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Highly stereoselective Pictet–Spengler reaction of D-tryptophan methyl ester with piperonal: convenient syntheses of Cialis (Tadalafil), 12a-*epi*-Cialis, and their deuterated analogues

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Abstract—The acid-catalyzed Pictet–Spengler reaction of D-tryptophan methyl ester with piperonal in acetic acid has been reported, the best stereoselectivity (*cis*/*trans* = 92:8) was obtained with benzoic acid as the catalyst. The Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with piperonal in various solvents has been extensively studied, the solvent-dependence of stereoselectivities could be principally attributed to the solubility-difference between *cis* and *trans* products **5**-HCl in the used solvent, the best stereoselectivity (*cis*/*trans* = 99:1) was obtained using nitromethane or acetonitrile as the solvent. A base-catalyzed epimerization at 12a-position of Cialis **1** (tadalafil) in a DMSO-containing solvent was also exploited. Cialis, 12a-*epi*-Cialis **2**, deuterium-labeled 3,3,12a-d_3-Cialis **3**, and 3,3,12a-d_3-12a-*epi*-Cialis **4** were efficiently synthesized from D-tryptophan methyl ester hydrochloride.

1. Introduction

Recently the asymmetric Pictet–Spengler reaction has attracted much attention,[1–8] because it is an important and useful tool to construct chiral synthons containing tetrahydroisoquinolines or tetrahydro-β-carbolines structural moieties. The acid-catalyzed Pictet–Spengler reaction of D-tryptophan methyl ester with piperonal is the key step in the synthesis of tadalafil (Cialis, **1**)[9–16] which is a cGMP specific Type V phosphodiesterase (PDE5) inhibitor similar to sildenafil (Viagra)[17] and vardenafil (Levitra)[18] and has an improved PDE5/PDE6 selectivity compared to sildenafil.[19–21] These three agents are well tolerated and have been approved as first-line therapy for ED in recent years.[22] However the moderate *cis* stereoselectivity of the particular Pictet–Spengler reaction of D-tryptophan methyl ester with piperonal[23] retards large-scale preparation of the pharmaceutically valuable compound **1**, hence an endeavor should be made to develop a practical synthesis of **1** with a high stereoselectivity. Herein, we would like to disclose the results of our recent studies aiming at efficient, highly stereoselective syntheses of **1** and its analogues. Our studies include: (a) the acid-catalyzed stereoselective Pictet–Spengler reaction of D-tryptophan methyl ester with piperonal in acetic acid; (b) the stereoselective Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with piperonal in various solvents; (c) the epimerization of compound **1** under basic conditions; (d) convenient syntheses of Cialis **1**, 12a-*epi*-Cialis **2**, deuterium-labeled 3,3,12a-d_3-Cialis **3**, and 3,3,12a-d_3-12a-*epi*-Cialis **4**.

2. Results and discussion

The trifluoroacetic acid-catalyzed Pictet–Spengler reaction of D-tryptophan methyl ester with piperonal in dichloromethane gives a mixture of *cis* and *trans* tetrahydro-β-carboline derivative **5** in a 3:2 ratio.[23] This disappointing stereoselectivity means that the mixture of *cis*-**5** and *trans*-**5** is hard to separate without chromatography, and may accordingly make the reaction hard to scale up.

We are interested in improving the stereoselectivity of the Pictet–Spengler reaction of D-tryptophan methyl ester with piperonal aiming at a practical synthesis of compound **1** and its analogues. At first, we tried the Pictet–Spengler reaction of D-tryptophan methyl ester with piperonal in acetic acid with or without an additive acid as a catalyst. Some results are included in Table 1. As can be seen in

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Table 1. Acids-catalyzed stereoselective Pictet–Spengler reaction of tryptophan methyl ester with piperonal in acetic acid at rt

Entry	Acids (catalyst)	Time (h)	Yield (%)	Selectivity (*cis*:*trans*)
1	None	85	80	55:45
2	Formic acid	20	78	70:30
3	Benzoic acid	16	86	92:8
4	Salicylic acid	22	84	74:26
5	D-Tartaric acid	28	65	85:15
6	L-Tartaric acid	28	67	86:14
7	CF_3COOH	24	70	72:28
8	HCl	22	82	80:20
9	H_3PO_4	24	60	64:36
10	H_3BO_3	30	70	75:25

Table 1, the reaction took a long time and gave a poor stereoselectivity in the absence of acid catalyst (Table 1, entry 1), the addition of acid catalyst improved the stereoselectivities and also increased the reaction rate, benzoic acid gave the best yield and the highest stereoselectivity (Table 1, entry 3).

Though the high selectivity (92:8) could be obtained with benzoic acid as a catalyst, it was not satisfactory yet and we should not stop searching for an alternative method to get better results. During the course of our study, a patent[24] issued to Lilly Icos described a modified Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with piperonal in isopropanol. Wu and his co-workers also reported an improved synthesis of **1**.[25] These two methods overcame the drawback of using the corrosive trifluoroacetic acid (TFA) and improved the yield and purity of the desired *cis*-**5**, however there are still some problems to be solved. For example, the nature and limitation of the modified Pictet–Spengler reaction should be further studied and better understood, the effect of solvents should also be studied in detail.

With these problems in mind, we then extensively studied the condition and possible mechanism of the reaction. Table 2 shows the solvent effect on the Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with piperonal. As can be seen in Table 2, 14 solvents have been examined to observe their impact on the yields and stereoselectivities. Isopropanol, butanol, pentanol, nitromethane, acetonitrile, 1,2-dichloroethane, and 1,2-dimethoxyethane were suitable solvents and gave good to high yields and excellent stereoselectivities (Table 2, entries 1, 2, 6–9, 14, and 15). Methanol, dimethylsulfoxide, and *N*,*N*-dimethylformamide gave only moderate yields and low stereoselectivities (Table 2, entries 3, 12, and 13). After comparing the experiments performed in various solvents, it was found that the solvents in which product hydrochloride salt of **5** (**5**-HCl) has low solubility gave better results than the solvents in which the product **5**-HCl has good solubility. For example, **5**-HCl has less solubility in nitromethane or acetonitrile, it did not dissolve during the reaction, and precipitated from the reaction solution, the best stereoselectivity was obtained. While product **5**-HCl dissolved in solvents such as methanol, DMSO, or DMF, the reaction mixture became a clear solution during the reaction, and the worst stereoselectivity was obtained. It

Table 2. The solvent effect on the Pictet–Spengler reaction of tryptophan methyl ester hydrochloride with piperonal

