Organic & Biomolecular Chemistry

PAPER

View Article Online
View Journal | View Issue

Cite this: *Org. Biomol. Chem.*, 2014, **12**, 5669

Efficient synthesis of mibefradil analogues: an insight into *in vitro* stability†

Ji Eun Lee,‡[a,b] Tae Hui Kwon,‡[a,c] Su Jin Gu,[a] Duck-Hyung Lee,[b] B. Moon Kim,[c] Jae Yeol Lee,[d] Jae Kyun Lee,[a] Seon Hee Seo,[a] Ae Nim Pae,[a,e] Gyochang Keum,[a,e] Yong Seo Cho*[a,e] and Sun-Joon Min*[a,e]

Received 6th March 2014,
Accepted 6th June 2014

DOI: 10.1039/c4ob00504j

www.rsc.org/obc

This article describes the synthesis and biological evaluation of a chemical library of mibefradil analogues to investigate the effect of structural modification on *in vitro* stability. The construction of the dihydrobenzopyran structure in mibefradil derivatives **2** was achieved through two efficient approaches based on a diastereoselective intermolecular Reformatsky reaction and an intramolecular carbonyl–ene cyclization. In particular, the second strategy through the intramolecular carbonyl–ene reaction led to the formation of a key intermediate **3** in a short and highly stereoselective way, which has allowed for practical and convenient preparation of analogues **2**. Using this protocol, we could obtain 22 new mibefradil analogues **2**, which were biologically tested for *in vitro* efficacies against T-type calcium channels and metabolic stabilities. Among the synthesized compounds, we found that analogue **2aa** containing a dihydrobenzopyran ring and a secondary amine linker showed high % remaining activities of the tested CYP enzymes retaining the excellent T-type calcium channel blocking activity. These findings indicated that the structural modification of **1** was effective for improving *in vitro* stability, *i.e.*, reducing CYP inhibition and metabolic degradation.

Introduction

Mibefradil **1** (Posicor, Roche®), the first selective T-type calcium channel blocker, was approved by the FDA in 1997 for the treatment of hypertension and chronic angina pectoris (Fig. 1).[1–3] Because of its high oral bioavailability and long half-life,[4] it was initially administered to patients as a suitable as well as convenient once-a-day medication. However, despite the excellent pharmacokinetic features, this drug was withdrawn from the market due to unfavorable cardiovascular side effects, which were derived from a drug–drug interaction likely resulting from an L-type calcium channel blockade.[5]

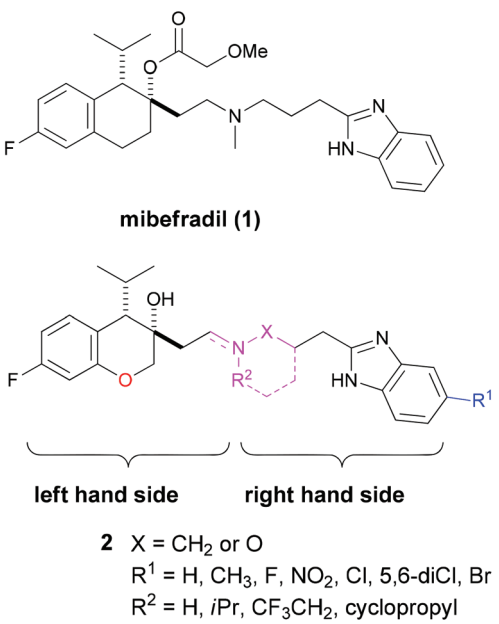


Fig. 1 Structures of mibefradil **1** and new analogues **2**.

A metabolic examination of mibefradil **1** in mammalian microsomes revealed that most metabolites are formed by a combination of enzymatic processes, such as cytochrome P450

[a]*Center for Neuro-Medicine, Brain Science Institute, Korea Institute of Science and Technology (KIST), Hwarangno 14-gil 5, Seongbuk-gu, Seoul 136-791, South Korea*
[b]*Department of Chemistry, Sogang University, Seoul, 121-742, South Korea*
[c]*Department of Chemistry, College of Natural Sciences, Seoul National University, Seoul 151-747, South Korea*
[d]*Department of Chemistry, Kyung Hee University, 26 Kyungheedae-ro, Dongdaemun-gu, Seoul, 130-701, South Korea*
[e]*Department of Biological Chemistry, Korea University of Science and Technology (UST), 176 Gajung-dong, 217 Gajungro, Yuseong-gu, Daejeon, 305-333, South Korea. E-mail: ys4049@kist.re.kr, sjmin@kist.re.kr*

†Electronic supplementary information (ESI) available. CCDC 980275. For ESI and crystallographic data in CIF or other electronic format see DOI: 10.1039/c4ob00504j

‡These authors equally contributed to this work.

(CYP450)-mediated oxidation at saturated and unsaturated carbons, CYP450-catalyzed dealkylation, and hydrolysis of the ester side-chain.[6] In particular, one of the most important metabolic processes is oxidation at the benzylic carbon of the tetrahydronaphthalene ring. Based on this study, we speculated that such a biotransformation might occur in CYP enzymes to generate some reactive metabolites, which, prior to release from the active site, could cause CYP inactivation.[7–9]

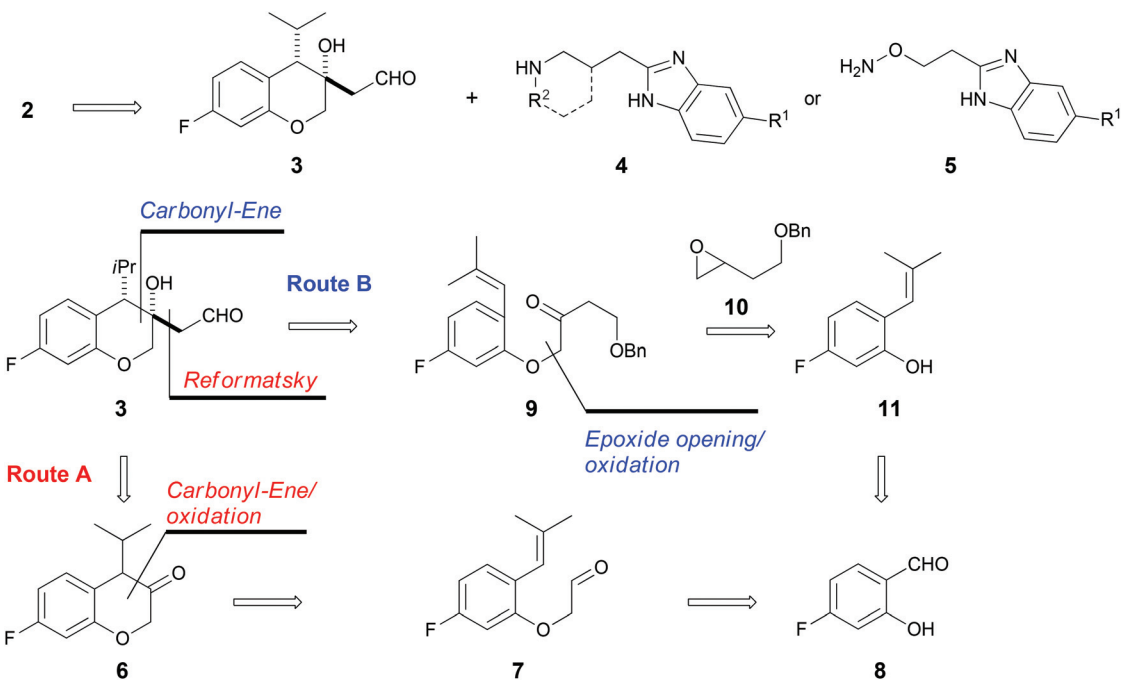
In order to reduce CYP inhibition without affecting channel blocking activity, we considered modifying the structure of mibefradil **1**. Several strategies can be used to possibly minimize the impact of CYP inhibition, such as changing the length of the chain linker, replacing the aromatic groups, or removing the labile functional groups.[10] Considering these methods, we have designed new mibefradil analogues **2** as shown in Fig. 1. First, we imagined that addition of oxygen to the site of hydroxylation could improve the stability of **1** by preventing further metabolic degradation. Second, alteration of the tertiary amine moiety to other preventive linkers such as bulkier tertiary amines or oximes would provide a reasonable approach to avoid the metabolic dealkylation process. In addition, the phenolic oxidation on the benzimidazole unit could be inhibited by introduction of R^1 substituents at the C5 position of benzimidazole. Thus, we anticipated that these structural changes could in turn diminish the formation of metabolic CYP inhibitors.

To investigate the correlation between structural modification and CYP inhibition, it is necessary to develop a practical synthetic method to obtain a focused library of mibefradil derivatives. In particular, an efficient and stereoselective synthesis of a dihydrobenzopyran ring system on the left side of compound **2** would be a crucial element for the preparation of the chemical library because there are two consecutive stereogenic centers in **2**, of which one is quaternary. Once a viable synthesis of the left hand side of **2** is established, we were assured that a set of the mibefradil analogues could be rapidly developed in a convergent way by combining it with various substituted benzimidazoles.

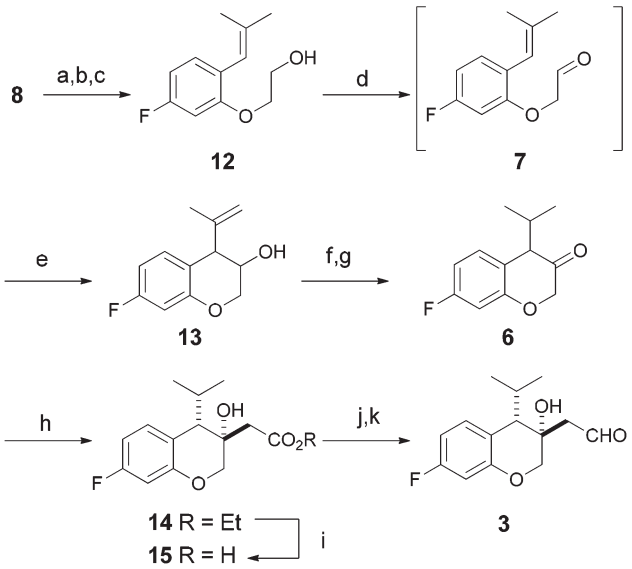
Herein, we report an efficient and stereoselective synthesis of mibefradil derivatives **2** and evaluation of their inhibitory activity against the CYP enzymes as well as their calcium channel blocking activities.

Results and discussion

Our synthetic plan to achieve diastereoselective synthesis of **2** is shown in Scheme 1. Coupling reactions of two fragments **3** and **4** or **5** through reductive amination or oxime formation would be performed at a late stage to afford our target molecules **2** (Scheme 2). We designed two synthetic approaches to construct the important structural framework of dihydrobenzopyran **3**. In route A, a metal-mediated Reformatsky reaction[11–14] would be the key strategy for installation of the quaternary center in the functionalized dihydrobenzopyran **3**. In this event, we proposed that the isopropyl group would thereby serve as a steric obstacle to direct stereoselective addition of an acetate unit to ketone **6**. The required chroman-3-one **6** would then be synthesized through an intramolecular carbonyl–ene reaction[15–19] of **7**, followed by oxidation. Alternatively, an intramolecular carbonyl–ene reaction of **9** in the second approach would result in ring formation and diastereo-



Scheme 1 Our synthetic plan to obtain mibefradil derivatives **2**.



