Bioorganic & Medicinal Chemistry 19 (2011) 663–676

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry

journal homepage: www.elsevier.com/locate/bmc

Synthesis and pharmacological evaluation of 4-(3,4-dichlorophenyl)-*N*-methyl-1,2,3,4-tetrahydronaphthalenyl amines as triple reuptake inhibitors

Liming Shao *, Fengjiang Wang, Scott C. Malcolm, Jianguo Ma, Michael C. Hewitt, Una C. Campbell, Larry R. Bush, Nancy A. Spicer, Sharon R. Engel, Lakshmi D. Saraswat, Larry W. Hardy, Patrick Koch, Rudy Schreiber †, Kerry L. Spear, Mark A. Varney ‡

Discovery and Early Clinical Research, Sepracor Inc., 84 Waterford Drive, Marlborough, MA 01752, USA

ARTICLE INFO

Article history:
Received 10 August 2010
Revised 10 October 2010
Accepted 13 October 2010
Available online 21 October 2010

Keywords:
Triple uptake inhibitor (TRI)
Depression
Antidepressant
Monoamine transporter

ABSTRACT

The present work describes a series of novel chiral amines that potently inhibit the in vitro reuptake of serotonin, norepinephrine and dopamine (triple reuptake inhibitors) and were active in vivo in a mouse model predictive of antidepressant like activity. The detailed synthesis and in vitro activity and ADME profile of compounds is described, which represent a previously undisclosed triple reuptake inhibitor chemotype.

1. Introduction

Commonly used antidepressants increase synaptic availability of biogenic amines by blocking their transport or reuptake into nerve terminals, which is the major mechanism for their physiological inactivation. Examples include norepinephrine and serotonin reuptake inhibitors (NSRIs) such as venlafaxine and milnacipram, which inhibit the reuptake of norepinephrine (NE) and serotonin (5-HT), selective serotonin reuptake inhibitors (SSRIs) such as fluoxetine (Prozac) and sertraline (Zoloft) and norepinephrine reuptake inhibitors (NRIs) such as reboxetine.[1] These compounds offer side-effect advantages over older monoamine oxidase inhibitors (MAOIs), which block metabolism of biogenic amines to increase synaptic availability, but offer no clear efficacy advantages. A major drawback to both reuptake inhibitors and MAOIs is the therapeutic lag[2] (slow onset) associated with their use. The patients must take the drugs for up to 3 weeks to achieve clinically meaningful symptom relief. Furthermore, the rate of patient response to the treatment is low.

One strategy to reduce therapeutic lag and improve efficacy is the addition of a dopamine component to a dual reuptake inhibitor to create a triple reuptake inhibitor (TRI). Dopamine reuptake inhibitors (e.g., buproprion) and dopamine agonists (e.g., pramipexole) are already used clinically as antidepressants[3] and to augment the effects of traditional antidepressants in treatment of refractory patients.[4] In addition, deficits in mesocorticolimbic dopaminergic function have been linked clinically and pre-clinically to anhedonia, which is a core symptom of depression.[5]

Medicinal chemistry publications and patents from the 1980s on sertraline analogs disclosed some of the first examples of TRIs. While sertraline had a (1*S*,4*S*)-*cis* configuration and shows selectivity for the serotonin transporter, the corresponding (1*R*,4*S*)-*trans* compound showed potent in vitro inhibition of the serotonin, norepinephrine and dopamine transporters in synaptosomal preparations of rat brain.[6] The TRI theory was further reduced to practice with DOV-21,947[7] and DOV-216,303,[8] which both potently inhibited the three monamine transporters (IC_{50} for 5-HT, NE and DA for DOV-21,947 = 12, 23, 96 nM and for DOV-216,303 = 14, 20, 78 nM, respectively), and dose-dependently reduced the duration of immobility in the forced swim and tail suspension tests in rats, two models that are highly predictive of antidepressant efficacy in humans (Fig. 1).

The body of clinical data for triple reuptake inhibitors is expanding, but has thus far produced mixed results. DOV Pharmaceuticals produced the first and only positive Phase II clinical result for a TRI in 2005. DOV-216,303 was shown in a multicenter, Phase II study of patients with moderate to severe major depressive disorder ($n = 67$) to be safe and as effective as citalopram.[9]

In 2009 GSK announced the results of two 10 week, placebo and active controlled studies ($n = 396$ and 504 patients) of their balanced TRI (equally potent on SERT, NET and DAT) GSK-372,475

* Corresponding author. Tel.: +1 508 357 7468; fax: +1 508 490 5454.
E-mail address: liming.shao@sepracor.com (L. Shao).
† Present address: Evotec, Schnacokenburgallee 114, Hamburg 22525, Germany.
‡ Present address: Cortex Pharmaceuticals, 15231 Barranca Parkway, Irvine, CA 92618, USA.