Entry	Solvent	Temp. (°C)	Time (h)	Yield (%)	Selectivity (*cis*:*trans*)
1	*i*-PrOH	83	8	92	97:3
2	*i*-PrOH	83	16	93	97:3
3	MeOH	65	19	71	46:54
4	EtOH	78	20	75	88:12
5	PrOH	97	10	79	90:10
6	BuOH	118	3	82	95:5
7	Pentanol	120	8	80	96:4
8	CH_3NO_2	101	4	94	99:1
9	CH_3CN	81	8	91	99:1
10	THF[a]	65	27	58[f]	95:5
11	$CHCl_3$	61	34	80	89:11
12	DMSO[b]	90	20	54[f]	50:50
13	DMF[c]	90	20	52[f]	51:49
14	DCE[d]	83	23	93	96:4
15	DME[e]	85	14	89	97:3

[a] Tetrahydrofuran.
[b] Dimethylsulfoxide.
[c] *N*,*N*-Dimethylformamide.
[d] 1,2-Dichloroethane.
[e] 1,2-Dimethoxyethane.
[f] The reactions were slow, 10–30% of the starting material was recovered.

was deduced that the solubility difference between *cis*- and *trans*-**5**-HCl played an important role and was the key factor for stereoselectivity, a good solvent should let the desired *cis* product *cis*-**5**-HCl precipitate from the solution and leave the undesired *trans* product *trans*-**5**-HCl dissolve in the mother liquor. The solubility of *cis*- and *trans*-**5**-HCl was measured in isopropanol, the ratio of solubility of *cis*- and *trans*-**5**-HCl in *i*-PrOH is 1:6.4 at room temperature (0.182 g/100 mL for *cis*, 1.17 g/100 mL for *trans*) and 1:11 at refluxing temperature (0.291 g/100 mL for *cis*, 3.218 g/100 mL for *trans*). The big difference of the solubility of *cis*- and *trans*-**5**-HCl favors good stereoselectivity.

It should be pointed out that when less polar solvents such as benzene, toluene, carbon tetrachloride, hexane, and pentane were used, the Pictet–Spengler reaction did not take place smoothly, and it took a long time and by-products were formed.

Some experiments helpful for understanding and elucidating the details of the Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with piperonal were performed. By monitoring the reaction by TLC, we found that the reaction produced both *cis*-**5**-HCl and *trans*-**5**-HCl in almost equal amounts at the beginning, and then the *trans*-**5**-HCl gradually transformed into *cis*-**5**-HCl in a solvent (CH_3CN, CH_3NO_2, etc.) with relatively less dissolving capacity for **5**-HCl. Especially when the starting material disappeared, the transformation went too fast, and finally produced almost only *cis*-**5**-HCl after suction leaving only a little of *trans*-**5**-HCl in the mother liquor. The above fact obviously suggested that an epimerization equilibrium between *cis*-**5**-HCl and *trans*-**5**-HCl existed during the reaction, the major driving force of the transformation of *cis*-**5**-HCl into *trans*-**5**-HCl was the large solubility difference between them. Several designed experiments summarized

in Table 3 also supported this suggestion. When we treated a mixture of *cis*- and *trans*-**5**-HCl of various ratios by refluxing for 8 h in different solvents, it was observed that the solvents (*i*-PrOH, CH_3CN, and CH_3NO_2) with a low dissolving capacity could transform the starting mixture of **5**-HCl with lower ratio (*cis*:*trans*) into product **5**-HCl with higher ratio (Table 3, entries 1, 2 and 6, 7), meanwhile the solvents (CH_3OH and DMSO) with a good dissolving capacity could transform the starting mixture of **5**-HCl with higher ratio into product **5**-HCl with lower ratio (Table 3, entries 4 and 5).

Table 3. The epimerization of a mixture of *cis*- and *trans*-**5**-HCl after refluxing for 8 h in different solvents

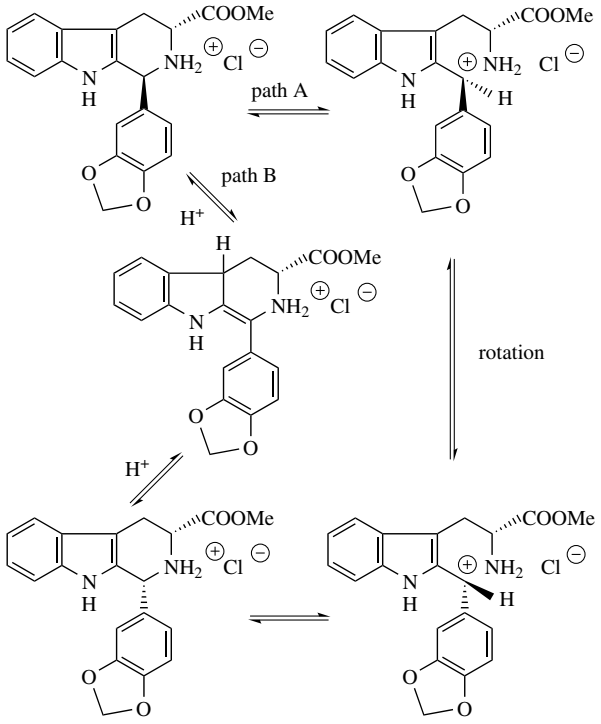
Entry	Starting **5** (*cis*:*trans*)	Solvent	Product **5** (*cis*:*trans*)	Yield (%)
1	46:54	*i*-PrOH	97:3	90
2	88:12	*i*-PrOH	97:3	89
3	97:3	*i*-PrOH	97:3	94
4	99:1	MeOH	47:53	70
5	99:1	DMSO[a]	52:48	65
6	46:54	CH_3CN	99:1	90
7	46:54	CH_3NO_2	99:1	92

[a] Stirring at 80 °C for 8 h.

The thermodynamic stability difference between *cis*- and *trans*-**5**-HCl could also significantly contribute to the high stereoselectivity, because the ratio of *cis*- and *trans*-**5**-HCl obtained in *i*-PrOH is 32:1 (Table 2, entries 1 and 2), while the solubility-difference is only 1:11. The *cis* isomer, in which 3,4-methylenedioxyphenyl and methoxycarbonyl groups on the piperidine ring are both in equatorial positions in the chair conformation, is probably more stable than the *trans* isomer.