Scheme 2 The first synthesis of aldehyde **3**: (a) K_2CO_3, $BrCH_2CH_2OTBS$, DMF, 80 °C, 64%; (b) $Ph_3PCH(CH_3)_2I$, *n*BuLi, THF; (c) TBAF, THF, 83% (2 steps); (d) SO_3·Pyr, iPr_2NEt, DMSO, CH_2Cl_2; (e) Me_2AlCl, CH_2Cl_2, −20 °C, 71% (2 steps); (f) 20% Pd/C, H_2, MeOH, 90%; (g) PCC, CH_2Cl_2, 81%; (h) ICH_2CO_2Et, Et_2Zn, O_2, Et_2O, rt, 72%; (i) LiOH, MeOH–H_2O (3 : 1), quant.; (j) $LiAlH_4$, THF, 0 °C, 96%; (k) Dess–Martin periodinane, CH_2Cl_2, 81%.

selection of **3** simultaneously, which might be of benefit to reduce the number of reaction steps. Ketone **9** would be prepared from nucleophilic addition of **11** to epoxide **10** and subsequent oxidation. Both precursors **7** and **11** would be readily accessible starting from 3-fluorosalicylic aldehyde **8**.

Synthesis of aldehyde 3

The synthesis of compound **3** *via* the first route is demonstrated in Scheme 2. 3-Fluorosalicylic aldehyde[20–22] **8** was treated with bromoethyl silyl ether[23] and K_2CO_3 in DMF to give *O*-alkylated product, which underwent the Wittig olefination with isopropylidene ylide and the silyl deprotection to afford the resulting primary alcohol **12** in good yields. The Swern oxidation of **12** was attempted to obtain aldehyde **7**, but it was susceptible to decomposition during flash column chromatography. To avoid such decomposition, we decided to conduct an oxidation reaction under mild conditions, followed by rapid cyclization without further purification. After screening several oxidation conditions, we discovered that the Parikh–Doering oxidation[24] of **12** gave the crude aldehyde **7** in a considerably high yield. Thus, the subsequent carbonyl–ene reaction immediately proceeded in the presence of dimethylaluminum chloride, which allowed for conversion of **7** to an inseparable 1 : 1.5 mixture of dihydrobenzopyranols **13**. Despite the low selectivity, it is noteworthy that the desired chromanol ring was successfully constructed through this ene reaction without the formation of any side products caused by a Friedel–Craft type acylation. Hydrogenation of alkene **13** followed by PCC oxidation produced the chroman-3-one derivative **6** in excellent yields.

With the ketone **6** in hand, we next investigated metal-induced Reformatsky reactions to synthesize the β-hydroxy ester **14**. In general, it is difficult to add carbon nucleophiles to ketones in the Reformatsky reaction due to their low reactivity. Initially, we attempted a typical Reformatsky reaction of ketone **6** with bromoacetate using zinc as a metal source, but we were not able to obtain any desired product. Instead, the indium-mediated reaction proceeded to give the desired β-hydroxy ester **14** exclusively in moderate yield.[25] The structure of ester **15a** was preliminarily elucidated by NOE experiments. In fact, the NOE correlation between the isopropyl group and the axial C2 proton indicated that the stereochemical relationship of the isopropyl group and the hydroxyl group was *cis*. However, the formation of **14** under these reaction conditions was not consistently reproducible on a large scale. While exploring other reaction conditions, we found that slow addition of ketone **6** to the solution of Et_2Zn and iodoacetate in Et_2O at ambient temperature[26,27] afforded the corresponding ester **14** as a single diastereomer in 72% yield. Thus, by following this protocol, we could efficiently obtain ester **14** on a large scale. The base-catalyzed hydrolysis of ester **14** yielded acid **15**, the structure of which was confirmed by X-ray crystallographic analysis (Fig. 2).[28] Finally, ester **14** was easily transformed to aldehyde **3** by sequential reduction and oxidation.

Alternative synthesis of aldehyde 3

In light of the efficiency for obtaining aldehyde **3** in the first approach, we started to explore a different approach to aldehyde **3** that would be amenable to reasonable scale-up as we proposed in Scheme 1. The second synthesis of aldehyde **3** is described in Scheme 3. Alkenylphenol **11** was first prepared in two steps and in 80% overall yields according to the modified literature procedure.[29] We attempted the reactions of **11** with epoxide **10**[30,31] to obtain the corresponding alcohol under different reaction conditions by varying the bases, solvents and temperature. Indeed, the best yield was achieved when we performed the reaction with two equivalents of epoxide **10** in CH_3CN at 90 °C using Cs_2CO_3 as a base. The ensuing secondary alcohol was then converted to the desired ketone **9** in 81%

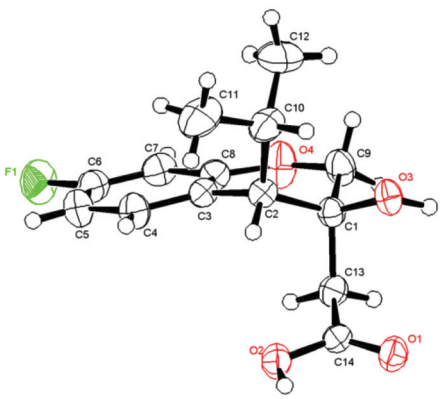
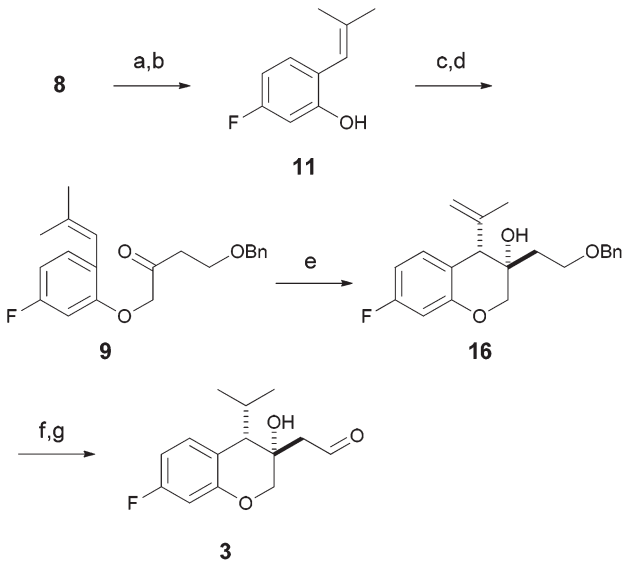


Fig. 2 X-ray crystal structure of the acid **15**.

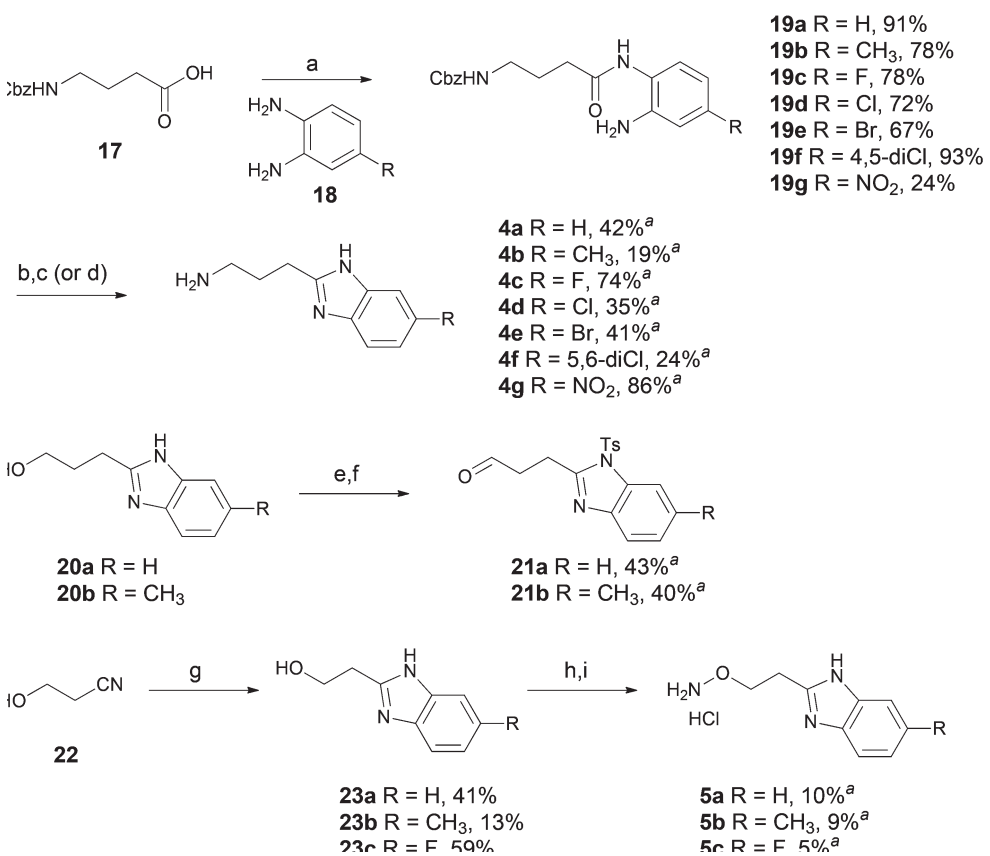


Scheme 3 The second approach to aldehyde **3**: (a) iPrMgBr, Et_2O, 86%; (b) μW (120W), hexane, 93%; (c) **10**, Cs_2CO_3, CH_3CN, 90 °C, 52%; (d) Dess–Martin periodinane, CH_2Cl_2, 94%; (e) $SnCl_4$, CH_2Cl_2, −78 °C to −40 °C, 53% (dr = 10 : 1); (f) H_2, Pd/C, MeOH, 86%; (g) Dess–Martin periodinane, CH_2Cl_2, 81%.

yield by DMP oxidation. The stereoselective intramolecular ene cyclization of **9** proved to be a significant challenge because there are few examples of intramolecular carbonyl–ene reactions using isolated ketones as the substrates for cyclization.[32,33] A preliminary evaluation of the carbonyl–ene reaction of **9** using $TiCl_4$ led to the formation of **16**; however, low yield and low selectivity were observed. After performing reaction optimization, we obtained the desired alcohol **16** with a high diastereoselectivity when a solution of **9** in CH_2Cl_2 was treated with $SnCl_4$ at −78 to −40 °C for 5.5 h. At last, concurrent debenzylation and hydrogenation followed by DMP oxidation proceeded to afford aldehyde **3**, which was identical to the compound synthesized from the first approach. In comparison to route A, this approach to **9** involved only 7 steps and provided the target aldehyde **3** in 14% overall yield. Thus, material for the construction of the mibefradil library was generally produced *via* this second route.

Synthesis of benzimidazole fragments

In order to use benzimidazoles containing various substituents for assembly of two fragments as we planned in Scheme 1, we prepared a set of amines **4**, aldehydes **21** and

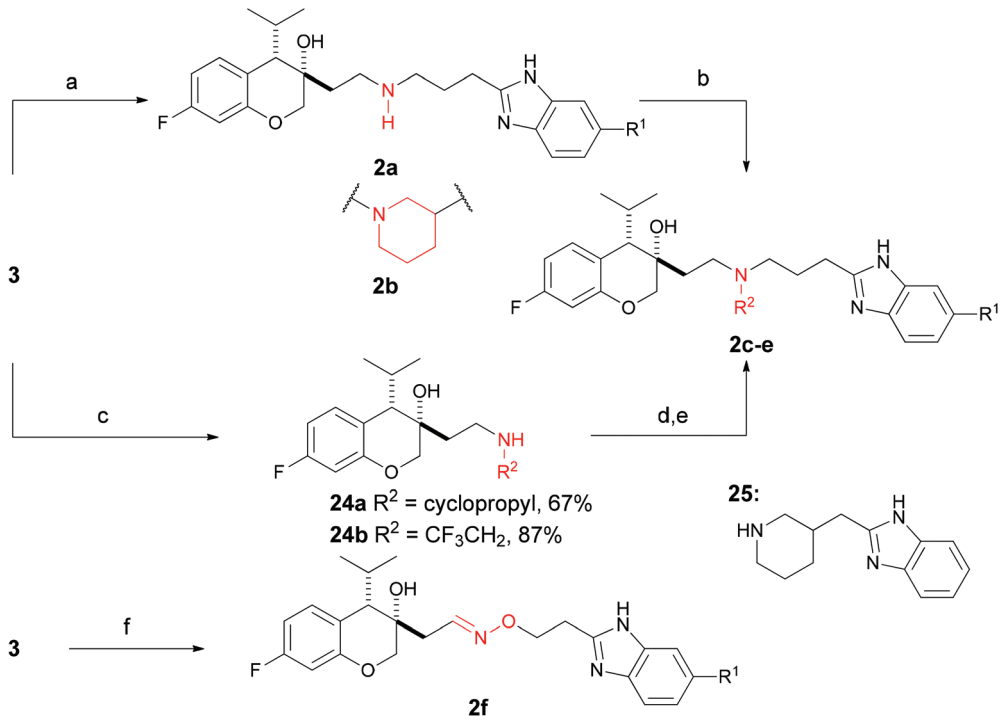


Scheme 4 Synthesis of benzimidazole fragments **4**, **21** and **5**: (a) $ClCO_2iBu$, Et_3N, diamines **18**, THF; (b) *p*TsOH·H_2O, toluene, Deans–Stark; (c) H_2, 10% Pd/C, MeOH (for **4a–d** and **4f**); (d) 6 N aq. HCl, 110 °C (for **4e** and **4g**); (e) *p*TsCl, DMAP, Et_3N, CH_2Cl_2–THF (1 : 1); (f) Dess–Martin periodinane, CH_2Cl_2; (g) diamines **18**, conc. HCl, reflux; (h) *N*-hydroxyphthalimide, DIAD, PPh_3, THF; (i) H_2NNH_2, EtOH, reflux, then 1 M HCl in Et_2O. [a]Isolated yields over two steps.