0968-0896/$ - see front matter © 2010 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmc.2010.10.034

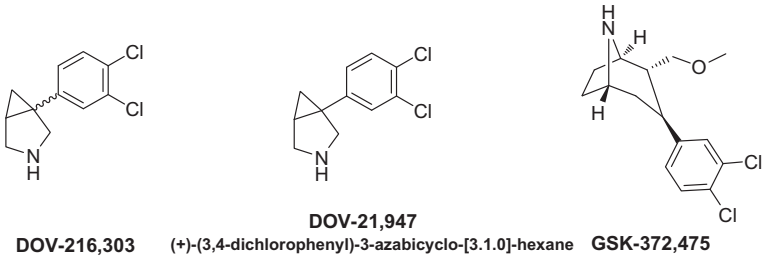


Figure 1. Structures of DOV-216,303, DOV-21,947 and reported structure of GSK-372,475.

(Fig. 1) in patients with major depressive disorder.[10] Neither study demonstrated a significant benefit for GSK-372,475 versus placebo, in comparison to a statistically significant improvement in MDD symptoms for both active controls (venlafaxine and paroxetine). The hypothesis of the investigators was that the onset of TRI related side effects thwarted the potential for observing efficacy. Also in 2009, Sepracor announced that their TRI SEP-225,289 did not meet its primary efficacy endpoint, reduction in symptoms of depression following 8 weeks of treatment, in a Phase II, 514 patients study. Positive control venlafaxine did separate from placebo and achieve a reduction in the symptoms of depression as judged by the 17 item HAM-D scale.[11] In this study, the measured serum concentrations of SEP-225,289 were far below expected levels of exposure for both doses studied and were well below exposure profiles observed in several Phase I studies. In addition, the side effect profile seen in the treatment group was similar to the placebo group and inconsistent with prior clinical observations. While the clinical data from GSK and Sepracor were disappointing with respect to advancement of a TRI to the marketplace, the results highlighted the difficulties inherent in fine-tuning three different neurotransmitters to create a clinically effective triple reuptake inhibitor. Significant challenges remain in the discovery of novel compounds that modulate 5-HT, NE and DA levels to varying degrees. The present work discloses our efforts to develop novel TRIs with variable levels of reuptake inhibition at the three neurotransmitters.

While *trans*-sertraline (**1**) and congener *cis*-sertraline (Zoloft) are known to be monoamine reuptake inhibitors (vida infra), novel chiral amines **2**, **3** or **4** had never, to our knowledge, been reported as monoamine reuptake inhibitors (Fig. 2). Our goal was to probe the SAR of these novel chiral amines as potential TRIs and create novel chemotypes that possessed varying degrees of reuptake inhibition at SERT, NET and DAT. The synthesis, detailed SAR of the scaffolds and their inhibition against SERT, NET and DAT are detailed below.

2. Results and discussion

2.1. Chemistry

Synthesis of the initial set of chiral amines (Scheme 1) began from commercially available α-tetralone **5**, which was reduced with sodium borohydride and dehydrated with H_2SO_4 to give alkene **6**. Dihydroxylation and treatment with *p*-TsOH gave the key β-tetralone intermediate **8**. Reductive amination provided amines **9** and **10**; Eschweiler–Clarke[12] alkylation of secondary amine **10** provided tertiary amine **11**.

The synthesis of the second set of amines also commenced from tetralone **5**, which was acylated with diethylcarbonate to give malonate derivative **12**, then reduced with triethylsilane/TFA to give ester **13** (Scheme 2). Reduction with LAH was followed by bromination (to **15**) and displacement with sodium azide to give **16**. Primary amine **17** was obtained as a mixtures of diastereomers after hydrogenation of the azide with Pd/C. The mono (**18**) and di-methyl amine (**19**) derivatives were synthesized via SN2 displacement of the alkyl bromide with methyl and dimethyl amine solutions, respectively.

