction mixture was stirred for 20 min in an ice-bath and then at room temp. for 1 h. After removal of the solvent under reduced pressure, the residue was extracted with chloroform. The crude product was purified by column chromatography on silica, using AcOEt–hexane (1:3) as eluent, to give a mixture of the stereoisomers of *quinone* **23** (0.0457 g, 53%) as red needles, mp 172–181 °C; ν_{max}(KBr)/cm^{-1} 2933, 1741, 1672, 1587, 1535, 1467, 1367, 1307, 1274, 1230, 1193, 1037, 999 and 914; δ_H(270 MHz; $CDCl_3$) 2.04 (3 H, s, $COCH_3$), 2.14 (3 H, s, $COCH'_3$), 3.98 (3 H, s, OCH'_3), 4.01 (3 H, s, OCH_3), 4.02 (3 H, s, OCH_3), 5.10 (2 H, s, OCH_2O-Ac), 5.14 (2 H, s, OCH_2OAc), 5.59 (2 H, s, OCH_2Ph), 5.67 (2 H, s, OCH_2Ph), 6.93 (1 H, d, *J* 1.1, 3′-H), 7.02 (1 H, d, *J* 1.1, 3-H), 7.24–7.70 (m), 7.73 (1 H, dd, *J* 8.1 and 1.1, 8′-H), 7.79 (1 H, dd, *J* 8.1 and 1.1, 8-H) and 7.94 (1 H, d, *J* 1.1, 1-H); *m*/*z* (EI) 497.1487 (M^+). $C_{29}H_{23}NO_7$ requires *M*, 497.1475.

2-Acetoxymethyl-11-amino-5-hydroxy-4,9-dimethoxybenzo[*b*]-fluoren-10-one 10

A solution of compound **23** (0.0216 g, 0.0434 mmol) in 2 cm^3 of glacial acetic acid was treated with zinc powder (0.0160 g, 0.243 mmol) and boiled for 150 min. After filtration of the mixture, the filtrate was made basic with 30% aq. ammonium hydroxide and extracted with chloroform. The combined extracts were washed with water, dried ($MgSO_4$) and evaporated. The residue was dissolved in acetonitrile, and the small amount of solid was filtered off. The filtrate was evaporated to leave title compound **10** (0.0047 g, 28%) as a purple solid, mp 288 °C; HPLC column: PRODIGY ODS-2 250 × 4.60 mm; mobile phase: MeCN–water, 60:40→90:10 during 20 min; flow rate: 0.8 cm^3 min^{-1}; pressure: 1800 psi; temp.: 20 °C; t_R: 17.40 min; ν_{max}(KBr)/cm^{-1} 3205, 1739, 1641, 1622, 1581, 1463, 1309, 1288, 1267, 1224, 1122, 1039, 1022 and 704; δ_H(270 MHz; $CDCl_3$) 2.01 (3 H, s, $COCH_3$), 3.83 (3 H, s, OCH_3), 4.15 (3 H, s, OCH_3), 4.96 (2 H, ABq, *J* 11.9, CH_2OAc), 6.79 (1 H, d, *J* 1.1, 3-H), 6.85 (1 H, d, *J* 1.1, 1-H), 7.01 (1 H, dd, *J* 8.1 and 1.1, 6-H), 7.50 (1 H, t, *J* 8.1, 7-H), 7.63 (1 H, dd, *J* 8.1 and 1.1, 8-H) and 10.81 (1 H, s, OH); δ_C(125 MHz; $CDCl_3$) 21.00, 56.14, 56.71, 66.42, 110.15, 114.61, 114.95, 117.62, 118.21, 121.25, 122.61, 131.47, 134.00, 135.68, 136.35, 139.84, 142.52, 151.73, 151.79, 161.21, 170.77 and 180.67; *m*/*z* (FAB, positive mode, matrix: 3-nitrobenzyl alcohol) 755 (2M − 31) and 695.

Acetylation of compound 10

Compound **10** was acetylated as in a similar manner to the preparation of compound **23**, and the products were purified by column chromatography on silica, using AcOEt–hexane (1:1) as eluent, to give 11-acetamido-2-acetoxymethyl-5-hydroxy-4,9-dimethoxybenzo[*b*]fluoren-10-one **24** and 11-acetamido-5-acetoxy-2-acetoxymethyl-4,9-dimethylbenzo[*b*]fluoren-10-one **25**.

Physical data of compound **24**: δ_H(270 MHz; $CDCl_3$) 2.14 (3 H, s, $COCH_3$), 2.47 (3 H, s, $COCH_3$), 3.94 (3 H, s, OCH_3), 4.15 (3 H, s, OCH_3), 5.11 (2 H, s, CH_2OAc), 6.90 (1 H, dd, *J* 8.1 and 1.1, 6-H), 7.10 (1 H, d, *J* 1.1, 3-H), 7.40 (1 H, d, *J* 1.1, 1-H), 7.49 (1 H, t, *J* 8.1, 7-H), 7.87 (1 H, dd, *J* 8.1 and 1.1, 8-H) and 9.83 (1 H, s, OH); *m*/*z* (EI) 435.1314 (M^+). $C_{24}H_{21}NO$ requires *M*, 435.1318; *m*/*z* (FAB, positive mode, matrix: 3-nitrobenzyl alcohol) 437.1 (M + 2).

Physical data of compound **25**: δ_H(270 MHz; $CDCl_3$) 2.13 (3 H, s, $COCH_3$), 2.49 (3 H, s, $COCH_3$), 2.51 (3 H, s, $COCH_3$), 3.95 (3 H, s, OCH_3), 3.99 (3 H, s, OCH_3), 5.12 (2 H, s, CH_2OAc), 6.90 (1 H, dd, *J* 8.1 and 1.1, 6-H), 7.08 (1 H, d, *J* 1.1, 3-H), 7.38 (1 H, d, *J* 1.1, 1-H) and 7.44–7.54 (2 H, m, 7- and 8-H); *m*/*z* (FAB, positive mode, matrix: 3-nitrobenzyl alcohol) 479.15 (M + 2).

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