O-alkylated hydroxyl amines **5** as shown in Scheme 4. *N*-CbZ protected amino acid **17**, prepared from the treatment of γ-aminobutyric acid with CbzCl and 4 N NaOH,[34] was transformed to amides **19** *via* mixed anhydride intermediates. Condensation of **19** with azeotrophic removal of water produced the corresponding benzimidazoles, which was subjected to Cbz deprotection by either hydrogenolysis or acid-catalyzed hydrolysis to afford free amines **4**. Considering a role reversal of two fragments for coupling reactions (see below), we synthesized aldehydes **21** as alternative components by conducting selective tosylation of readily available benzimdazoles **20**[35] and consecutive DMP oxidation. At this time, we should point out that all attempted direct oxidations of **20** using different reagents such as SO_3·pyr, PCC, DMP, TPAP/NMO or Swern failed without blocking nitrogen of the benzimidazole moiety in **20**. On the other hand, *O*-alkylated hydroxyl amines **5** were also prepared in three steps: the elaboration of 3-hydroxypropionitrile **22** to hydroxyethyl benzimidazoles **23** through condensation with phenylenediamines **18** under acidic conditions, Mitsunobu reaction and hydrazinolysis followed by *in situ* salt formation. Although the yields of several reactions were relatively low, the whole process provided reasonable amounts of materials for the next steps.

Complete synthesis of mibefradil analogues

The complete synthesis of mibefradil derivatives **2** is illustrated in Scheme 5. Reductive amination of **3** with amines **4** and **25** produced the first series of analogues **2a** and **2b**. The second series of compounds **2c** (R^2 = iPr) were obtained by subsequent reductive amination of **2a** with acetone in the presence of acetic acid. However, conversion of **2a** to tertiary amines **2d** (R^2 = cyclopropyl) or **2e** (R^2 = CF_3CH_2) using reductive amination, metal-catalyzed coupling reactions or direct *N*-alkylations[36,37] proved to be problematic. As indicated in the previous section, aldehydes **21** were regarded as surrogate fragments for coupling reactions. Thus, reductive aminations of **3** with cyclopropylamine and trifluoroethylamine gave secondary amines **24**, which could be transformed into the desired **2d** and **2e** by second reductive amination with aldehydes **21** and subsequent removal of the tosyl group.[38] Towards the last analogues of mibefradil, condensation reactions of aldehydes **3**



R^1 =	H	CH_3	F	Cl	Br	5,6-diCl	NO_2
2a	**2aa**, 91%	**2ab**, 31%	**2ac**, 26%	**2ad**, 26%	**2ae**, 14%	**2af**, 46%	**2ag**, 24%
2b	**2ba**, 47%						
2c (R^2 = *i*Pr)	**2ca**, 47%	**2cb**, 49%	**2cc**, 25%	**2cd**, 14%	**2ce**, 29%	**2cf**, 47%	**2cg**, 39%
2d (R^2 = cyclopropyl)	**2da**, 36%[a]	**2db**, 57%[a]					
2e (R^2 = CF_3CH_2)	**2ea**, 58%[a]						
2f	**2fa**, 47%	**2fb**, 49%	**2fc**, 25%	**2fd**, 14%			

Scheme 5 Reagents and conditions: (a) **4** or **25**, $NaBH_3CN$, AcOH, MeOH; (b) acetone, $NaBH_3CN$, AcOH, MeOH (from **2a** only); (c) R^2NH_2, AcOH, $NaBH_3CN$, MeOH; (d) **21**, AcOH, $NaBH_3CN$, MeOH; (e) TBAF, THF; (f) **5**, K_2CO_3, MeOH. [a]Isolated yields over two steps.

with **5** in the presence of K_2CO_3 provided a series of oximes **2f** in moderate yields.

Channel blocking activity and *in vitro* stability

In vitro screening of mibefradil analogues **2** was performed with HEK293 cells which stably expressed T-type calcium channel α_{1G} and α_{1H} subtypes using an FDSS6000 instrument.[39] As summarized in Table 1, 19 and 18 compounds out of the total 22 new analogues respectively displayed higher than 50% inhibitory activities against each T-type calcium channel (α_{1G} and α_{1H}) at 10 μM, which indicated that the overall structural modification of mibefradil **1** in this study did not significantly reduce the T-type calcium channel blockade as we expected. In particular, the structural change of the tertiary amine moiety to other functional groups such as a secondary amine (**2a**), a larger tertiary amine (**2b–e**), or oxime (**2f**) exerted a great influence on the channel blocking activities, whereas the installation of various substituents to the benzimidazole moiety in **2** did not induce any critical change of inhibitory activities against T-type calcium channels.

After we confirmed the *in vitro* potencies of our compounds **2**, we evaluated their inhibitory activities against five CYP450 isozymes including CYP2D6 and CYP3A4, two of the most abundant CYP enzymes in human liver. As shown in Table 2, we measured the remaining activity of the CYP enzymes after treatment with the selected compounds **2** using CYP450 screening kits.[40] In general, the blocking activities of several compounds against the CYP enzymes were considerably improved in comparison with those of mibefradil **1**. In particular, compound **2aa** containing dihydrobenzopyran and a secondary amine linker exhibited significantly increased CYP remaining activities. Although the inhibitory values of the selected **2c** (R^2 = iPr) against CYP3A4 isozyme were highly reduced relative to that of mibefradil **1**, we could not observe any enhancement of those activities against CYP2D6. In contrast, the CYP inhibitory activities of the selected oximes **2f** were increased over that of **1**, which might be a consequence of the hydrolysis of oxime due to its liability to acid, but this is not clearly conclusive at this point. The overall result implied that our approach to structural modification of mibefradil **1** was significantly effective in reducing its CYP inactivation.

Table 1 *In vitro* T-type calcium channel blocking activities[a] of mibefradil analogues **2**

Compds	α1G (10 μM)	α1H (10 μM)	Compds	α1G (10 μM)	α1H (10 μM)
2aa	71.20	66.55	**2ce**	67.42	65.61
2ab	66.04	47.90	**2cf**	53.67	62.28
2ac	65.19	60.54	**2cg**	64.35	69.56
2ad	60.02	55.32	**2da**	46.85	44.56
2ae	62.59	56.20	**2db**	47.56	44.16
2af	59.76	58.37	**2ea**	11.76	38.10
2ag	59.69	66.73	**2fa**	53.29	55.33
2ba	68.37	70.76	**2fb**	59.28	52.70
2ca	64.57	59.16	**2fc**	58.65	54.53
2cb	63.63	54.58	**2fd**	60.37	52.11
2cc	61.74	64.41	**1**	79.19	80.21
2cd	63.49	59.10			

[a] Ca^{2+} flux assay; % Inhibition value was obtained at 10 μM (see the detail assay procedure in the Experimental section).

Table 2 CYP inhibition[a] and microsomal stability[b] of the selected compounds **2**

	% CYP isozyme remaining activity[c]					
Compds	1A2	2D6	2C9	3A4	2C19	HLM[d] % Remaining @ 30 min
2aa	59.46	84.25	109.58	39.91	55.00	75.32
2ab	16.01	2.75	33.48	25.71	19.94	37.23
2ac	59.86	3.27	85.69	26.20	40.46	83.26
2ad	19.43	1.86	5.91	42.97	5.52	—
2ae	43.07	3.95	21.15	54.99	17.44	—
2af	26.36	1.81	14.69	47.45	4.57	—
2ag	58.75	5.15	51.54	121.08	28.64	—
2ba	49.06	8.68	90.38	31.65	44.26	12.89
2ca	77.56	10.56	110.18	46.94	56.71	—
2cc	111.44	7.79	55.57	45.55	49.66	5.72
2da	56.02	4.73	26.39	19.66	9.45	—
2db	56.71	8.07	54.52	12.05	12.76	1.19
2fa	52.27	9.54	11.45	30.26	9.92	—
2fb	19.42	5.61	5.72	10.86	11.22	0.11
2fc	39.74	8.07	8.09	10.81	8.75	0.38
2fd	16.84	3.38	3.05	8.52	2.93	0.30
1	99.00	3.79	24.70	4.82	39.70	56.66

[a] Each data represent the mean of triplicate experiments. [b] Each data represent the mean of duplicated experiments. [c] The remaining activity of each isozyme was obtained at a concentration of 10 μM using fluorogenic Vivid® CYP450 screening kits. [d] HLM, human liver microsomes.

In addition to the CYP inhibition experiments, we examined the metabolic stability of the representative compounds **2** of each series in human hepatic microsome. The result showed that the metabolic stabilities of the tested compounds **2** were also dependent on the structural modification of the tertiary amine moiety of **1** (Table 2). In fact, compound **2aa** turned out to be highly stable, remaining up to 75% at 30 min after treatment of **2aa** in pooled human hepatic microsomes, while other compounds having piperidine (**2ba**), isopropyl amine (**2cc**), cyclopropyl amine (**2db**), and oxime (**2fc**) appeared to be rapidly decomposed. Given the comprehensive data of **2aa** with regard to calcium channel blocking activities, CYP inhibition, and metabolic stability, we therefore concluded that the structural revision of **1** into **2aa** by insertion of oxygen at the benzylic position of the tetrahydronaphthalene ring and replacement of the tertiary amine by a secondary amine could effectively improve the metabolic stability of mibefradil **1** in both the CYP enzymes and the hepatic microsomes, maintaining the *in vitro* efficacy against T-type calcium channels.

Conclusion

In this article, we have established a synthesis of the chemical library of mibefradil analogues **2** which is based on the construction of the dihydrobenzopyran structure by means of the

diastereoselective intermolecular Reformatsky reaction and the intramolecular carbonyl–ene cyclization. These protocols provided efficient access to the fragment **3** at the left hand side of **2**, which served as the immediate precursor for coupling reactions with amines or aldehydes. In particular, the second strategy through the carbonyl–ene reaction led to the formation of aldehyde **3** within only 7 steps in a highly stereoselective manner, which has allowed for practical and convenient preparation of mibefradil analogues **2**. Accordingly, the synthesis of **2** has been successfully achieved by simple or dual reductive amination strategies using aldehyde **3** as a switchable substrate.

A total of 22 mibefradil derivatives were synthesized and evaluated for *in vitro* activities. Most compounds **2** were confirmed to be potent T-type calcium channel blockers comparable to mibefradil **1**. Moreover, *in vitro* stabilities of the selected compounds **2** in the CYP enzymes and the human hepatic microsomes were also significantly improved. In particular, we found that analogue **2aa**, which has a dihydrobenzopyran ring and a secondary amine linker, showed high % remaining activities of the tested CYP enzymes, retaining the high inhibitory activity against T-type calcium channels. These findings highlight the importance of the structural modification of **1** for reducing CYP inhibition and metabolic degradation.