A possible mechanism for the epimerization between *cis*- and *trans*-**5**-HCl is proposed in Scheme 1. There are two reasonable pathways[1,26,27] for acid-catalyzed epimerization of *cis* and *trans* intermediates **5**-HCl. Path A involves scission of the carbon–nitrogen bond, rotation of the carbon–carbon bond, and reformation of a carbon–nitrogen bond. Path B involves double migration of the double bond catalyzed by acid. If the epimerization follows path B, then deuterium would have been incorporated into the tetrahydro-β-carboline at the C-1 position upon protonation of the olefinic intermediate in the deuterated media. If the epimerization follows path A, then deuterium would not have been incorporated into the tetrahydro-β-carboline because there was no exchange of a proton at all between the tetrahydro-β-carboline scaffold and the deuterated media. When we operated the reaction in a mixed solvent of acetonitrile and heavy water (20:1), we did not get any evidence of the incorporation of a deuterium atom at the C-1 position of the tetrahydro-β-carboline by the analysis of ^1H NMR and MS, which implies that the epimerization of *cis*- and *trans*-**5**-HCl follows the mechanism of path A exclusively.

It is noteworthy that the scope of the above-described solvent-dependent highly stereoselective Pictet–Spengler reaction is quite limited. Unlike D-tryptophan methyl ester

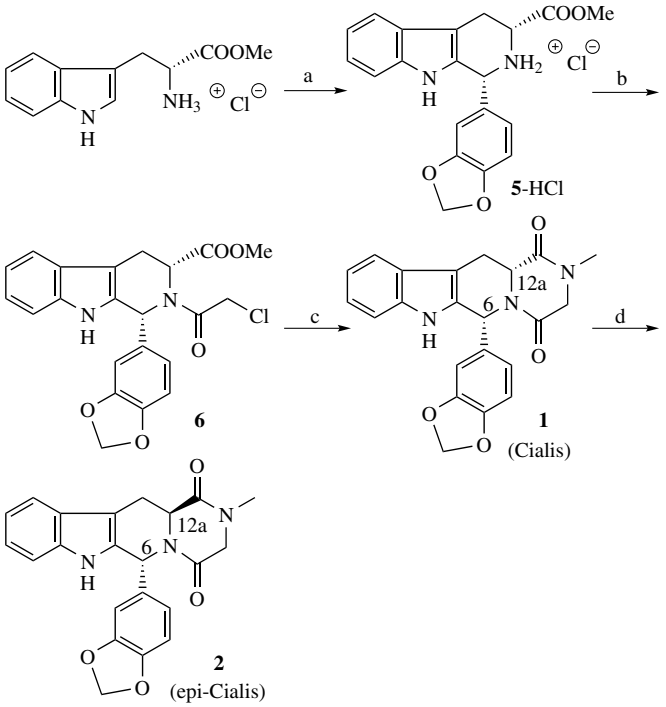


Scheme 1. Possible mechanism for acid-catalyzed epimerization of *cis* and *trans* intermediate **5**-HCl.

hydrochloride, the Pictet–Spengler reaction of D-tryptophan ethyl ester hydrochloride or D-tryptophan propyl ester hydrochloride with piperonal gave only a moderate *cis*/*trans* stereoselectivity in isopropanol, 1,2-dimethoxyethane, nitromethane or acetonitrile, probably due to the low solubility-difference of the corresponding *cis* and *trans* products. When we tried the Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with other aldehydes such as benzyl aldehyde, 2-chlorobenzyl aldehyde, salicylaldehyde, or vanillin in isopropanol, 1,2-dimethoxyethane, nitromethane or acetonitrile, only moderate *cis*/*trans* stereoselectivity could be obtained.

Scheme 2 depicts an efficient and stereospecific synthesis of Cialis **1** as well as 12a-*epi*-Cialis **2**. Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with equal molar piperonal by refluxing for 4 h in nitromethane afforded *cis*-**5**-HCl in 98% ee and 94% yield. The hydrochloride salt of *cis* tetrahydro-β-carboline derivative *cis*-**5**-HCl was directly treated with 1.5 equiv of chloroacetyl chloride in dichloromethane at 0 °C in the presence of 3 equiv of triethylamine to form *N*-chloroacetyl tetrahydro-β-carboline derivative **6** in 92% yield. Then compound **6** reacted with 5 equiv of methylamine overnight in DMF at room temperature to furnish Cialis **1** in 95% yield.

When we operated the reaction of compound **6** with methylamine in DMSO, we found that the reaction produced both **1** and its epimer 12a-*epi*-Cialis **2**. The amount of **2** increased as the temperature elevated. Jiang et al.[28] have also observed that a small amount of 12a-*epi*-Cialis **2** formed during the cyclization of compound **6** with methylamine in methanol at 50 °C because of the simultaneous



Scheme 2. Synthesis of Cialis and 12a-*epi*-Cialis. Reagents and conditions: (a) 1 equiv of piperonal, refluxing in nitromethane for 4 h, 94%; (b) 1.5 equiv of chloroacetyl chloride and 3 equiv of triethylamine, 0–5 °C for 2 h in CH_2Cl_2, 92%; (c) 5 equiv of methylamine, rt overnight in DMF, 95%; (d) 2 equiv of DBU, 83 °C for 5 h in a mixed solvent of DMSO and *i*-PrOH (1:5), 98%.

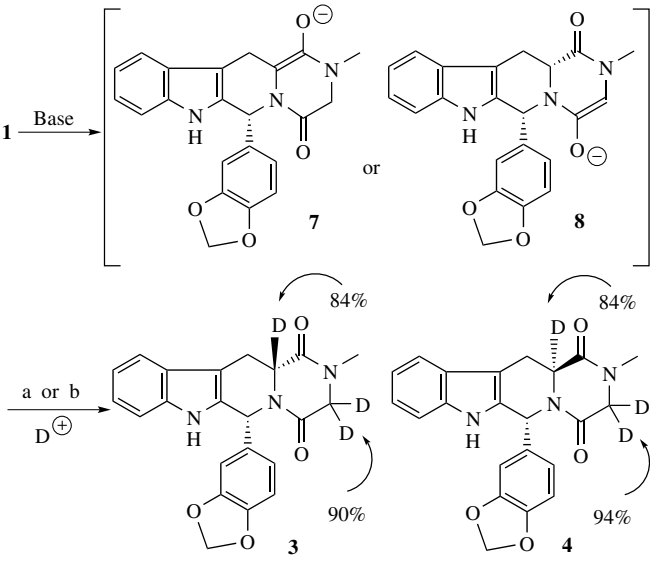
epimerization of **1**. To understand the epimerization of **1** at the C-12a position more clearly, we tried the epimerization of **1** in different solvents using various bases as the catalyst. Table 4 summarizes the outcomes of epimerization of **1** into **2**. It should be pointed out that DMSO was crucial for the epimerization, the reaction took place smoothly in DMSO or a mixed solvent containing DMSO, while it was very slow in other solvents. When a strong base such as potassium hydroxide or potassium *tert*-butoxide was used, the reaction was fast, but the yield was not high (Table 4, entries 1 and 2). A weak base such as DBU or potassium carbonate turned out to be a suitable catalyst, and the yield was high (Table 4, entries 3–7). Cialis **1** was almost quantitatively transformed into 12a-*epi*-Cialis **2** in a mixed solvent (DMSO–*i*-PrOH = 1:5) in the presence of 2 equiv of DBU after refluxing for 5 h.