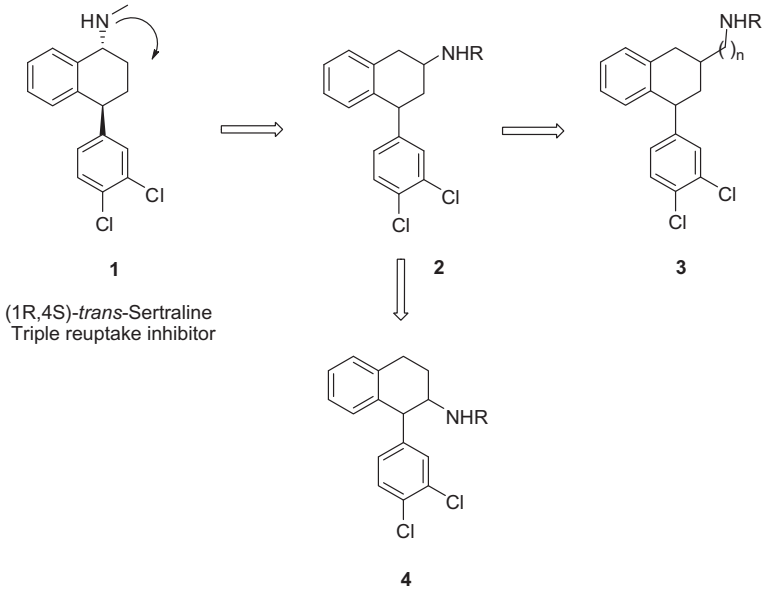
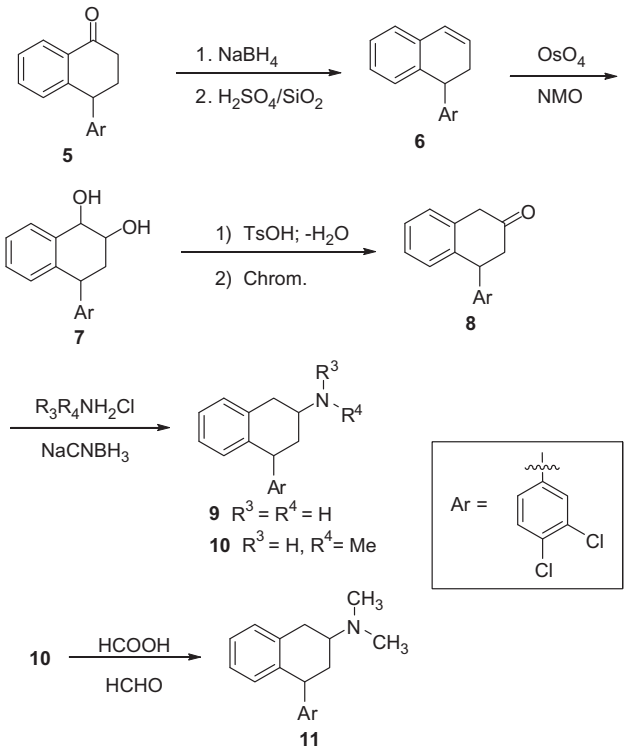


Figure 2. Novel chiral amines **2**, **3** and **4**.



Scheme 1. Synthesis of chiral amines **9**–**11**.

Assignments of relative stereochemistries were made using NMR techniques (determination of *cis*- and *trans*-configurations, optionally using literature reports for similar compounds). Absolute stereochemistries of selected compounds were determined by synthesis of key intermediates from commercially-available (*S*)-α-tetralone as outlined in Scheme 3, below. Correlations were made using chiral HPLC analyses. For example, authentic samples could be spiked into enantiomeric and/or diastereomeric mixtures to allow for a correlation of retention times and structures. Using the synthetic methods outlined in Scheme 3 and described above, the absolute configuration of compounds **9a–d, 10a–d**, **11a–d**, **17a–d** and **18a–d** (Schemes 3 and 4) could be determined.

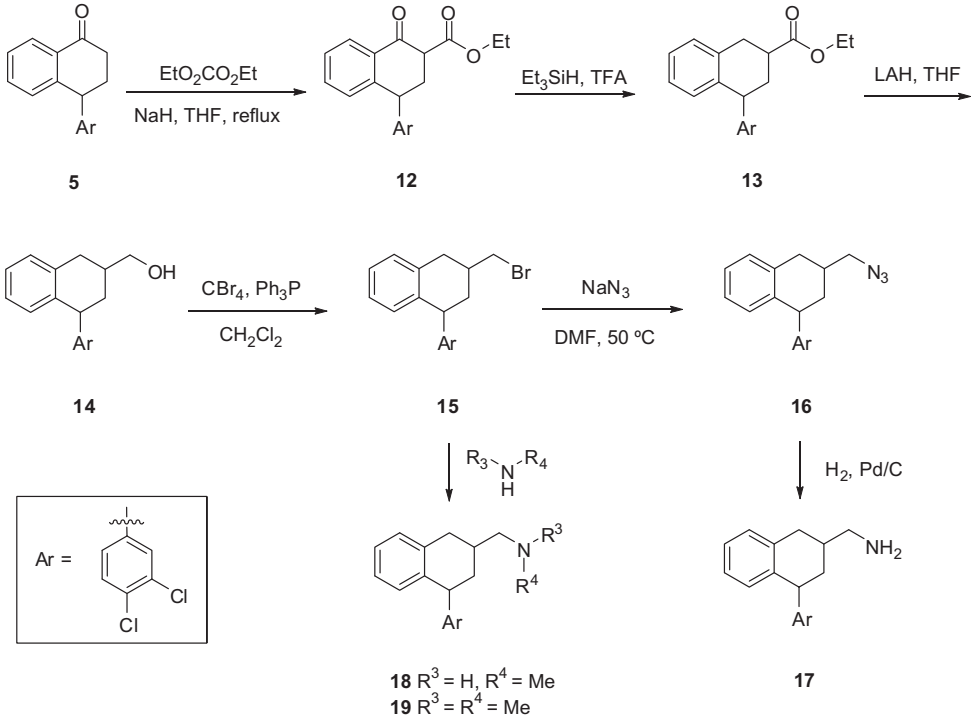
We also chose to evaluate the one-carbon extended version of compounds **17**–**19**, with an ethylene linker between the tetralone and amine moieties (Scheme 5). Starting from the same β-tetralone **8**, we carried out a Wittig reaction with dimethyl cyanomethylphosphonate, followed by hydrogenation and reduction to give racemic **21**. Installation of a BOC group allowed chiral separation to be carried out (Chiral Technologies OD column and a solvent system of 90% hexanes/10% IPA/0.1% diethylamine); removal of the BOC group gave single enantiomer (*cis*) compounds **21a** and **21b**.