Experimental section

General

All reactions were carried out under dry nitrogen unless otherwise indicated. Commercially available reagents were used without further purification. Solvents and gases were dried according to standard procedures. Organic solvents were evaporated with reduced pressure using a rotary evaporator. Analytical thin layer chromatography (TLC) was performed using glass plates precoated with silica gel (0.25 mm). TLC plates were visualized by exposure to UV light (UV), and then visualized with a *p*-anisaldehyde stain followed by brief heating on a hot plate. Flash column chromatography was performed using silica gel 60 (230–400 mesh, Merck) with the indicated solvents. ^{1}H and ^{13}C spectra were recorded on Bruker 300, Bruker 400 or Varian 300 NMR spectrometers. ^{1}H NMR spectra are represented as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet), integration, and coupling constant (*J*) in hertz (Hz). ^{1}H NMR chemical shifts are reported relative to $CDCl_3$ (7.26 ppm). ^{13}C NMR was recorded relative to the central line of $CDCl_3$ (77.0 ppm). HPLC data were acquired from a Waters Alliance System with a UV detector set to 254 and 280 nm. Samples were injected (10 μL) onto a Waters Sunfire 4.6 × 150 mm, 5.0 μM, C18 column maintained at 25.8 °C. A linear gradient from 30% to 100% **B** (MeCN) in 20 min was followed by pumping 100% **B** for another 10 minutes with **A** being H_2O + 0.1 M NH_4OAc (or NH_4HCO_2). The flow rate was 1.0 mL min^{-1}.

2-(5-Fluoro-2-(2-methylprop-1-enyl)phenoxy)ethanol (12). A solution of aldehyde **8** (7.74 g, 55.2 mmol) and K_2CO_3 (7.63 g, 55.2 mmol) in anhydrous DMF (60 mL) was heated at 80 °C for 30 min. 2-Bromoethoxy *tert*-butyldimethylsilane (16.94 g, 70.8 mmol) was added and the reaction mixture was stirred at the same temperature for an additional 3 h. The solvent was removed under reduced pressure to give the crude material, which was dissolved in ethyl acetate (100 mL). The resulting solution was treated with aqueous NH_4Cl (300 mL) and was extracted with ethyl acetate (3 × 200 mL). The combined organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The crude residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 20) to afford the corresponding ether (10.00 g, 64%) as a colorless oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 10.40 (s, 1H), 7.84–7.85 (m, 1H), 6.70–6.74 (m, 2H), 4.14 (t, J = 4.6 Hz, 2H), 4.01 (t, J = 4.6 Hz, 2H), 0.88 (s, 9H), 0.07 (s, 6H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 188.2, 167.6 (d, J = 254.4 Hz), 163.2 (d, J = 11.0 Hz), 130.6 (d, J = 11.5 Hz), 121.9, 108.3 (d, J = 22.1 Hz), 100.9 (d, J = 25.5 Hz), 70.5, 61.6, 25.8, 18.29, −5.4. HRMS-CI (*m*/*z*): $[M + H]^+$ calcd for $C_{15}H_{24}FO_3Si$ 299.1479, found 299.1479.

To a solution of isopropyltriphenylphosphonium iodide (19.7 g, 45.6 mmol) in THF (80 mL) was slowly added a solution of *n*-BuLi (31.7 mL, a 1.6 M solution in *n*-hexane, 50.6 mmol) at 0 °C. After the reaction was stirred for 30 min, a solution of the above aldehyde (7.56 g, 25.3 mmol) in THF (20 mL) was added. The reaction mixture was stirred for 2 h at room temperature and quenched with NH_4Cl (150 mL). The resulting solution was extracted with ethyl acetate (3 × 150 mL), washed with brine, and dried over anhydrous $MgSO_4$. The solvent was removed under reduced pressure to give the crude alkene, which was used for the next step without further purification; ^{1}H-NMR (300 MHz, $CDCl_3$ δ 7.09–7.15 (m, 1H), 6.60–6.65 (m, 2H), 6.26 (s, 1H), 4.00 (m, 4H), 1.91 (s, 3H), 1.79 (s, 3H), 0.93 (s, 9H), 0.12 (s, 6H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 162.3 (d, J = 324.5 Hz), 157.7 (d, J = 127.0 Hz), 135.2, 131.4 (d, J = 127.0 Hz), 123.9 (d, J = 47.0 Hz), 120.3, 106.5 (d, J = 27.4 Hz), 100.0 (d, J = 34.0 Hz), 70.1, 62.2, 26.8, 26.2, 19.7, 18.7, −5.1. HRMS-EI (*m*/*z*): $[M]^+$ calcd for $C_{18}H_{29}FO_2Si$ 324.1921, found 324.1926.

To a solution of the above alkene (8.22 g, 25.3 mmol) in distilled THF (60 mL) was added a solution of tetrabutylammonium fluoride (30.6 mL, 1.0 M solution in THF, 30.6 mmol) at 0 °C. After stirring for 2 h, the reaction mixture was quenched with NH_4Cl (200 mL). The solvent was partially removed under reduced pressure. The resulting mixture was extracted with CH_2Cl_2 (3 × 150 mL). The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 4) to afford alcohol **12** (4.44 g, 83%, 2 steps) as a pale yellow oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.10–7.13 (m, 1H), 6.58–6.67 (m, 2H), 6.21 (s, 1H) 4.04 (t, J = 3.7 Hz, 2H), 3.95 (t, J = 3.7 Hz, 2H), 2.24 (s, 1H), 1.91 (s, 3H), 1.77 (s, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.1 (d, J = 243.5 Hz), 156.9 (d, J = 9.3 Hz), 135.8, 131.1 (d, J = 9.3 Hz), 123.787, 119.5, 107.0 (d, J = 20.8 Hz), 100.2 (d, J = 25.2 Hz), 70.0, 61.3, 26.5, 19.5. HRMS-EI (*m*/*z*): $[M]^+$ calcd for $C_{12}H_{15}FO_2$ 210.1056, found 210.1054.

2-(5-Fluoro-2-(2-methylprop-1-enyl)phenoxy)acetaldehyde (7). To a solution of the alcohol **12** (628 mg, 2.99 mmol) in dry CH_2Cl_2 (15 mL) were sequentially added DMSO (4 mL), iPr_2NEt (8 mL) and SO_3·pyridine (2.38 g, 14.9 mmol) at 0 °C. The reaction mixture was allowed to stir at the same temperature for 1 h. It was the quenched with NH_4Cl (10 mL), acidified with 3 N HCl until pH 2 was reached, and finally extracted with diethyl ether (3 × 45 mL) and washed with brine. The organic layers were dried over anhydrous $MgSO_4$ and concentrated *in vacuo* to afford the crude aldehyde **7**, which was used for the next step without purification; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.83 (s, 1H), 7.13–7.16 (m, 1H), 6.66–6.70 (m, 1H), 6.43–6.46 (m, 1H), 6.26 (s, 1H), 4.52 (s, 2H), 1.91 (s, 3H), 1.77 (s, 3H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 199.2, 162.2 (d, J = 326.5 Hz), 156.2, 136.6, 131.8 (d, J = 12.0 Hz), 124.2, 119.5, 108.1 (d, J = 28.0 Hz), 100.5 (d, J = 34.1 Hz), 73.4, 26.8, 19.8. HRMS-EI (m/z): $[M]^+$ calcd for $C_{12}H_{13}FO_2$ 208.0899, found 208.0898.

7-Fluoro-4-(prop-1-en-2-yl)chroman-3-ol (13). To a solution of the above aldehyde **7** (622 mg, 2.99 mmol) in dry CH_2Cl_2 (15 mL) cooled to −20 °C was added a solution of Me_2AlCl (2.99 mL, 1.0 M in *n*-hexane). The reaction mixture was stirred at the same temperature for 30 min, quenched with $NaHCO_3$ (20 mL) at 0 °C and filtered through a pad of Celite®. The filtrate was extracted with CH_2Cl_2 (3 × 30 mL). The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 5) to afford an inseparable mixture of alcohols **13** (440 mg, 71%, 2 steps, dr = 1.5 : 1) as a pale yellow oil; *major isomer*: ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.01–7.06 (m, 1H), 6.55–6.63 (m, 2H), 5.22 (d, J = 5.0 Hz, 1H), 4.83 (d, J = 0.7 Hz, 1H), 4.03–4.28 (m, 3H), 3.70 (d, J = 4.3 Hz, 1H), 1.87 (s, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.1 (d, J = 243.0 Hz), 155.3 (d, J = 12.2 Hz), 144.4, 131.6 (d, J = 9.5 Hz), 117.2, 116.5, 108.2 (d, J = 21.4 Hz), 103.6 (d, J = 21.5 Hz), 68.5, 63.7, 47.7, 23.0. HRMS-EI (m/z): $[M]^+$ calcd for $C_{12}H_{13}FO_2$ 208.0899, found 208.0898; *minor isomer*: ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.93–6.98 (m, 1H), 6.54–6.65 (m, 2H), 5.12 (d, J = 13.2 Hz, 1H), 4.79 (d, 0.7 Hz, 1H), 3.90–4.25 (m, 3H), 3.41 (d, J = 6.1 Hz, 1H), 1.70 (s, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.3 (d, J = 243.2 Hz), 155.0 (d, J = 11.8 Hz), 144.5, 131.2 (d, J = 9.6 Hz), 117.6, 116.9, 108.5 (d, J = 21.5 Hz), 103.6 (d, J = 21.9 Hz), 67.7, 64.8, 52.3, 19.3.

7-Fluoro-4-isopropylchroman-3-one (6). To a solution of alkene **13** (730 mg, 3.54 mmol) in MeOH (10 mL) was added Pd/C (146 mg, 10% (w/w)) at room temperature. The reaction mixture was stirred under a hydrogen atmosphere (with the aid of a hydrogen balloon) for 3 h. It was filtered through a pad of Celite® and the solvent was removed under reduced pressure to afford isopropylchroman-3-ol (663 mg, 90%) as a pale yellow liquid; *major isomer*: ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.13–7.16 (m, 1H), 6.56–6.68 (m, 2H), 4.31–4.32 (m, 1H), 4.22 (dd, J = 4.2, 11.2 Hz, 1H), 4.09 (dt, J = 1.0, 11.2 Hz, 1H), 2.82 (m, 1H), 2.41–2.46 (m, 1H), 1.16 (d, J = 6.8 Hz, 3H), 1.05 (d, J = 7.0 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.0 (d, J = 242.7 Hz), 155.1 (d, J = 11.7 Hz), 130.0 (d, J = 9.5 Hz), 118.3, 108.1 (d, J = 21.2 Hz), 103.8 (d, J = 24.1 Hz), 71.0, 64.8, 44.2, 27.7, 21.0, 19.4. HRMS-EI (m/z): $[M]^+$ calcd for $[C_{12}H_{15}FO_2]^+$ 210.1056, found, 210.1055; *minor isomer*: ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.98–7.03 (m, 1H), 6.53–6.63 (m, 2H), 4.12–4.14 (m, 3H), 3.93–4.02 (m, 1H), 2.43 (d, J = 7.8 Hz, 1H), 2.10 (s, 1H), 1.63–1.70 (m, 1H), 0.96 (d, J = 6.6 Hz, 6H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 162.5 (d, J = 242.3 Hz), 154.5 (d, J = 12.0 Hz), 132.9 (d, J = 9.5 Hz), 118.0, 107.9 (d, J = 21.0 Hz), 103.9 (d, J = 24.1 Hz), 67.7, 65.2, 48.6, 33.1, 21.5, 20.3.