Table 4. The base-catalyzed epimerization of Cialis at the C-12a position to form 12a-epi-Cialis **2**

Entry	Solvent (ratio)	Base (equiv)	Condition	Yield (%)
1	DMSO	KOH (2)	rt, 4 h	82
2	DMSO	*t*-BuOK (3)	0 °C, 0.5 h	83
3	DMSO	DBU[a] (3)	70 °C, 10 h	91
4	DMSO (1) THF (5)	DBU (2)	70 °C, 10 h	96
5	DMSO (4) H_2O (1)	KCO_3 (2)	65 °C, 15 h	95
6	DMSO (1) DME[b] (9)	DBU (3)	85 °C, 9 h	97
7	DMSO (1) *i*-PrOH (5)	DBU (2)	83 °C, 5 h	98

[a] 1,8-Diazabicyclo[5,4,0]undec-7-ene.
[b] 1,2-Dimethoxyethane.

We supposed that the base-catalyzed epimerization of Cialis **1** proceeded via an enolate **7** by enolization because the proton adjacent to the carbonyl group at C-12a position was acidic (Scheme 3). To verify this assumption, we tried the epimerization of **1** in a deuterated solvent. When the reaction was performed in the mixed deuterated solvent (DMSO-d_6–THF–D_2O = 1.5:6:1) using anhydrous K_2CO_3 as a base and refluxing at 65 °C for 16 h, we found out that not only the acidic proton on C-12a was replaced by a deuterium atom, but also two acidic protons on C-3 were also replaced by two deuterium atoms due to the formation of another enolate **8** under the basic condition, thus 3,3,12a-d_3-12a-*epi*-Cialis **4** could be obtained in 97% yield. If we want to prepare deuterated Cialis, we let the reaction to stop at the mid-point before Cialis **1** is totally transformed into 12a-*epi*-Cialis **4**. When the reaction was performed in a mixed deuterated solvent (DMSO-d_6–THF–D_2O = 4.5:18:1.5) in the presence of 1 equiv of anhydrous K_2CO_3, and the refluxing was allowed to continue only for 6.5 h, rapid cooling and quenching with an acid afforded 3,3,12a-d_3-Cialis **3** in 33% yield and 3,3,12a-d_3-12a-*epi*-Cialis **4** in 65% yield. Two deuterated compounds **3** and **4** may be useful for measuring the plasma concentration of these drugs using GC–MS techniques.[29–32] Percentages of deuterium-labeling in **3** and **4** obtained from the above procedure were higher than 83% at the C-12a position and 90% at the C-3 position. Unfortunately when we tried to purify **3** and **4** by recrystallization in some solvents, almost no change in the purity was obtained at all. For example, even after two recrystallizations in 1,2-dichloro-



Scheme 3. Preparation of deuterated Cialis **3** and deuterated 12a-*epi*-Cialis **4**. Reagents and conditions: (a) 1 equiv of K_2CO_3, DMSO-d_6–THF–D_2O = 1.5:6:1, 65 °C for 16 h, to form **4** in 95% yield; (b) 1 equiv of K_2CO_3, DMSO-d_6–THF–D_2O = 4.5:18:1.5, 65 °C for 6.5 h, to form **3** in 33% yield and **4** in 65% yield.

ethane, only 84% of C-12a position and 94% of C-3 position in **4** were occupied by deuterium atoms, and 84% of C-12a position and 90% of C-3 position in **3** were occupied by deuterium atoms.

3. Conclusion

In summary, reaction conditions, possible mechanism, scope, and limitation of the highly stereoselective Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with piperonal were studied extensively. The solvent-dependence of the yield and *cis* stereoselectivity of the Pictet–Spengler reaction was revealed and rationalized. Cialis was efficiently synthesized from D-tryptophan methyl ester hydrochloride in three steps in 82% overall yield, and 12a-*epi*-Cialis was obtained from Cialis in an almost quantitative yield in a solvent containing DMSO under basic conditions. A base-catalyzed epimerization of Cialis in a deuterated solvent was also studied, and deuterium-labeled 3,3,12a-d_3-Cialis and 3,3,12a-d_3-12a-*epi*-Cialis were prepared.

4. Experimental

4.1. General methods

Melting points are uncorrected. NMR spectra were acquired on Bruker AM-500, chemical shifts of ^{1}H NMR were given on the delta scale as parts per million (ppm) with tetramethylsilane (TMS) as the internal standard. IR spectra were recorded on Nicolet Magna IR-550. Mass spectra were recorded on HP5989A. Optical rotation was measured on WZZ-1S automatic polarimeter at room temperature. Column chromatography was performed on silica gel. HPLC analysis was performed with an Agilent/HP 1100 series equipped with an Alltech ELSD 2000ES detector. All solvents were purified by standard procedures. All chemicals were analytically pure. The D-tryptophan methyl ester hydrochloride was prepared according to a known procedure.[24]

4.2. Typical procedure of acid-catalyzed Pictet–Spengler reaction of D-tryptophan methyl ester with piperonal in acetic acid

A solution of D-tryptophan methyl ester (8.57 g, 39.27 mmol) in 50 mL of acetic acid was transferred into a three-necked round-bottom flask equipped with a stirring bar and a thermometer. The mixture was cooled down to 0 °C by an ice bath, piperonal (6.50 g, 43.30 mmol) and benzoic acid (1.12 g, 9.17 mmol) were then added. After stirring was continued at 0–5 °C for about 16 h, TLC showed that the reaction was complete. The products were allowed to partition between dichloromethane (200 mL) and 10% aqueous ammonia, the aqueous solution was adjusted to pH 9–10. After the extraction of aqueous solution once more with dichloromethane (100 mL), the organic extracts were combined and dried over anhydrous magnesium sulfate. Evaporation of the solvent gave a residue which was purified by chromatography (eluant: CH_2Cl_2–CH_3OH = 99:1) to give the *cis*-product (1*R*,3*R*)-1-(3,4-methylenedioxyphenyl)-2,3,4,9-tetrahydro-9*H*-pyrido[3,4-*b*]-indole-3-carboxylic methyl ester *cis*-**5** (10.88 g, 31.05 mmol) in 79% yield and *trans*-product (1*S*,3*R*)-1-(3,4-methylenedioxyphenyl)-2,3,4,9-tetrahydro-9*H*-pyrido[3,4-*b*]indole-3-carboxylic methyl ester *trans*-**5** (0.95 g, 2.71 mmol) in 7% yield.