Ketone **22** was the starting point for the synthesis of the final set of chiral amines **24a**–**d** (Scheme 6). Pd-catalyzed arylation with 1-bromo-3,4-dichlorobenzene and reductive amination with methyl amine hydrochloride and sodium cyanoborohydride delivered the crude target compounds, which could be resolved by chiral HPLC with the OD column and two different solvent systems (see Supplementary data for details). The relative (*cis*/*trans*) configurations were determined based on ^{1}H NMR coupling patterns and by comparison to reported values for similar compounds.[13] The absolute configurations of **24a**–**d** were assigned arbitrarily.

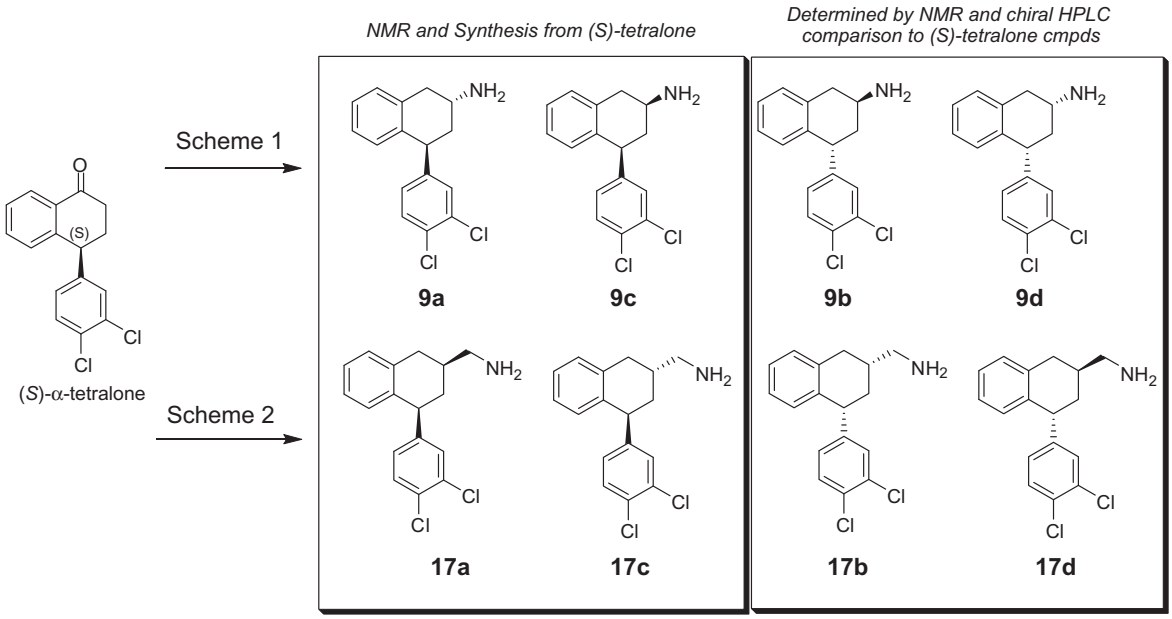
2.1.1. In vitro pharmacology

Table 1 details the pharmacological characterization of our novel triple reuptake inhibitors – in vitro data on inhibition of human recombinant serotonin,[14] norepinephrine[15] and dopamine[16] transporters.