To a solution of a mixture of the above alcohols (200 mg, 0.95 mmol) in dry CH_2Cl_2 (50 mL) was added pyridinium chlorochromate (615 mg, 2.85 mmol) at 0 °C. The reaction mixture was allowed to stir at room temperature for 2 h. The resulting solution was filtered through a pad of Celite® and silica gel and the solvent was removed under reduced pressure. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 10) to afford ketone **6** (160 mg, 81%) as an oil; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.00–7.02 (m, 1H), 6.74–6.80 (m, 2H), 4.58 (d, J = 18.1 Hz, 1H), 4.27 (dd, J = 1.3, 18.1 Hz, 1H), 3.13 (d, J = 7.4 Hz, 1H), 2.20 (m, 1H), 0.98 (d, J = 6.8 Hz, 3H), 0.84 (d, J = 6.8 Hz, 3H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 209.4, 162.7 (d, J = 326 Hz), 155.3 (d, J = 15.0 Hz), 131.0 (d, J = 13.0 Hz), 120.6 (d, J = 4.0 Hz), 110.1 (d, J = 29.0 Hz), 105.6 (d, J = 32.0 Hz), 72.72, 58.0, 32.4, 20.5, 20.0. HRMS-EI (m/z): $[M]^+$ calcd for $C_{12}H_{13}FO_2$ 208.0899, found 208.0899.

(±)-(3*S*,4*S*)-Ethyl 2-(7-fluoro-3-hydroxy-4-isopropylchroman-3-yl)acetate (14). In a two neck round bottom flask equipped with a $CaCl_2$ tube, diethyl ether (8 mL) and ethyl iodoacetate (0.26 mL, 2.16 mmol) were added at room temperature. Et_2Zn (4.32 mL of a 1 M solution in *n*-hexane) was added and immediately the addition of a solution of ketone **7** (225 mg, 1.08 mmol) in diethyl ether (3 mL) was started using a syringe pump over a 50 min period. Then, a new portion of Et_2Zn (4.32 mL of a 1.0 M solution in *n*-hexane) was added. The resulting solution was stirred for 1.5 h and quenched with NH_4Cl (20 mL). It was extracted with diethyl ether (3 × 20 mL). The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 10) to afford β-hydroxy ester **14** (230 mg, 72%); ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.92–6.96 (m, 1H), 6.51–6.61 (m, 2H), 4.33 (s, 1H), 4.17 (q, J = 5.9 Hz, 2H) 4.06 (d, J = 11.0 Hz, 1H), 3.91 (dd, J = 2.1, 11.0 Hz, 1H), 2.56 (m, 1H), 2.55 (d, J = 16.8 Hz, 1H), 2.45 (d, J = 16.8 Hz, 1H), 2.32–2.36 (m, 1H),), 1.25 (t, J = 7.1 Hz, 3H) 1.13 (d, J = 6.9 Hz, 3H), 0.64 (d, J = 7.0 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 173.2, 162.4 (d, J = 242.3 Hz), 153.3 (d, J = 11.8 Hz), 131.8 (d, J = 9.4 Hz), 118.2, 107.4 (d, J = 21.5 Hz), 103.3 (d, J = 24.3 Hz), 69.0, 68.1, 61.1, 50.8, 41.8, 28.3, 25.0, 21.1, 14.1. HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{16}H_{22}FO_4$ 297.1502, found 297.1510.

(±)-(3*S*,4*S*)-2-(7-Fluoro-3-hydroxy-4-isopropylchroman-3-yl)-acetic acid (15). To a solution of β-hydroxy ester **14** (230 mg, 0.78 mmol) in MeOH (3 mL) and H_2O (1 mL) was added LiOH·H_2O (98 mg) at room temperature. The reaction mixture was stirred at the same temperature for 15 h and quenched

with 3 N HCl until pH 2 was reached. It was extracted with ethyl acetate (3 × 30 mL), washed with brine, and dried over anhydrous $MgSO_4$. The solvent was completely removed under reduced pressure to give carboxylic acid **15** (208 mg, quant.) as a solid; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.96–6.98 (m, 1H), 6.53–6.63 (m, 2H), 4.08 (d, J = 11.1 Hz, 1H), 3.97 (dd, J = 2.1, 11.1 Hz, 1H), 2.65 (d, J = 17.0 Hz, 1H), 2.61–2.62 (m, 1H), 2.57 (d, J = 17.0 Hz, 1H), 1.14 (d, J = 7.0 Hz, 3H), 0.66 (d, J = 7.0 Hz, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 178.1, 162.5 (d, J = 243.0 Hz), 153.2 (d, J = 11.2 Hz), 131.8 (d, J = 9.5 Hz), 118.0, 107.7 (d, J = 21.5 Hz), 103.5 (d, J = 24.2 Hz), 69.1, 67.9, 50.7, 41.7, 28.4, 25.0, 21.1. HRMS-CI (m/z): $[M + H]^+$ calcd for $C_{14}H_{18}FO_4$ 269.1189, found 269.1187.

(±)-(3*S*,4*S*)-2-(7-Fluoro-3-hydroxy-4-isopropylchroman-3-yl)-acetaldehyde (3). To a suspension of the lithium aluminium hydride (184 mg, 4.92 mmol) in dry THF (16.4 mL) was added β-hydroxy ester **14** (487 mg, 1.64 mmol) at 0 °C. The reaction mixture was allowed to stir at room temperature for 2 h. The reaction mixture was quenched with 30% NaOH (30 mL) for 20 min. The resulting solution was extracted with ethyl acetate (3 × 30 mL), washed with brine, and dried over anhydrous $MgSO_4$. The solvent was completely removed under reduced pressure. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 4) to afford the corresponding diol (400 mg, 96%) as a colorless oil. The spectroscopic data is identical to that of **16a** (see below).

Dess–Martin periodinane (226 mg, 533 μmol) was added to a solution of the above diol (90.3 mg, 355 μmol) in CH_2Cl_2 (4 mL). After stirring for 2 h, the reaction mixture was quenched with $Na_2S_2O_3$ (10 mL) and sat. $NaHCO_3$ (10 mL). The resulting mixture was extracted with Et_2O (3 × 15 mL). The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 4) to afford aldehyde **3** (72.4 mg, 81%) as a pale brown oil; ^{1}H-NMR (300 MHz, $CDCl_3$)δ 9.77 (s, 1H), 6.95 (dd, J = 6.4, 8.4 Hz, 1H), 6.60 (td, J = 2.5, 8.3 Hz, 1H), 6.53 (dd, J = 2.6, 10.5 Hz, 1H), 4.02 (d, J = 11.0 Hz, 1H), 3.93 (dd, J = 2.0, 11.0 Hz, 2H), 3.61 (s, 1H), 2.72 (dd, J = 18.7, 39.9 Hz, 2H), 2.60–2.58 (m, 1H), 2.35–2.24 (m, 1H), 1.12 (d, J = 7.0 Hz, 3H), 0.62 (d, J = 7.0 Hz, 3H).^{13}C-NMR (100 MHz, $CDCl_3$) δ 203.5, 162.4 (d, ^1J = 243 Hz), 153.3 (d, ^3J = 12 Hz), 131.8 (d, ^3J = 9 Hz), 118.2 (d, ^4J = 3 Hz), 107.6 (d, ^2J = 21 Hz), 103.4 (d, ^2J = 24 Hz), 70.1, 68.1, 50.8, 28.1, 25.1, 21.1. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{14}H_{18}FO_3$ 253.1235, found 253.1201.

5-Fluoro-2-(2-methylprop-1-en-1-yl)phenol (11). To a solution of aldehyde **8** (1.97 g, 14.0 mmol) in Et_2O (12 mL) cooled to 0 °C was slowly added isopropylmagnesium bromide (56 mL, 1.0 M in THF) for 30 min. The reaction was monitored by TLC, quenched with saturated aqueous NH_4Cl (50 mL) after completion of the reaction, and extracted with Et_2O (3 × 75 mL). The organic extracts were washed with water and brine, dried over $MgSO_4$, and concentrated under reduced pressure. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 4) to afford isobutyl alcohol (2.22 g, 86%) as a pale brown oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 8.45 (s, 1H), 6.82 (dd, J = 6.5, 8.3 Hz, 1H), 6.54 (td, J = 1.8, 5.4 Hz, 1H), 6.50 (dd, J = 2.8, 8.3 Hz, 1H), 4.48 (d, J = 7.0 Hz, 1H), 3.08 (s, 1H), 2.10–1.98 (m, 1H), 1.02 (d, J = 6.7 Hz, 3H), 0.83 (d, J = 6.8 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.9 (d, ^1J = 243 Hz), 157.0 (d, ^3J = 12 Hz), 129.1 (d, ^3J = 10 Hz), 122.0 (d, ^4J = 3 Hz), 106.2 (d, ^2J = 21 Hz), 104.4 (d, ^2J = 24 Hz), 81.5, 34.5, 19.1, 18.1. LRMS-EI (m/z): $[M]^+$ calcd for $C_{10}H_{13}FO_2$ 184.09, found 184.

In a sealed tube, a solution of the above isobutyl alcohol (952 mg, 5.17 mmol) in *n*-hexane (2 mL) was stirred using a microwave (170 °C, 120 W) for 15 min. The reaction mixture was treated with water (30 mL) and extracted with CH_2Cl_2 (3 × 30 mL). The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 6) to afford alkene **11** (800 mg, 93%) as a red brown oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.97 (dd, J = 7.0, 7.8 Hz, 1H), 6.65–6.57 (m, 2H), 6.04 (s, 1H), 5.27 (d, J = 1.4 Hz, 1H), 1.93 (d, J = 1.2 Hz, 3H), 1.66 (d, J = 0.9 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 165.5 (d, ^1J = 240 Hz), 153.9 (d, ^3J = 12 Hz), 114.4, 130.6 (d, ^3J = 10 Hz), 120.6 (d, ^4J = 3 Hz), 117.8, 107.1 (d, ^2J = 21 Hz), 102.4 (d, ^2J = 25 Hz), 25.7, 19.3. LRMS-EI (m/z): $[M]^+$calcd for $C_{10}H_{11}FO$ 166.08, found 166.

4-(Benzyloxy)-1-(5-fluoro-2-(2-methylprop-1-en-1-yl)phenoxy)-butan-2-one (9). To a solution of alkene **11** (220 mg, 1.32 mmol) in anhydrous CH_3CN (13 mL) were added epoxide **10** (472 mg, 2.65 mmol) and Cs_2CO_3 (647 mg, 1.99 mmol) at room temperature. The reaction mixture was allowed to warm to reflux for 21 h. The reaction solution was extracted with CH_2Cl_2 and washed with brine. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 4) to afford the corresponding alcohol (0.24 g, 52%) as a brown oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.38–7.26 (m, 5H), 7.11 (t, J = 7.6 Hz, 1H), 6.64 (td, J = 2.3, 8.3 Hz, 1H), 6.58 (dd, J = 2.2, 10.8 Hz, 1H), 6.19 (s, 1H), 4.55 (s, 1H), 4.22 (s, 1H), 3.94–3.87 (m, 2H), 3.78–3.67 (m, 2H), 2.96 (s, 1H) 1.97–1.88 (m, 5H), 1.76 (s, 1H).^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.1 (d, ^1J = 243 Hz), 156.9 (d, ^3J = 9 Hz), 138.0, 135.6, 131.0 (d, ^3J = 9 Hz), 128.5, 127.8, 127.7, 123.7 (d, ^4J = 4 Hz), 119.5, 106.8 (d, ^2J = 20 Hz), 100.2 (d, ^2J = 26 Hz), 73.3, 72.5, 68.9, 67.8, 33.1, 26.5, 19.5. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{21}H_{26}FO_3$ 345.1861, found 345.1861.