cis-**5**: mp 153–155 °C (lit.[23] 154–155 °C), $[\alpha]_D^{20} = +25.2$ (*c* 1.0, $CHCl_3$) {lit.[23] $[\alpha]_D^{20} = +24.4$ (*c* 1.0, $CHCl_3$)}, ^{1}H NMR (acetone-d_6) δ 8.01 (s, 1H), 7.48 (dd, $J = 6.9$ Hz; 1.2 Hz, 1H), 7.24 (dd, $J = 7.1$ Hz; 0.9 Hz, 1H), 6.96–7.05 (m, 2H), 6.93 (dd, 7.9 Hz; 1.6 Hz, 1H), 6.85 (d, $J =$ 1.6 Hz, 1H), 6.81 (d, $J = 7.9$ Hz, 1H), 5.97 (d, $J = 0.8$ Hz, 1H), 5.96 (d, $J = 0.9$ Hz, 1H), 5.21 (s, 1H), 3.90 (dd, $J = 11.1$ Hz; 4.1 Hz, 1H), 3.76 (s, 3H), 3.07–3.13 (m, 1H), 2.85–2.92 (m, 1H). MS (*m*/*z*, relative intensity) 350 (M^+, 100), 333(23), 291(46), 274(24), 263(49), 233(25), 204(32), 169(23), 144(16), 115(11), 77(5), 44(4). IR (KBr film) 3315, 2930, 1700, 1479, 1435, 1238, 1039, 740 cm^{-1}.

trans-**5**: mp 186–188 °C (lit.[23] 188 °C), $[\alpha]_D^{20} = +33.1$ (*c* 1.0, $CHCl_3$) {lit.[23] $[\alpha]_D^{20} = +32.4$ (*c* 1.0, $CHCl_3$)}, ^{1}H NMR (acetone-d_6) δ 9.72 (s, 1H), 7.48 (d, $J = 7.8$ Hz, 1H), 7.27 (d, $J = 8.0$ Hz, 1H), 6.97–7.07 (m, 2H), 6.81 (d, $J =$ 0.8 Hz, 1H), 6.72–6.78 (m, 2H), 5.95 (d, $J = 0.9$ Hz, 1H), 5.94 (d, 0.9 Hz, 1H), 5.34 (s, 1H), 3.91 (dd, $J = 6.9$ Hz; 5.3 Hz, 1H), 3.65 (s, 3H), 3.10–3.17 (m, 1H), 2.96–3.03 (m, 1H). MS (*m*/*z*, relative intensity) 350 (M^+, 86), 333(34), 289(52), 274(39), 262(45), 233(44), 204(100), 169(58), 144(33), 115(34), 102(19), 77(10), 63(14). IR (KBr film) 3316, 2893, 1705, 1479, 1438, 1238, 1040, 744 cm^{-1}.

4.3. Typical procedure for a Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with piperonal in different solvents (for example, in nitromethane)

A powder of D-tryptophan methyl ester hydrochloride (10.00 g, 39.26 mmol) was added into a solution of piperonal (5.90 g, 39.30 mmol) in 100 mL of nitromethane. The suspension was then heated to reflux, refluxing was continued for 4 h while stirring, and the reaction was monitored by TLC. After the reaction was complete, the mixture was gradually cooled to room temperature and then to 0 °C. After standing in a refrigerator for about 2 h, the product as a pale yellow solid was collected on a Büchner funnel by suction and washed with a small amount of fresh nitromethane. The product was dried in a warm air to produce (1*R*,3*R*)-1-(3,4-methylenedioxyphenyl)-2,3,4,9-tetrahydro-9*H*-pyrido[3,4-*b*]indole-3-carboxylic methyl ester hydrochloride **5**-HCl (14.27 g, 36.89 mmol) in 94% yield, mp 239–241 °C, $[\alpha]_D^{20} = +92.9$ (*c* 1.1, MeOH). ^{1}H NMR (DMSO-d_6) δ 10.84 (s, 1H), 10.68 (s, 1H), 10.23 (s, 1H), 7.53 (d, $J = 7.8$ Hz, 1H), 7.28 (d, $J = 8.1$ Hz, 1H), 7.11 (dd, $J = 7.4$ Hz; 7.7 Hz, 1H), 7.01–7.07 (m, 3H), 6.99 (s, 1H), 6.09 (s, 2H), 5.80–5.90 (m, 1H), 4.70–4.80 (m, 1H), 3.83 (s, 3H), 3.24–3.39 (m, 2H). MS (*m*/*z*, relative intensity) 351 (M^++1, 21), 350 (M^+, 100), 333(24), 291(46), 274(24), 263(48), 233(25), 204(32), 169(23), 144(16), 115(11), 77(5). IR (KBr film) 3204, 2910, 2524, 2422, 2387, 2280, 1742, 1569, 1506, 1492, 1450, 1331, 1307, 1257, 1238, 1096,

1039, 932, 814, 747, 613 cm^{-1}. HPLC analysis after neutralization of the sample showed that it contained 99% of *cis*-isomer and 1% of *trans*-isomer. HPLC details: column: Fuji silysia C_{18}, 4.6 × 200 mm; temperature: 25 °C; mobile phase: $CH_3CN–H_2O = 50:50$; wave length of UV Det. = 280 nm; injection volume: 10 μL; flow rate: 1 mL/min; retention time: 11.1 min for *trans*-isomer, 12.5 min for *cis*-isomer.

4.4. Pictet–Spengler reaction of D-tryptophan methyl ester hydrochloride with piperonal in a deuterated solvent

A powder of D-tryptophan methyl ester hydrochloride (2.00 g, 7.85 mmol) was added into a solution of piperonal (1.30 g, 8.66 mmol) in a mixed solvent of 20 mL of acetonitrile and 1 mL of D_2O. The suspension was then heated at reflux; refluxing was continued for 32 h while stirring. The mixture was allowed to cool to room temperature and then to 0 °C. The pale yellow solid was collected on a Büchner funnel by suction and washed twice with fresh acetonitrile. The product (2.66 g, 6.88 mmol) was then obtained in 88% yield after drying. ^{1}H NMR and MS analyses of this product showed that it was identical with the sample (**5**-HCl) which was obtained from the reaction in a non-deuterated solvent.