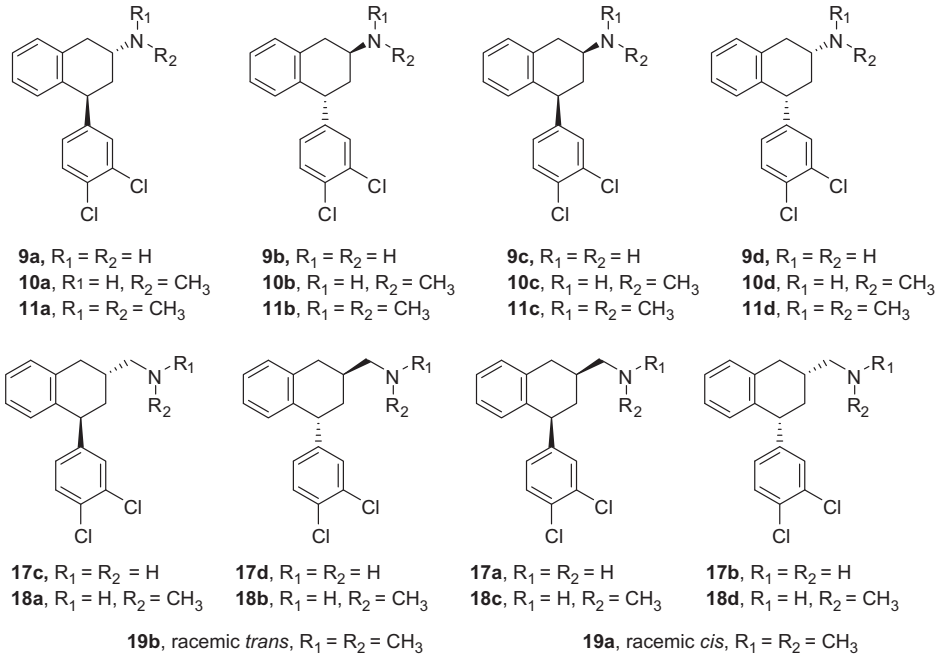
For the first set of chiral amines (**9a**–**d**, **10a**–**d** and **11a**–**d**), potency against SERT increased as the substitution on the amine increased, with *trans* mono-methyl amines **10a** (IC_{50} for 5-HT, 6 nM) and cis dimethyl amine **11c** (3 nM) as standouts. For NET, mono-methyl amines such as **10a** (IC_{50} for NE, 27 nM), **10c** (IC_{50}



Scheme 2. Synthesis of amines **17**–**19**.



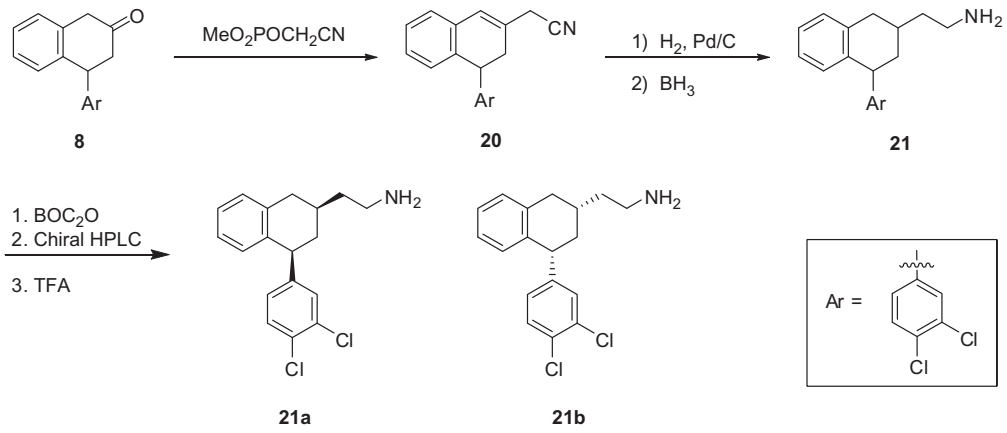
Scheme 3. Synthesis of **9a**, **9c**, **17a** and **17c** using (*S*)-α-tetralone starting material, and determination of configuration of **9b**, **9d**, **17b** and **17d** by analogy and comparison via chiral HPLC.



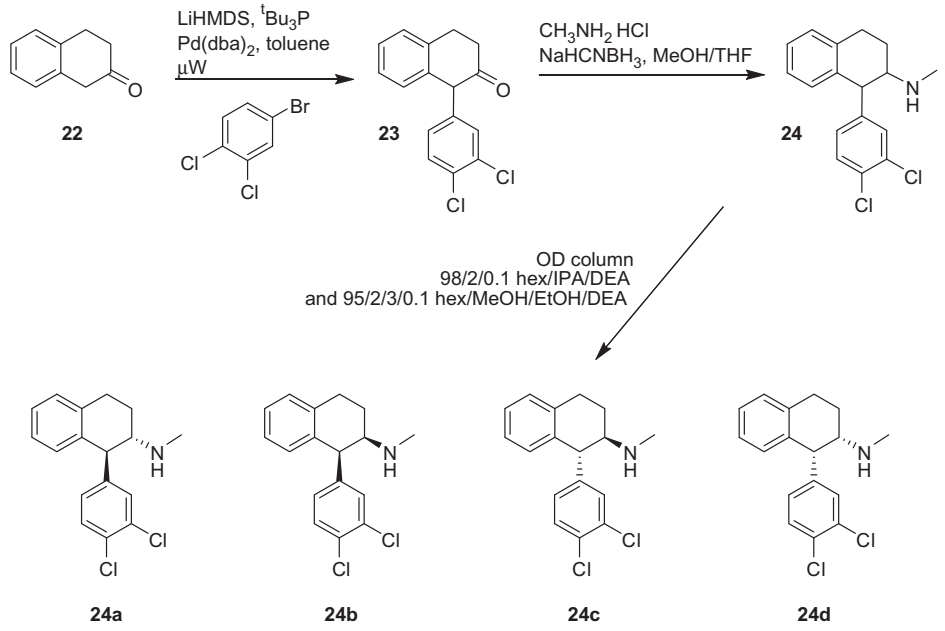
Scheme 4. Absolute configuration of amines **9**, **10**, **11**, **17** and **18a–d** and relative configuration of racemic compounds **19a–b**.