Dess–Martin periodinane (1.11 g, 2.62 mmol) was added to a solution of the above alcohol (600 mg, 1.74 mmol) in CH_2Cl_2 (18 mL). After stirring for 1 h, the reaction mixture was quenched with $Na_2S_2O_3$ (20 mL) and sat. $NaHCO_3$ (20 mL). The resulting mixture was extracted with Et_2O (3 × 60 mL). The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 10) to afford the ketone **9** (565 mg, 95%) as a colorless oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.37–7.27 (m, 5H), 7.16 (dd, J = 7.2, 8.0 Hz, 1H), 6.68 (td, J = 2.4, 8.3 Hz, 1H), 6.45 (dd, J = 2.4, 10.6 Hz, 1H), 6.30 (s, 1H), 4.59 (s, 2H), 4.54 (s, 2H), 3.81 (t, J = 6.1 Hz, 2H), 2.85 (t, J = 6.1 Hz, 2H), 1.94 (d, J =

1.1 Hz, 3H), 1.80 (d, J = 1.0 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 205.2, 162.0 (d, ^1J = 244 Hz), 156.1 (d, ^3J = 9 Hz), 137.9, 135.9, 131.3 (d, ^3J = 9 Hz), 128.5, 127.8, 127.7, 123.8 (d, ^4J = 3 Hz), 119.5, 107.4 (d, ^2J = 21 Hz), 100.4 (d, ^2J = 25 Hz), 73.6, 73.3, 64.9, 39.5, 26.5, 19.5. LRMS-EI (m/z): $[M]^+$ calcd for $C_{21}H_{23}FO_3$ 342.16, found 342.

(±)-(3*S*,4*S*)-3-(2-(Benzyloxy)ethyl)-7-fluoro-4-(prop-1-en-2-yl)-chroman-3-ol (16). To a solution of ketone **9** (32.4 mg, 94.6 μmol) in dry CH_2Cl_2 (1 mL) was added $SnCl_4$ (27.1 mg, 104 μmol) at −78 °C. The reaction mixture was allowed to stir at the same temperature for 2.5 h. The mixture was allowed to warm up to −40 °C over 3 h. After completion of the reaction (monitored by TLC), it was quenched with saturated aqueous $NaHCO_3$ (10 mL), extracted with CH_2Cl_2 (3 × 10 mL) and washed with brine. The organic layers were dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 4) to afford alcohol **16** (17.1 mg, 53%) as a colorless oil; *major isomer*: ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.39–7.26 (m, 5H), 6.96–6.92 (m, 1H), 6.60 (td, J = 2.6, 8.4 Hz, 1H), 6.55 (dd, J = 2.6, 8.4 Hz, 1H), 5.13 (t, J = 1.6 Hz, 1H), 4.66 (t, J = 0.8 Hz, 1H), 4.54 (d, J = 2.2 Hz, 2H), 4.11 (d, J = 11.0 Hz, 1H), 3.90 (dd, J = 1.4, 11.0 Hz, 1H), 3.83–3.72 (m, 2H), 3.61 (s, 1H), 3.43 (s, 1H), 2.09–2.02 (m, 1H), 1.84–1.77 (m, 4H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.1 (d, ^1J = 243 Hz), 154.3 (d, ^3J = 12 Hz), 146.4, 137.4, 131.5 (d, ^3J = 10 Hz), 128.6, 128.0, 127.9, 119.11 (d, ^4J = 3 Hz), 117.4, 108.3 (d, ^2J = 21 Hz), 103.4 (d, ^2J = 25 Hz), 73.7, 69.7, 69.0, 66.8, 53.2, 36.3, 23.4. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{21}H_{24}FO_3$ 343.1704, found 343.1705; *minor isomer*: ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.38–7.28 (m, 5H), 6.98 (dd, J = 6.8, 8.2 Hz, 1H), 6.62–6.55 (m, 2H), 5.02 (t, J = 1.5 Hz, 1H), 4.81 (s, 1H), 4.5 (s, 2H), 4.03 (d, J = 0.6 Hz, 2H), 3.86–3.74 (m, 2H), 3.72 (s, 1H), 3.49 (s, 1H), 2.00–1.93 (m, 1H), 1.76–1.70 (m, 1H), 1.67 (d, J = 0.5 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$)δ 162.1 (d, ^1J = 243 Hz), 154.3 (d, ^3J = 12 Hz), 144.5, 137.4, 131.6 (d, ^3J = 10 Hz), 128.6, 128.0, 127.9, 118.5 (d, ^4J = 3 Hz), 117.2, 108.3 (d, ^2J = 21 Hz), 103.4 (d, ^2J = 25 Hz), 73.6, 70.3, 69.8, 66.5, 54.6, 33.5, 22.3. LRMS-EI (m/z): $[M]^+$ calcd for $C_{21}H_{23}FO_3$ 342.16, found 342.

(±)-(3*S*,4*S*)-7-Fluoro-3-(2-hydroxyethyl)-4-isopropylchroman-3-ol (16a). To a solution of the alkene **16** (17.1 mg, 50.0 μmol, only major) in MeOH (1 mL) was added Pd/C (5 mg, 10% (w/w)) at room temperature. The reaction mixture was stirred under a hydrogen atmosphere (with the aid of a hydrogen balloon) for 7 h. It was filtered through a pad of Celite® and the solvent was removed under reduce pressure. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 2) to afford the alcohol **16a** (10.9 mg, 86%) as a colorless oil; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.94 (dd, J = 8.9, 11.1 Hz, 1H), 6.55 (td, J = 3.4, 11.1 Hz, 1H), 6.49 (dd, J = 3.3, 13.6 Hz, 1H), 4.07 (d, J = 11.0 Hz, 1H), 3.97 (dd, J = 2.4, 8.2 Hz, 1H), 4.03–3.95 (m, 2H), 2.59 (s, 1H), 2.38–2.27 (m, 1H), 1.84–1.68 (m, 2H), 1.15 (d, J = 7.0 Hz, 3H), 1.14 (d, J = 7.0 Hz, 3H), 0.64 (d, J = 7.0 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.3 (d, ^1J = 242 Hz), 153.6 (d, ^3J = 12 Hz), 131.6 (d, ^3J = 10 Hz), 119.0 (d, ^4J = 3 Hz), 107.1 (d, ^2J = 21 Hz), 103.2 (d, ^2J = 24 Hz), 71.2, 68.0, 59.6, 50.8, 38.5, 27.8, 25.4, 21.1. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{14}H_{20}FO_3$ 255.1391, found 255.1379.

(±)-(3*S*,4*S*)-3-(2-((3-(1*H*-Benzo[*d*]imidazol-2-yl)propyl)amino)-ethyl)-7-fluoro-4-isopropylchroman-3-ol (2aa). Acetic acid was added to a solution of amine **4a** (98.2 mg, 561 μmol) in anhydrous MeOH (4.5 mL) until pH 6 was reached. To the resulting solution was added sodium cyanoborohydride (26.4 mg, 420 μmol) and aldehyde **3** (72.4 mg, 280 μmol). The reaction mixture was stirred at room temperature for 18 h. The reaction was monitored by TLC, quenched with saturated aqueous $NaHCO_3$ (10 mL) after completion of the reaction, and extracted with CH_2Cl_2 (3 × 10 mL). The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (CH_2Cl_2–MeOH–H_2O–NH_4OH = 80 : 20 : 1 : 1) to afford amine **2aa** (104 mg, 91%) as a brown oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.48–7.46 (m, 2H), 7.19–7.16 (m, 2H), 6.91 (dd, J = 6.6, 8.4 Hz, 1H), 6.55 (td, J = 2.6, 8.3 Hz, 1H), 6.47 (dd, J = 2.5, 10.2 Hz, 1H), 4.06 (d, J = 11.0 Hz, 1H), 3.90 (dd, J = 1.8, 11.0 Hz, 1H), 2.94–2.88 (m, 4H), 2.71–2.58 (m, 2H), 2.53 (s, 1H), 2.37–2.29 (m, 1H), 1.96 (m, J = 6.3 Hz, 2H), 1.69–1.54 (m, 2H), 1.13 (d, J = 7.0 Hz, 3H), 0.61 (d, J = 6.9 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.3 (d, ^1J = 242 Hz), 154.4, 153.5 (d, ^3J = 12 Hz), 138.2, 131.7 (d, ^3J = 10 Hz), 122.4, 118.9 (d, ^4J = 3 Hz), 114.6, 107.1 (d, ^2J = 21 Hz), 103.1 (d, ^2J = 2.5 Hz), 71.1, 67.8, 51.6, 47.7, 44.9, 34.8, 27.8, 27.1, 26.5, 25.5, 21.0. HPLC: 99.5%, RT 12.02 min. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{24}H_{31}FN_3O_2$ 412.2395, found 412.2398.

(±)-(3*S*,4*S*)-3-(2-(3-((1*H*-Benzo[*d*]imidazol-2-yl)methyl)piperidin-1-yl)ethyl)-7-fluoro-4-isopropylchroman-3-ol (2ba). By following the same procedure as that used for the synthesis of **2aa**, the reaction of amine **25** (56 mg, 261 μmol), sodium cyanoborohydride (16 mg, 261 μmol), and aldehyde **3** (44 mg, 174 μmol) in MeOH (2 mL) gave an inseparable 1 : 1 mixture of amines **2ba** (37 mg, 47%) after purification by column chromatography on silica gel (CH_2Cl_2–MeOH = 10 : 1) as an oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.57 (m, 2H + 2H′), 7.26–7.19 (m, 2H + 2H′), 6.96 (dd, J = 6.8, 8.4 Hz, 1H), 6.94 (dd, J = 6.8, 8.4 Hz, 1H′), 6.57 (td, J = 2.4, 8.4 Hz, 1H), 6.55 (td, J = 2.4, 8.4 Hz, 1H′), 6.51–6.47 (m, 1H + 1H′) 4.16 (d, J = 10.7 Hz, 1H), 4.10 (d, J = 11.2 Hz, 1H′), 3.94–3.86 (m, 1H + 1H′), 3.05–2.73 (m, 3H + 3H′), 2.62–2.21 (m, 8H + 8H′) 1.82–1.40 (m, 4H + 4H′), 1.25 (d, J = 6.4 Hz, 3H), 1.15 (d, J = 6.8 Hz, 3H′), 0.69 (d, J = 7.2 Hz, 3H), 0.67 (d, J = 8.0 Hz, 3H′). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.4 (d, ^1J = 242 Hz, C), 162.3 (d, ^1J = 242 Hz, C′), 153.6 (d, ^3J = 11 Hz, C), 153.5 (d, ^3J = 11 Hz, C′), 131.8 (d, ^3J = 9 Hz, C), 131.7 (d, ^3J = 10 Hz, C′), 122.2 (d, ^4J = 2 Hz, C), 119.0 (d, ^4J = 2 Hz, C′), 118.8 (C + C′), 114.5 (C + C′), 107.1 (d, ^2J = 22 Hz, C), 107.0 (d, ^2J = 22 Hz, C′), 103.1 (d, ^2J = 24 Hz, C), 103.1 (d, ^2J = 25 Hz, C′), 71.6 (C + C′), 68.7 (C + C′), 67.2 (C + C′), 54.8 (C), 54.7 (C′), 53.4 (C + C′), 52.4 (C + C′), 51.0 (C + C′), 35.5 (C), 35.3 (C′), 32.1 (C), 31.7 (C′), 29.8 (C), 29.4 (C′), 28.1 (C), 27.8 (C′), 25.7 (C), 25.5 (C′), 21.2 (C), 21.1 (C′). HPLC: 97.6%, RT 8.67 min. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{27}H_{35}FN_3O_2$ 452.2708, found 452.2704.