4.5. (1*R*,3*R*)-1-(3,4-Methylenedioxyphenyl)-2-chloroacetyl-2,3,4,9-tetrahydro-9*H*-pyrido[3,4-*b*]indole-3-carboxylic methyl ester 6

Compound **5**-HCl (14.27 g, 36.89 mmol), triethylamine (11.20 g, 110.68 mmol) and 200 mL of dichloromethane were added into a three-necked round-bottom flask which was equipped with a mechanical stirrer and a thermometer. A powder of **5**-HCl dissolved after stirring for about 15 min at room temperature, a clear solution was obtained and then cooled to 0 °C by an ice bath. Chloroacetyl chloride (6.25 g, 55.34 mmol) was added slowly within 30 min keeping the inner temperature below 5 °C. The reaction continued for around 2 h at 0–5 °C and was monitored by TLC. After the reaction was complete, an aqueous solution of potassium carbonate (10%, 160 mL) was added and stirred vigorously. The organic phase was separated and dried over anhydrous magnesium sulfate. Removal of solvents under reduced pressure gave a crude product which was purified by recrystallization in the mixed solvent of methanol and water (8:2) to afford the compound **6** (14.49 g, 33.95 mmol) in 92% yield, mp 230–232 °C (lit.[23] 233 °C), $[\alpha]_D^{20} = -126.0$ (*c* 1.1, $CHCl_3$) {lit.[23] $[\alpha]_D^{20} = -125.4$ (*c* 1.17, $CHCl_3$)}. ^{1}H NMR (DMSO-d_6) δ 10.87 (s, 1H), 7.54 (d, $J = 7.8$ Hz, 1H), 7.27 (d, $J = 8.0$ Hz, 1H), 7.09 (dd, $J = 7.2$ Hz; 7.8 Hz, 1H), 7.02 (dd, $J = 7.4$ Hz; 7.4 Hz, 1H), 6.80 (d, $J = 8.1$ Hz, 1H), 6.75 (s, 1H), 6.63 (s, 1H), 6.45 (d, $J = 8.1$ Hz, 1H), 5.97 (d, $J = 10.1$ Hz, 2H), 5.20 (d, $J = 6.7$ Hz, 1H), 4.84 (d, $J = 13.9$ Hz, 1H), 4.43 (d, $J = 13.9$ Hz, 1H), 3.46 (d, $J = 15.9$ Hz, 1H), 3.07 (dd, $J = 15.9$ Hz; 6.9 Hz, 1H), 3.02 (s, 3H). MS (*m*/*z*, relative intensity) 428 (M^++2, 5), 426 (M^+, 15), 391(20), 349(100), 331(5), 289(32), 274(13), 262(6), 231(4), 204(13), 169(6), 144(2), 115(3), 102(2), 77(3). IR (KBr film) 3245, 2945, 1734, 1657, 1501, 1486, 1435, 1414, 1309, 1285, 1236, 1211, 1157, 1037, 934, 870, 806, 745, 692 cm^{-1}.

4.6. (6*R*,12a*R*)-2,3,6,7,12,12a-Hexahydro-2-methyl-6-(3,4-methylenedioxyphenyl)-pyrazino-[2′,1′:6,1]pyrido[3,4-*b*]indole-1,4-dione Cialis, 1

Compound **6** (14.49 g, 33.95 mmol) was dissolved in 150 mL of DMF. An aqueous solution of methylamine (30%, 17.57 g, 169.70 mmol) was added dropwise at room temperature. After the addition was finished, the reaction was continued overnight at rt and monitored by TLC. When the reaction was complete, 500 mL of deionized water was added slowly while the mixture was rapidly stirred. Rapid stirring was continued for 2 h, and then the suspension was filtered through a Büchner funnel by suction and the cake was washed successively with deionized water and isopropanol. The title compound **1** (12.51 g, 32.13 mmol) was obtained as an off-white solid in 95% yield after drying. It was pure enough, but for increased purity, compound **1** can be recrystallized from acetic acid. Mp 300–301 °C (lit.[23] 302–303 °C), $[\alpha]_D^{20} = +71.5$ (*c* 1.0, $CHCl_3$) {lit.[23] $[\alpha]_D^{20} = +71.0$ (*c* 1.0, $CHCl_3$)}. ^{1}H NMR (DMSO-d_6) δ 11.02 (s, NH on the indole ring), 7.54 (d, 7.8 Hz, 1H), 7.29 (d, $J = 8.0$ Hz, 1H), 7.05 (dd, $J = 7.8$ Hz; 7.2 Hz, 1H), 6.99 (dd, $J = 7.8$ Hz; 7.1 Hz, 1H), 6.86 (s, 1H), 6.77 (s, 2H), 6.12 (s, 1H), 5.91 (s, 2H), 4.39 (dd, $J = 4.2$ Hz; 11.6 Hz, 1H), 4.17 (d, $J = 17.2$ Hz, 1H), 3.94 (d, $J = 17.1$ Hz, 1H), 3.51 (dd, $J = 4.5$ Hz; 15.8 Hz, 1H), 2.96 (dd, $J = 11.8$ Hz; 15.8 Hz, 1H), 2.92 (s, 3H). ^{13}C NMR (DMSO-d_6) δ 166.88, 166.56, 147.06, 146.08, 137.00, 136.23, 133.96, 125.78, 121.25, 119.34, 118.89, 118.10, 111.32, 107.98, 107.00, 104.78, 100.91, 55.52, 55.34, 51.47, 32.86, 23.16. MS (*m*/*z*, relative intensity) 390 (M^++1, 21), 389 (M^+, 100), 317(5), 289(7), 268(10), 262(37), 233(9), 204(9), 169(4), 115(1), 102(1). IR (KBr film) 3326, 2902, 1676, 1649, 1489, 1437, 1400, 1323, 1269, 1241, 1150, 939, 922, 746 cm^{-1}. Anal. Calcd for $C_{22}H_{19}N_3O_4$: C, 67.86; H, 4.92; N, 10.79. Found: C, 68.08; H, 4.63; N, 10.63.