for NE, 45 nM) and **10d** (IC_{50} for NE, 73 nM) showed good potency, while the primary amines provided some of the least potent compounds in the initial set with respect to NET (i.e., compounds **9b** and **9c**). Mono-methyl amines such as **10b** (IC_{50} for DA, 62 nM) and **10d** (IC_{50} for DA, 72 nM) provided good potency against DAT; the primary and tertiary amines were generally worse than the secondary amines against DAT for the first set of compounds. In terms of triple reuptake inhibitor development potential, compound **10a** and **10c** showed the most potential, with excellent potency against SERT and NET and slightly decreased potency against DAT. Clinically this might be a benefit in that patients would achieve symptom relief without a risk of dependence.

There was a pronounced effect of chirality at the dichlorophenyl aromatic position with respect to SERT reuptake inhibition in the first series of compounds. Compounds derived from (*S*)-tetralone showed potent inhibition at SERT compared with (*R*)-tetralone derived analogs, regardless of substitution of the amine portion of the compound and cis/trans configuration of the amine and dichlorophenyl moieties. The clear preference for the (*S*)-derived compounds at SERT was quite striking and must be due to the serotonin transporter having a binding pocket that was nicely occupied by the (*S*)-dichlorophenyl tetralone pharmacophore. This result was also consistent with what the Pfizer group found in their investigations of sertraline. Only the *cis*-(1*S*,4*S*) and *trans*-(1*R*,4*S*)



Scheme 5. Synthesis of amino-ethyl derivatives **21a** and **21b**.



Scheme 6. Synthesis of δ-tetralones **24a–d**.

compounds showed reuptake inhibition activity.[6a] The (*R*)-tetralone derived compounds showed moderately increased inhibition of the dopamine transporter compared with their (*S*)-tetralone derived congeners, although the effect was less pronounced than the SERT effect described above. The influence of chirality on NET was less clear in the first series of compounds, as was any preference for a *cis* or *trans* relationship between the dichlorophenyl and amine moieties (compare *trans*-**10a** to *cis*-**10c**).

For the amino-methyl derivatives **17–19**, the primary and secondary amines exhibited excellent potency, with a number of analogs active against all three transporters. The most potent SERT inhibitors were found among the primary amines, with **17a** (IC_{50} for 5-HT, 2 nM) and **17c** (IC_{50} for 5-HT, 1 nM) in the low single digits. The secondary amine **18a** was also among the best SERT inhibitors (IC_{50} for 5-HT, 9 nM). NET inhibition was more varied, with both primary amine **17a** (IC_{50} for NE, 28 nM) and secondary amine **18a** (IC_{50} for NE, 72 nM) providing compounds with potencies <100 nM. For the dopamine transporter, the most potent inhibitors were found amongst the primary amines of the second set, with **17a** and **17c** as standouts. As for TRI potential, primary amine **17a** and **17c** and secondary amine **18a** looked the best, with serotonin and norepinephrine reuptake more potent than dopamine reuptake inhibition in vitro. Methylene extended compounds **21a** and **21b** were both potent inhibitors against the three transporters, although **21a** was superior across all three transporters.

For the amino-methyl series (compounds **17–19**), the chirality at the dichlorophenyl attachment showed a significant impact on SERT activity with analogs derived from (*S*)-tetralone being more potent against SERT, and similar results were also obtained for potency against NET for primary and secondary amines (i.e., compounds **17a–d** and **18a–b**). For potency against all three transporters, primary amine **17a** was the best compound, while secondary amines such as **18a** showed ratios that might be more favorable clinically to avoid risks of dependence while providing maximum amounts of postsynaptic serotonin and norepinephrine.

The δ-tetralone compounds **24a–d** did not show potent inhibition against all three transporters in general, with only compound **24a** showing potential as a selective sertotonin and dopamine

Table 1

Compound	IC_{50}[a] (nM)		
	5-HT	NE	DA
Sertraline (zoloft®)[21]	3	825	310
1 (*trans*-sertraline)[6a]	50[b]	22[b]	60[b]
9a	46	124	350
9b	1830	731	408
9c	84	855	894
9d	108	174	175
10a	6	27	114
10b	125	117	62
10c	8	45	281
10d	107	73	72
11a	7	167	454
11b	108	174	176
11c	3	164	273
11d	20	98	319
17a	2	28	11
17b	19	257	111
17c	1	92	45
17d	61	371	92
18a	9	72	125
18b	54	126	103
18c	23	210	111
18d	16	372	484
19a mixture of *cis*-enantiomers	311	565	332
19b mixture of *trans*-enantiomers	970	309	339
21a	1	26	32
21b	79	158	50
24a	42	766	91
24b	1956	112	559
24c	2495	1642	212
24d	2827	7262	1616

[a] SERT, NET and DAT reuptake inhibition assays were run with at least $n = 2$ and five concentrations to generate inhibition curves, from which IC_{50}'s were determined. Assay performance was monitored by the use of SERT reference compound fluoxetine ($IC_{50} = 5.2 \pm 0.6$ nM), NET reference compound nisoxetine ($IC_{50} = 8.3 \pm 1.0$ nM) and DAT reference compound nomifensine ($IC_{50} = 30.2 \pm 2.2$ nM).