(±)-(3*S*,4*S*)-3-(2-((3-(1*H*-Benzo[*d*]imidazol-2-yl)propyl)(isopropyl)amino)ethyl)-7-fluoro-4-isopropylchroman-3-ol (2ca). To a solution of amine **2aa** (33.8 mg, 82.1 μmol) in anhydrous MeOH (1 mL) were added acetic acid (9.40 μL, 164 μmol), sodium cyanoborohydride (10.3 mg, 164 μmol) and acetone (24.1 μL, 329 μmol). The reaction mixture was stirred for 19.5 h at room temperature. The reaction was monitored by TLC, quenched with saturated aqueous $NaHCO_3$ (10 mL) after completion of the reaction, and extracted with CH_2Cl_2 (3 × 15 mL). The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated under reduced pressure. The resulting residue was purified by flash column chromatography on silica gel (CH_2Cl_2–MeOH = 10 : 1) to afford amine **2ca** (17.3 mg, 46%) as a colorless oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.52–7.48 (m, 2H), 7.23–7.19 (m, 2H), 6.92 (dd, J = 6.6, 8.6 Hz, 1H), 6.55 (td, J = 2.5, 8.3 Hz, 1H), 6.48 (dd, J = 2.4, 10.4 Hz, 1H), 4.11 (d, J = 10.8 Hz, 1H), 3.93 (d, J = 2.2, 11.0 Hz, 1H), 3.22 (m, J = 6.8 Hz, 1H), 3.03–2.90 (m, 2H), 2.77 (t, J = 6.0 Hz, 2H), 2.63–2.58 (m, 3H), 2.45–2.38 (m, 1H), 2.10–2.04 (m, 2H), 1.15 (d, J = 6.8 Hz, 3H), 1.08 (d, J = 6.4 Hz, 6H), 0.64 (d, J = 6.8 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$)δ 162.4 (d, ^1J = 242 Hz), 154.5, 153.6 (d, ^3J = 11 Hz), 138.3, 131.6 (d, ^3J = 9 Hz), 122.3, 118.7 (d, ^4J = 3 Hz), 114.6, 107.0 (d, ^2J = 22 Hz), 103.1 (d, ^2J = 24 Hz), 71.3, 68.4, 51.4, 49.5, 48.6, 45.5, 32.2, 27.7, 26.5, 28.8, 25.5, 20.9, 17.3, 16.6. HPLC: 98.7%, RT 15.47 min. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{27}H_{37}FN_3O_2$ 454.2864, found 454.2867.

(±)-(3*S*,4*S*)-3-(2-(Cyclopropylamino)ethyl)-7-fluoro-4-isopropylchroman-3-ol (24a). To a solution of the aldehyde **3** (31.8 mg, 126 μmol) in dry MeOH (1.5 mL) were sequentially added cyclopropylamine (14.4 mg, 252 μmol) and sodium cyanoborohydride (11.9 mg, 189 μmol) at room temperature. The reaction was allowed to stir for 18 h at the same temperature. The reaction mixture was quenched with $NaHCO_3$ (10 mL), extracted with CH_2Cl_2 (3 × 10 mL) and washed with brine. The organic layers were dried over anhydrous $MgSO_4$ and concentrated *in vacuo.* The resulting residue was purified by flash column chromatography on silica gel (CH_2Cl_2–MeOH = 10 : 1) to afford amine **24a** (24.8 mg, 67%) as a colorless oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.92 (dd, J = 6.7, 8.4 Hz, 1H), 6.54 (td, J = 2.6, 8.4 3.03–2.96 (m, 2H), 2.49 (m, 1H), 2.33–2.25 (m, 1H), 2.12–2.07 (m, 1H), 1.64–1.58 (m, 1H), 1.53–1.47 (m, 1H), 1.13 (d, J = 6.9 Hz, 3H), 0.60 (d, J = 7.0 Hz, 3H), 0.52–0.40 (m, 4H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.2 (d, ^1J = 242 Hz), 153.7 (d, ^3J = 11 Hz), 131.7 (d, ^3J = 10 Hz), 119.3 (d, ^4J = 3 Hz), 106.8 (d, ^2J = 22 Hz), 102.5 (d, ^2J = 24 Hz), 70.6, 68.1, 51.8, 45.4, 35.6, 30.6, 27.8, 25.5, 21.1, 6.3, 6.1.

(±)-(3*S*,4*S*)-3-(2-((3-(1*H*-Benzo[*d*]imidazol-2-yl)propyl)(cyclopropyl)amino)ethyl)-7-fluoro-4-isopropylchroman-3-ol (2da). To a solution of amine **24a** (24.8 mg, 85.0 μmol) in anhydrous MeOH (1 mL) were added acetic acid (9.68 μL, 169 μmol), sodium cyanoborohydride (10.6 mg, 169 μmol) and aldehyde **21a** (38.4 mg, 117 μmol). The reaction was stirred for 17.5 h at room temperature. The reaction mixture was monitored by TLC, quenched with saturated aqueous $NaHCO_3$ (10 mL) after completion of the reaction, and extracted with CH_2Cl_2 (3 × 10 mL). The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (EtOAc–*n*-hexane = 1 : 1) to afford tertiary amine (33.8 mg, 66%) as a colorless oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 8.04–8.02 (m, 1H), 7.80 (d, J = 8.4 Hz, 1H), 7.64 (d, J = 6.4 Hz, 1H), 7.37–7.30 (m, 2H), 7.29–7.26 (m, 2H), 6.92 (dd, J = 6.8, 8.4 Hz, 1H), 6.75 (s, 1H), 6.55 (td, J = 2.4, 8.4 Hz, 1H), 6.49 (dd, J = 2.4, 10.4 Hz, 1H), 4.00 (d, J = 11.2 Hz, 1H), 3.90 (dd, J = 1.8, 11.0 Hz, 1H), 3.15 (t, J = 7.4 Hz, 1H), 2.98–2.84 (m, 4H), 2.52 (s, 1H), 2.38 (s, 3H), 2.35–2.21 (m, 3H), 1.79 (s, 1H), 1.64 (s, 2H), 1.11 (d, J = 6.8 Hz, 3H), 0.59 (d, J = 7.2 Hz, 7H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.3 (d, ^1J = 242 Hz), 154.4, 153.7 (d, ^3J = 12 Hz), 146.0, 141.9, 135.5, 133.2, 131.7 (d, ^3J = 10 Hz), 130.3, 126.8, 124.8, 124.7, 119.8, 119.3 (d, ^4J = 3 Hz), 113.6, 106.7 (d, ^2J = 21 Hz), 103.0 (d, ^2J = 24 Hz), 70.6, 68.4, 54.5, 51.8, 51.7, 37.4, 32.2, 28.0, 27.7, 25.5, 23.7, 25.5, 23.7, 21.7, 21.0, 6.9, 6.2.

To a solution of the above product (18.2 mg, 30.0 μmol) in distilled THF (0.6 mL) was added a solution of tetrabutylammonium fluoride (300 μL, 1.0 M solution in THF, 30.6 mmol) at room temperature. The reaction mixture was allowed to warm to reflux for 4 h. The resulting mixture was extracted with Et_2O. The organic layers were washed with brine, dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica gel (CH_2Cl_2–MeOH = 10 : 1) to afford amine **2da** (7.40 mg, 55%) as a pale colorless oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.57–7.55 (m, 2H), 7.26–7.21 (m, 2H), 6.93 (dd, J = 6.6, 8.5 Hz, 1H), 6.56 (td, J = 2.6, 8.3 Hz, 1H), 6.49 (dd, J = 2.6, 10.2 Hz, 1H), 4.10 (d, J = 10.9 Hz, 1H), 3.93 (dd, J = 2.1, 10.8 Hz, 1H), 2.99–2.87 (m, 4H), 2.70–2.66 (m, 2H), 2.57 (s, 1H), 2.42–2.34 (m, 1H), 2.89–2.18 (m, 2H), 1.78–1.72 (m, 1H), 1.62 (t, J = 5.9 Hz, 2H), 1.18 (d, J = 7.0 Hz, 3H), 0.70–0.59 (m, 7H). ^{13}C-NMR (75 MHz, $CDCl_3$) δ 162.4 (d, ^1J = 241 Hz), 154.4, 153.5 (d, ^3J = 12 Hz), 138.6, 131.6 (d, ^3J = 9.8 Hz), 122.3, 118.8 (d, ^4J = 3 Hz), 114.7, 107.0 (d, ^2J = 22 Hz), 103.1 (d, ^2J = 24 Hz), 75.6, 68.3, 54.3, 53.0, 51.4, 37.9, 32.2, 27.8, 26.6, 25.6, 25.2, 21.0, 6.7, 6.2. HPLC: 96.1%, RT 19.06 min. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{27}H_{35}FN_3O_2$ 452.2708, found 452.2710.

(±)-(3*S*,4*S*)-3-(2-((3-(1*H*-Benzo[*d*]imidazol-2-yl)propyl)(2,2,2-trifluoroethyl)amino)ethyl)-7-fluoro-4-isopropylchroman-3-ol (2ea). By following the same procedure as that used for the synthesis of **2da**, the reaction of amine **24b** (38.8 mg, 116 μmol), acetic acid (13.3 μL, 231 μmol), sodium cyanoborohydride (10.9 mg, 174 μmol) and aldehyde **21a** (44.8 mg, 136 μmol) in MeOH (1 mL) gave a tertiary amine intermediate (57.2 mg, 76%) after purification by column chromatography on silica gel (CH_2Cl_2–*n*-hexane = 1 : 2) as a colorless oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 8.05–8.03 (m, 1H), 7.80 (d, J = 8.4 Hz, 1H), 7.66–7.64 (m, 1H), 7.38–7.26 (m, 4H), 6.93 (dd, J = 6.7, 8.4 Hz, 1H), 6.55 (td, J = 2.6, 8.3 Hz, 1H), 6.49 (dd, J = 2.5, 10.2 Hz, 1H), 4.04 (d, J = 11.1 Hz, 1H), 3.91 (dd, J = 1.9, 11.1 Hz, 1H), 3.20–3.11 (m, 1H), 2.94–2.89 (m, 2H), 2.88–2.83 (m, 2H), 2.55 (s, 1H), 2.38–2.32 (m, 4H), 2.18–2.11 (m, 2H), 1.71–1.56 (m, 2H), 1.10 (d, J = 7.0 Hz, 3H), 0.60 (d, J = 6.9 Hz, 3H).^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.3 (d, ^1J = 242 Hz), 153.9, 153.6 (d, ^3J = 12 Hz), 146.1, 141.8, 135.4,

133.2, 131.7 (d, ^3J = 9 Hz), 130.3, 126.8, 125.4 (d, ^1J = 279 Hz), 124.9, 124.7, 119.8, 119.1 (d, ^4J = 3 Hz), 113.6, 106.9 (d, ^2J = 21 Hz), 103.1 (d, ^2J = 24 Hz), 70.5, 68.3, 54.7 (q, ^2J = 30 Hz), 53.3, 51.4, 51.3, 33.2, 27.7, 27.2, 25.3, 23.8, 21.6, 21.0.

The reaction of the above tertiary amine (57.2 mg, 88.3 μmol) and tetrabutylammonium fluoride (883 μmol, 1.0 M solution in THF) in THF (1 mL) gave amine **2ea** (16.0 mg, 57%) after purification by column chromatography on silica gel (33.0 mg, 76%) as a yellow oil; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.56 (s, 2H), 7.26–7.22 (m, 2H), 6.93 (dd, J = 6.6, 8.4 Hz, 1H), 6.56 (td, J = 2.6, 8.3 Hz, 1H), 6.49 (dd, J = 2.6, 10.2 Hz, 1H), 4.12 (d, J = 11.0 Hz, 1H), 3.90 (dd, J = 2.1, 11.0 Hz, 1H), 3.05 (q, J = 9.4 Hz, 2H), 2.94 (t, J = 7.1 Hz, 2H), 2.87 (t, J = 5.7 Hz, 2H), 2.67 (t, J = 6.6 Hz, 2H), 2.58 (s, 1H), 2.44–2.37 (m, 1H), 2.11–2.04 (m, 2H), 1.65–1.53 (m, 2H), 1.15 (d, J = 7.0 Hz, 3H), 0.64 (d, J = 6.9 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.4 (d, ^1J = 242 Hz), 154.0, 153.5 (d, ^3J = 12 Hz), 131.6 (d, ^3J = 9 Hz), 130.0, 125.3 (q, ^1J = 277 Hz), 122.4, 118.7 (d, ^4J = 3 Hz), 115.7, 107.1 (d, ^2J = 22 Hz), 103.1(d, ^2J = 24 Hz), 71.3, 68.2, 55.0 (q, ^2J = 31 Hz), 53.2, 52.4, 51.2, 33.1, 27.8, 26.0, 25.5, 25.3, 20.9. HPLC: 96.6%, RT 17.68 min. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{26}H_{32}F_4N_3O_2$ 494.2425, found 494.2426.