4.7. (6*R*,12a*S*)-2,3,6,7,12,12a-Hexahydro-2-methyl-6-(3,4-methylenedioxyphenyl)-pyrazino-[2′,1′:6,1]pyrido[3,4-*b*]indole-1,4-dione 2

Compound **1** (5.00 g, 12.84 mmol) was first dissolved in dimethylsulfoxide (20 mL), isopropanol (100 mL) and 1,8-diazabicyclo[5,4,0]undec-7-ene (3.91 g, 25.68 mmol) were then added. The reaction solution was heated at reflux at 83 °C, and refluxing was continued for about 5 h. Isopropanol was removed by distillation under reduced pressure, the residue was then cooled to room temperature. A dilute HCl aqueous solution (100 mL) was gradually added while stirring and an off-white solid precipitated. The off-white solid was collected by suction and washed three times with water. After drying in a warm air, the crude product was purified by flash chromatography to give compound **2** (4.89 g, 12.56 mmol) in 98% yield, mp 295–296 °C (lit.[23] 287–289 °C for the sample containing 0.25 toluene), $[\alpha]_D^{20} = -303.1$ (*c* 1.2, $CHCl_3$) {lit.[23] $[\alpha]_D^{20} = -293.4$ (*c* 1.2, $CHCl_3$) for the sample containing 0.25 toluene}. ^{1}H NMR (DMSO-d_6) δ 11.06 (s, NH on the indole ring),

7.49 (d, $J = 7.7$ Hz, 1H), 7.31 (d, $J = 8.1$ Hz, 1H), 7.10 (dd, $J = 7.2$ Hz; 7.9 Hz, 1H), 7.01 (dd, $J = 7.7$ Hz; 7.2 Hz, 1H), 6.86 (d, $J = 8.0$ Hz, 1H), 6.82 (s, 1H), 6.75 (d, $J = 1.4$ Hz, 1H), 6.60 (dd, $J = 1.1$ Hz; 8.0 Hz, 1H), 5.99 (d, $J = 6.5$ Hz, 2H), 4.24 (d, $J = 17.6$ Hz, 1H), 4.07 (dd, $J = 4.1$ Hz; 11.8 Hz, 1H), 4.03 (d, $J = 17.7$ Hz, 1H), 3.25 (dd, $J = 4.2$ Hz; 15.4 Hz, 1H), 2.95 (dd, $J = 12.1$ Hz; 14.8 Hz, 1H), 2.84 (s, 3H). ^{13}C NMR (DMSO-d_6) δ 164.78, 162.37, 147.69, 147.33, 136.20, 132.92, 130.25, 125.94, 121.78. 118.99, 118.16, 111.36, 108.42, 108.21, 107.55, 101.32, 52.09, 50.94, 50.73, 32.69, 26.74. MS (m/z, relative intensity) 390 (M^++1, 17), 389 (M^+, 100), 360(1), 317(4), 289(7), 268(7), 262(26), 233(8), 204(8), 169(3), 115(1), 102(1). IR (KBr film) 3314, 2924, 1711, 1659, 1623, 1488, 1468, 1450, 1328, 1291, 1267, 1237, 1159, 1037, 931, 875, 789, 740, 607 cm^{-1}. Anal. Calcd for $C_{22}H_{19}N_3O_4$: C, 67.86; H, 4.92; N, 10.79. Found: C, 67.82; H, 4.83; N, 10.73.

4.8. (6*R*,12a*S*)-2,3,6,7,12,12a-Hexahydro-2-methyl-6-(3,4-methylenedioxyphenyl)-3,3,12a-trideutero-pyrazino-[2′,1′:6,1]pyrido[3,4-*b*]indole-1,4-dione 4

Compound **1** (0.50 g, 1.28 mmol), DMSO-d_6 (1.5 mL), THF (6 mL), D_2O (1 mL), and anhydrous potassium carbonate (177 mg, 1.28 mmol) were mixed together. The mixture was stirred and then heated at 65 °C. A clear solution was obtained, and the stirring was continued at this temperature for 16 h. Afterwards THF was removed by distillation under reduced pressure, the residual oil was cooled to room temperature and diluted with 25 mL of 0.5 M HCl aqueous solution. The white solid precipitated and was filtered to give crude product which was purified by flash chromatography to afford compound **4** (0.48 g, 1.22 mmol) in 95% yield. After two recrystallizations in 1,2-dichloroethane, compound **4** was characterized as follows: $[\alpha]_D^{20} = -268.0$ (c 1.2, $CHCl_3$). ^{1}H NMR ($CDCl_3$) δ 8.08 (s, NH on the indole ring), 7.53 (d, $J = 7.8$ Hz, 1H), 7.32 (d, $J = 8.0$ Hz), 7.22 (dd, $J = 7.9$ Hz; 8.1 Hz, 1H), 7.16 (dd, $J = 7.2$ Hz; 7.1 Hz, 1H), 6.97 (s, 1H), 6.80 (s, 1H), 6.70 (s, 2H), 5.92 (d, $J = 3.6$ Hz, 2H), 4.34 (dd, $J = 4.1$ Hz; 11.9 Hz, 0.16H at C-6 position), 4.13 (s, 0.06H at C-3 position), 3.98 (s, 0.06H at C-3 position), 3.54 (d, $J = 15.4$ Hz, 1H), 2.99 (s, 3H), 2.94 (d, $J = 15.6$ Hz, 1H). MS (m/z, relative intensity) 393 (M^++1, 28), 392 (M^+, 100), 391 (M^+−1, 24), 363(2), 333(1), 318(7), 290(14), 271(14), 262(73), 233(29), 204(30), 170(15), 141(3), 116(6), 102(6), 89(2), 77(1), 63(1), 46(4). IR (KBr film) 3325, 2918, 1662, 1642, 1620, 1554, 1501, 1487, 1441, 1403, 1273, 1239, 1095, 1038, 924, 872, 823, 745, 653, 606, 425 cm^{-1}. HRMS(EI) m/z calcd for $C_{22}H_{16}D_3N_3O_4$ (M^+), 392.1564; found: 392.1526.

4.9. (6*R*,12a*R*)-2,3,6,7,12,12a-Hexahydro-2-methyl-6-(3,4-methylenedioxyphenyl)-3,3,12a-trideutero-pyrazino-[2′,1′:6,1]pyrido[3,4-*b*]indole-1,4-dione 3