[b] IC_{50} values for *trans*-sertraline were for inhibition of monoamine uptake into rat brain synaptosomes. Sertotonin and dopamine taken from corpus striatum, norepinephrine from hypothalamus.

reuptake inhibitor (SDRI). Moving the amino group to the C-3 position on the tetralone scaffold appeared to be negative with respect to broad activities against the three transporters.

2.1.2. In vitro ADME: microsomal stability, CYP and hERG inhibition

The secondary amines **10a**–**10d**, **18a**–**18c**, racemic primary amines **21a** and **21b** and δ-tetralones **24a–d** were chosen for profiling in CYP and hERG inhibition and microsomal stability assays based on their potency against the three transporters. All compounds profiled showed exceptional stability in human and mouse liver microsomes, with $t_{1/2}$ exceeding 300 min in almost every case. In the CYP inhibition assay, the compounds began to differentiate; significant (IC_{50} <1 μM) CYP2D6 inhibition was seen for compounds **10a**, **10c**, **18a–c**, and **21a** and potent inhibition for **24a–d** (Table 2). We were not that surprised by the CYP2D6 inhibition for our test compounds—structurally related *cis* sertraline shows mild inhibition of CYP2D6 and can result in a 10–50% elevation of plasma levels of a co-administered CYP2D6 substrate such as dextromethmorphan.[17] There was also a clear correlation between CYP2D6 inhibition and the configuration of the dichlorophenyl aromatic moiety for compounds **10a–d**, which were most closely structurally related to sertraline. Compounds **10a** and **10c**, with an (*S*)-configuration of the dichlorophenyl aromatic (like sertraline and *trans*-sertraline), showed significant (nanomolar) 2D6 inhibition, while the (*R*)-configured congeners **10b** and **10d** showed micromolar inhibition. For the other series of compounds it was more difficult to correlate the CYP2D6 inhibition with any one structural feature. The profiled compounds showed minimal inhibition against CYP1A, 2C9 and 3A4, although **18b** and **21b** exhibited some CYP2C19 inhibitory activity (IC_{50} ~2 μM). Several of the compounds also had significant hERG inhibition; **10b**, **10d**, **18a** and **18**c all had IC_{50}'s around 2 μM and could pose a concern from a safety perspective. When the in vitro ADME data were viewed as a whole, **10d** and **21b** stood out as having the best profiles. Although both showed some CYP inhibition (CYP2C19 inhibition for **21b** and CYP2D6 for **10d**) and both also showed moderate hERG inhibition, the liabilities were not viewed as fatal flaws. The potent reuptake inhibition for the two compounds could provide a reasonable therapeutic index and might make it possible to avoid concentrations in vivo that could make the CYP and hERG inhibition a safety concern. With this in mind, compound **10d** was chosen as the better of the two based on potency against the three transporters and profiling in the mouse tail suspension test[18] was initiated. The assay is not a model of depression, but is sensitive to the effects of several classes of antidepressants, including tricyclics, SSRIs and SNRIs.

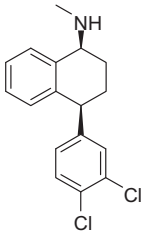
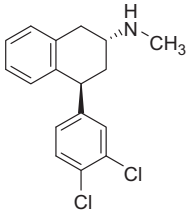
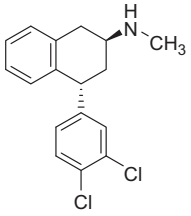
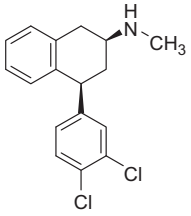
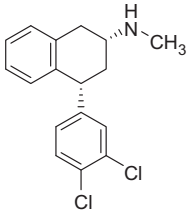
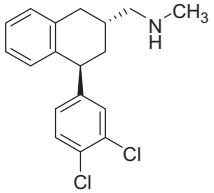
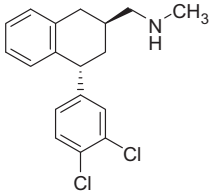
2.1.3. Pharmacology