(±)-(3*S*,4*S*)-2-(7-Fluoro-3-hydroxy-4-isopropylchroman-3-yl)acetaldehyde *O*-(2-(1*H*-benzo[*d*]imidazol-2-yl)ethyl) oxime (2fa). To a solution of aldehyde **3** (68 mg, 0.27 mmol) in MeOH (1 mL) were added K_2CO_3 and *N*-alkoxyamine **5a** (38 mg, 0.18 mmol) at 0 °C. The reaction mixture was stirred for 30 min at room temperature. The solvent of the reaction mixture was removed under reduced pressure. The resulting mixture was extracted with EA and washed with brine. The organic layer was dried over anhydrous $MgSO_4$ and concentrated *in vacuo*. The resulting residue was purified by flash column chromatography on silica-gel (EtOAc–*n*-hexane = 1 : 1) to give oxime **2fa** (21 mg, 28%); ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.53–7.54 (m, 2H), 7.48 (t, J = 6.0 Hz, 1H), 7.19–7.25 (m, 2H) 6.95 (dd, J = 6.6, 8.3 Hz, 1H), 6.51–6.62 (m, 2H), 4.40–4.47 (m, 2H), 4.06 (d, J = 11.0 Hz, 1H), 3.87 (dd, J = 1.8, 11.0 Hz, 1H), 3.21–3.36 (m, 2H), 2.59 (s, 1H), 2.31–2.46 (m, 3H), 1.13 (d, J = 7.0 Hz, 3H), 0.66 (d, J = 6.9 Hz, 3H). ^{13}C-NMR (100 MHz, $CDCl_3$) δ 162.4 (d, J = 242.0 Hz), 153.5 (d, J = 12.0 Hz), 152.2, 149.1, 137.5, 131.7 (d, J = 9.0 Hz), 122.8, 118.5 (d, J = 3.0 Hz), 114.7, 107.5 (d, J = 21.0 Hz), 103.4 (d, J = 24.0 Hz),70.9, 70.4, 68.2, 50.8, 38.5, 29.1, 28.3, 25.1, 21.0. HPLC: 95.1%, RT 17.34 min. HRMS-ESI (m/z): $[M + H]^+$ calcd for $C_{23}H_{27}FN_3O_3$ 412.2031, found 412.2030.

In vitro assay for T-type calcium channels blockade

HEK293 cells which stably express both α_{1G} (or α_{1H}) and Kir2.1 subunits were grown in Dulbecco's modified Eagle's medium supplemented with 10% (v/v) fetal bovine serum, penicillin (100 U mL^{-1}), streptomycin (100 μg mL^{-1}), geneticin (500 μg mL^{-1}), and puromycin (1 μg mL^{-1}) at 37 °C in a humid atmosphere of 5% CO_2 and 95% air. Cells were seeded onto 96-well black wall clear bottom plates at a density of 4×10^4 cells per well and were used the next day for the high-throughput screening (HTS) FDSS6000 assay. For the FDSS6000 assay, cells were incubated for 60 min at room temperature with 5 μM fluo3/AM and 0.001% Pluronic F-127 in a Hepes-buffered solution composed of (in mM): 115 NaCl, 5.4 KCl, 0.8 $MgCl_2$, 1.8 $CaCl_2$, 20 Hepes, and 13.8 glucose (pH 7.4). During the fluorescence-based FDSS6000 assay, a_{1G} T-type Ca^{2+} channels were activated using a high concentration of KCl (70 mM) in 10 mM $CaCl_2$ contained Hepes-buffered solution and the increase in $[Ca^{2+}]_i$ by KCl-induced depolarization was detected. During the whole procedure, cells were washed using a BIO-TEK 96-well washer. All data were collected and analyzed using FDSS6000 and related software (Hamamatsu, Japan).

CYP inhibition assay

The test compound (40 μL of a 25 μM solution of the compound in distilled water) was seeded onto a 96-well plate and then 50 μl of the Master Pre-Mix (CYP450 BACULOSOMES® Reagent and Regeneration System) was added. The plate was incubated for 20 min at 37 °C to allow the compound to interact with CYP450. The reaction was initiated by adding Vivid® CYP450 substrate/NADP$^+$ buffer (10 μL). The remaining enzyme activity was measured by reading the amount of fluorescent product using a fluorescent plate reader. The reference inhibitor for each CYP isozyme is indicated in Table 2.

Microsomal stability assay

Human liver microsomes (0.5 mg mL^{-1}) and the test compound (1 μM) in potassium phosphate buffer (0.1 M, pH 7.4) were incubated at 37 °C. Reactions were initiated by the addition of β-NADPH (1.2 mM) and continued at 37 °C. After incubation for 30 min, the reaction was stopped by addition of acetonitrile with the internal standard. The samples were analyzed using LC–MS/MS. Mibefradil was used as a reference standard. The results are expressed as the percentage of drug remaining after incubation.

Acknowledgements

This research was supported by the Korea Institute of Science and Technology (KIST-2E24670, 2E25023, 2E24510, 2V03370) and the National Research Foundation of Korea (NRF-2013R1A1A2005550) funded by the Ministry of Education, Science and Technology.

Notes and references

1 W. Osterrieder and M. Holck, *J. Cardiovasc. Pharmacol.*, 1989, **13**, 754–759.
2 S. K. Mishra and K. Hermsmeyer, *Circ. Res.*, 1994, **75**, 144–148.
3 I. Bezprozvanny and R. W. Tsien, *Mol. Pharmacol.*, 1995, **48**, 540–549.
4 H. A. Welker, H. Eggers, C. Kleinbloesem, K. Erb, K. Breithaupt, R. Butzer and G. G. Belz, *Eur. J. Clin. Pharmacol.*, 1989, **36**(suppl. A), A304.
5 M. Spoendlin, J. Peters, H. Welker, A. Bock and G. Thiel, *Nephrol., Dial., Transplant.*, 1998, **13**, 1787–1791.

6 H. R. Wiltshire, B. M. Sutton, G. Heeps, A. M. Betty, D. W. Angus, S. R. Harris, E. Worth and H. A. Welker, *Xenobiotica*, 1997, **27**, 557–571.
7 C. M. Brown, B. Reisfeld and A. N. Mayeno, *Drug Metab. Rev.*, 2008, **40**, 1–100.
8 E. Fontana, P. M. Dansette and S. M. Poli, *Curr. Drug Metab.*, 2005, **6**, 413–454.
9 B. K. Park, A. Boobis, S. Clarke, C. E. P. Goldring, D. Jones, J. G. Kenna, C. Lambert, H. G. Laverty, D. J. Naisbitt, S. Nelson, D. A. Nicoll-Griffith, R. S. Obach, P. Routledge, D. A. Smith, D. J. Tweedie, N. Vermeulen, D. P. Williams, I. D. Wilson and T. A. Baillie, *Nat. Rev. Drug Discovery*, 2011, **10**, 292–306.
10 E. H. Kerns and L. Di, in *Drug-like Properties: Concept, Structure, Design and Methods: from ADME to Toxicity Optimization*, Elsevier Academic Press, Amsterdam, 1st edn, 2008, pp. 197–208.
11 R. Ocampo and W. R. Dolbier Jr., *Tetrahedron*, 2004, **60**, 9325–9374.
12 A. Fürstner, *Synthesis*, 1989, 571.
13 M. W. Rathke and P. Weipert, in *Comprehensive Organic Synthesis*, ed. B. M. Trost and I. Fleming, Pergamon, New York, 1991, vol. 2, p. 277.
14 A. Fürstner, in *Organozinc Reagents*, ed. P. Knochel and P. Jones, Oxford University Press, New York, 1999, p. 287.
15 W. Oppolzer and V. Snieckus, *Angew. Chem., Int. Ed. Engl.*, 1978, **17**, 476–486.
16 M. L. Clarke and M. B. France, *Tetrahedron*, 2008, **64**, 9003–9031.
17 K. Mikami, M. Terada, S. Narisawa and T. Nakai, *Synlett*, 1992, 255–265.
18 K. Mikami and M. Shimizu, *Chem. Rev.*, 1992, **92**, 1021–1050.
19 B. B. Snider, *Acc. Chem. Res.*, 1980, **13**, 426–432.
20 G. Casiraghi, G. Casnati, M. Cornia, A. Pochini, G. Puglia, G. Sartori and R. Ungaro, *J. Chem. Soc., Perkin Trans. 1*, 1978, 318–321.
21 J. H. Byun, H. Kim, Y. Kim, I. Mook-Jung, D. J. Kim, W. K. Lee and K. H. Yoo, *Bioorg. Med. Chem. Lett.*, 2008, **18**, 5591–5593.
22 Y. Li, K. J. Jardine, R. Tan, D. Song and V. M. Dong, *Angew. Chem., Int. Ed.*, 2009, **48**, 9690–9692.
23 J. Vader, H. Sengers and A. de Groot, *Tetrahedron*, 1989, **45**, 2131–2142.
24 J. R. Parikh, W. von and E. Doering, *J. Am. Chem. Soc.*, 1967, **89**, 5505–5507.
25 H. Schick, R. Ludwig, K. Kleiner and A. Kunath, *Tetrahedron*, 1995, **51**, 2939–2946.
26 N. A. Fernández-Ibánez, B. Macia, A. J. Minnaard and B. L. Feringa, *Chem. Commun.*, 2008, 2571–2573.
27 K. Kanai, H. Wakabayashi and T. Honda, *Org. Lett.*, 2000, **2**, 2549–2551.
28 CCDC-980275 contains the supplementary crystallographic data for compound **15**.
29 A. Arduini, A. Pochini and R. Ungaro, *Synthesis*, 1984, 950–953.
30 C. D. Poulter and M. Muehlbacher, *J. Org. Chem.*, 1988, **53**, 1026–11030.
31 T. D. Nelson, J. D. Rosen, J. H. Smitrovich, J. Payack, B. Craig, L. Matty, M. A. Huffman and J. McNamara, *Org. Lett.*, 2005, **7**, 55–58.
32 Y. J. Zhao, B. Li, L. J. S. Tan, Z. L. Shen and T. P. Loh, *J. Am. Chem. Soc.*, 2010, **132**, 10242–10244.
33 C. E. Davis and R. M. Coates, *Angew. Chem., Int. Ed.*, 2002, **41**, 491–493.
34 M. Tegoni, L. Ferretti, F. Sansone, M. Remelli, V. Bertolasi and F. Dallavalle, *Chem. – Eur. J.*, 2007, **13**, 1300–1308.
35 A. A. Shazhenov, Ch. Sh. Kadyrov and P. Kurbanov, *Chem. Heterocycl. Compd.*, 1972, **8**, 580–583.
36 M. L. Gillaspy, B. A. Lefker, W. A. Hada and D. J. Hoover, *Tetrahedron Lett.*, 1995, **36**, 7399–7402.
37 S. B. Benard, L. Neuville and J. Zhu, *Chem. Commun.*, 2010, **49**, 3393–3395.
38 A. Yasuhara and T. Sakamoto, *Tetrahedron Lett.*, 1998, **39**, 595–596.
39 The Functional Drug Screening System (FDSS6000), manufactured by Hamamatsu Photonics, was used for quantitatively analyzing calcium mobility by way of measuring the fluorescence intensity.
40 We purchased Vivid® CYP450 Screening Kits from Invitrogen and measured the remaining enzyme activity by following the protocol provided by the company. For more details, see: B. D. Marks, R. W. Smith, H. A. Braun, T. A. Goossens, M. Christenson, M. S. Ozers, C. S. Lebakken and O. V. Trubetskoy, *Assay Drug Dev. Technol.*, 2002, **1**, 73–81.