Compound **1** (1.50 g, 3.85 mmol), DMSO-d_6 (4.5 mL), THF (18 mL), D_2O (1.5 mL), and anhydrous potassium carbonate (532 mg, 3.85 mmol) were mixed together. The mixture was stirred and then heated to 65 °C. A clear solution was obtained, and the stirring was continued at this temperature for 6.5 h. After THF was removed by distillation under reduced pressure, the residual oil was cooled to room temperature and diluted with 45 mL of 0.5 M HCl aqueous solution. The white solid precipitated and was filtered to give crude product which was purified by flash chromatography (eluent: ethyl acetate–dichloromethane–petroleum ether = 1:2:6) to afford compound **3** (502 mg, 1.28 mmol) in 33% yield and compound **4** (990 mg, 2.52 mmol) in 65% yield. After two recrystallizations in 1,2-dichloroethane, compound **3** was characterized as follows: $[\alpha]_D^{20} = +87.1$ (c 1.0, $CHCl_3$). ^{1}H NMR ($CDCl_3$) δ 7.84 (s, 1H), 7.61 (d, $J = 7.0$ Hz, 1H), 7.27 (d, $J = 9.0$ Hz, 1H), 7.13–7.21 (m, 2H), 6.86 (dd, $J = 1.7$ Hz; 8.0 Hz, 1H), 6.73 (d, $J = 1.7$ Hz, 1H), 6.69 (d, $J = 8.0$ Hz, 1H), 6.15 (s, 1H), 5.87 (dd, $J = 1.4$ Hz; 11.6 Hz, 1H), 4.30 (dd, $J = 4.3$ Hz; 11.6 Hz, 0.16H at C-6 position), 4.09 (s, 0.10H at C-3 position), 3.93 (s, 0.10H at C-3 position), 3.78 (d, $J = 16.0$ Hz, 1H), 3.21 (d, $J = 15.9$ Hz, 1H), 3.04 (s, 3H). MS (m/z, relative intensity) 393 (M^++1, 24), 392 (M^+, 100), 391 (M^+−1, 47), 363(2), 333(1), 318(10), 306(2), 290(11), 276(6), 276(6), 271(20), 262(74), 251(2), 243(4), 233(30), 218(3), 204(2), 170(21), 141(2), 131(3), 116(6), 102(8), 89(2), 77(2), 63(2), 46(6). IR (KBr film) 3323, 2902, 1672, 1645, 1626, 1488, 1423, 1396, 1329, 1242, 1155, 1099, 1038, 938, 920, 790, 745, 694, 625, 475, 435 cm^{-1}. HRMS(EI) m/z calcd for $C_{22}H_{16}D_3N_3O_4$ (M^+), 392.1564; found: 392.1565.

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References

1. Cox, E. D.; Cook, J. M. *Chem. Rev.* **1995**, *95*, 1797.
2. Chrzanowska, M.; Rozwadowska, M. D. *Chem. Rev.* **2004**, *104*, 3341.
3. Seayad, J.; Seayad, A. M.; List, B. *J. Am. Chem. Soc.* **2006**, *128*, 1086.
4. Taylor, M. S.; Jacobsen, E. N. *J. Am. Chem. Soc.* **2004**, *126*, 10558.
5. Cesati, R. R., III; Katzenellenbogen, J. A.; Clingan, P. D.; Mills, T. J.; Price, R. A.; Pritchard, R. G. *Eur. J. Org. Chem.* **2004**, 1887.
6. Tsuji, R.; Nakagawa, M.; Nishida, A. *Tetrahedron: Asymmetry* **2003**, *14*, 177.
7. Yu, J.; Wearing, X. Z.; Cook, J. M. *Tetrahedron Lett.* **2003**, *44*, 543.
8. Bonnet, D.; Ganesan, A. *J. Comb. Chem.* **2002**, *4*, 546.
9. Dunn, P. J. *Org. Process Res. Dev.* **2005**, *9*, 88.
10. Sorbera, L. A.; Martin, L.; Leeson, P. A.; Castaner, J. *Drugs Future* **2001**, *26*, 15.
11. Daugan, A. C.-M. European Patent, EP 0740668.
12. Daugan, A.; Grondin, P.; Ruault, C.; Le Monnier de Gouville, A.-C.; Coste, H.; Kirilovsky, J.; Hyafil, F.; Labaudiniere, R. *J. Med. Chem.* **2003**, *46*, 4525.
13. Daugan, A.; Grondin, P.; Ruault, C.; Le Monnier de Gouville, A.-C.; Coste, H.; Linget, J. M.; Kirilovsky, J.; Hyafil, F.; Labaudiniere, R. *J. Med. Chem.* **2003**, *46*, 4533.

14. Orme, M. W.; Sawyer, J. C.; Schultze, L. M. World Patent WO 02/036593.
15. Revell, J. D.; Srinivasan, N.; Ganesan, A. *Synlett* **2004**, 1428.
16. Lohray, B. B.; Lohray, V. B.; Patel, S. I. World Patent WO 05/068464.
17. Montorsi, F.; McDermott, T. E. D.; Morgan, R.; Olsson, A.; Schultz, A.; Kirkeby, H. J., et al. *Urology* **1999**, *53*, 1011.
18. Stark, S.; Sachse, R.; Liedl, T.; Hensen, J.; Rohde, G.; Wensing, G.; Horstmann, R.; Schrott, K. M. *Eur. Urol.* **2001**, *40*, 181.
19. Patterson, B.; Bedding, A.; Jewell, H.; Payne, C.; Mitchell, M. *Eur. Urol. Suppl.* **2002**, *1*, 152.
20. Corbin, J. D.; Francis, S. H. *Int. J. Clin. Pract.* **2002**, *56*, 453.
21. Manallack, D. T.; Hughes, R. A.; Thompson, P. E. *J. Med. Chem.* **2005**, *48*, 3449.
22. Brock, G. *Eur. Urol. Suppl.* **2002**, *1*, 12.
23. Daugan, A. C.-M. U.S. Patent 5,859,006.
24. Orme, M. W.; Martinelli, M. J.; Doecke, C. W.; Pawlak, J. M. World Patent WO 04/011463.
25. Zhang, Y.; He, Q.; Ding, H.; Wu, X.; Xie, Y. *Org. Prep. Proced. Int.* **2005**, *37*, 99.
26. Cox, E. D.; Hamaker, L. K.; Li, J.; Yu, P.; Czerwinski, K. M.; Deng, L.; Bennett, D. W.; Cook, J. M. *J. Org. Chem.* **1997**, *62*, 44.
27. Repke, D. B.; Artis, D. R.; Nelson, J. T.; Wong, E. H. F. *J. Org. Chem.* **1994**, *59*, 2164.
28. Jiang, W.; Alford, V. C.; Qiu, Y.; Bhattacharjee, S.; John, T. W.; Haynes-Johnson, D.; Kraft, P. J.; Lundeen, S. G.; Sui, Z. *Bioorg. Med. Chem.* **2004**, *12*, 1505.
29. Allen, J.; Voges, R. *Synthesis and Applications of Isotopically Labelled Compounds*; Wiley: New York, 1994.
30. Guaran, A.; Danza, G.; Bartolucci, G.; Marrucci, D. S.; Serio, M. J. *J. Chromatgr., B* **1995**, *674*, 197.
31. Soine, W. H.; Hudson, J. E.; Shoulders, B. A.; Smith, R. V. *J. Pharm. Sci.* **1980**, *69*, 1040.
32. Khanapure, S. P.; Shi, X.-X.; Powell, W. S.; Rokach, J. *J. Org. Chem.* **1998**, *63*, 4098.