The HCl salt of **10d** was prepared and dosed 3, 10 and 30 mg/kg po in male mice in the tail suspension test (TST). A 120 min pretreatment time was used based on exposure work prior to the assay which showed maximal brain levels 120 min after po dosing. **10d** showed a nice dose dependent reduction in immobility, which was statistically significant compared to vehicle at 10 and 30 mg/kg doses (Fig. 3a). The positive control, desipramine, also showed a statistically significant reduction in immobility at 100 mg/kg po. Brain and plasma samples taken from the testing animals showed that the brain to plasma ratio was close to 40 at the 30 mg/kg dose, with almost 44 μM of **10d** in the brain (Table 3). Even given the high degree of protein binding we measured for compound **10d** (99% protein binding using mouse plasma protein), the brain free fraction for compound **10d** was estimated to be ~440 nM, which exceeded the IC_{50} at all three transporters. Plasma protein binding can be a surrogate for brain binding if the compound freely diffuses into the brain and no efflux is present (no efflux as judged by A→B/B→A permeability in the CaCO-2 cell line was seen in the compound series, data not shown).[19] The effect of compound **10d** (Fig. 3b), in the TST was also not due to a general locomotor activation effect; the compounds did not significantly increase spontaneous locomotor activity in vivo in the first 5 min at the 10 or 30 mg/kg dose. The 5 min time point was significant because that is the amount of time the compounds were evaluated in the TST.

3. Conclusions

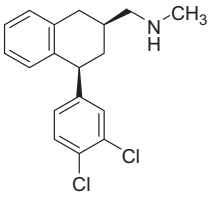
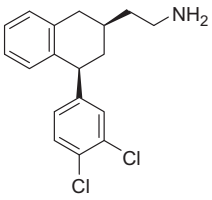
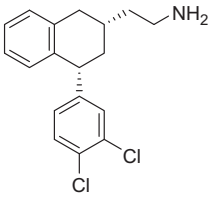
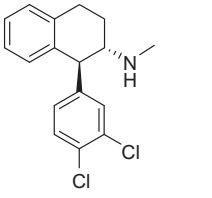
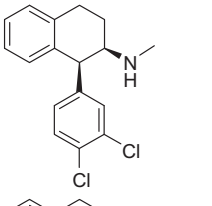
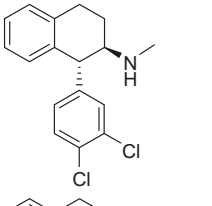
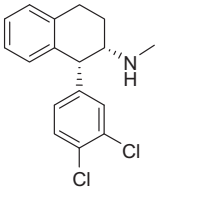
The goal of the present work was to explore the potential of novel triple reuptake inhibitors as treatments for depression. We succeeded in that goal by showing a wide range of chiral amines with potent reuptake inhibition at SERT, NET and DAT. CYP and hERG inhibition emerged as a problem area for the designed compounds, with most of the compounds showing potent inhibition of CYP2D6 and several showing IC_{50}'s <2 μM for the hERG potassium channel. Compound **10d** was shown to be a promising candidate for further optimization due to its potent inhibition of SERT, NET and DAT, good stability in vitro and relatively clean profile against the CYPs and hERG. In vivo testing of

Table 2
In vitro microsomal stability, CYP and hERG inhibition

Structure	Compound	Microsomal stability $t_{1/2}$ (min)		CYP450 inhibition IC_{50} (μM)					*h*ERG IC_{50} (μM)
		Human	Mouse	1A	2C9	2C19	2D6	3A4	
	**Sertraline**	>120	nt[a]	>10	>10	0.31	1.4	0.8	nt
	**10a**	>300	>300	>25	>25	>25	0.067	>25	nt
	**10b**	>300	120	>25	>25	5.26	1.16	>25	2.32
	**10c**	>300	>300	>25	>25	>25	0.020	>25	5.27
	**10d**	>300	>300	>25	>25	10.15	2.78	17.1	1.60
	**18a**	>300	>300	>25	>25	7.69	0.041	20.1	1.61
	**18b**	>300	93	>25	>25	2.17	0.917	>25	nt

(*continued on next page*)

Table 2 (*continued*)

Structure	Compound	Microsomal stability $t_{1/2}$ (min)		CYP450 inhibition IC_{50} (μM)					*h*ERG IC_{50} (μM)
		Human	Mouse	1A	2C9	2C19	2D6	3A4	
	**18c**	>300	>300	>25	>25	6.02	0.154	>25	2.0
	**21a**	>300	>300	>25	>25	3.76	0.265	>25	13.1
	**21b**	>300	>300	20.7	>25	1.64	13.8	>25	1.51
	**24a**	106	101	nt	>10	nt	0.008	nt	nt
	**24b**	55	55	nt	7.6	nt	0.033	nt	nt
	**24c**	95	20	nt	>10	nt	0.057	nt	nt
	**24d**	55	6	nt	>10	nt	0.495	nt	nt

Human microsomal stability data for sertraline taken from Ref. 17. CYP inhibition for sertraline was determined at CEREP, using assay codes 900-1 (1A2), -4 (2C9), -6 (2D6), -2 (2C19) and -3 (3A4). Microsomal stability, CYP and hERG inhibition for all compounds other than sertraline determined at Cyprotex using their standard protocols, with midazolam as the reference substrate for CYP3A4.

[a] nt, not tested.

compound **10d** in the mouse tail suspension test confirmed its potential as an antidepressant; the compound was active at 10 and 30 mg/kg po and its activity was not due to a general locomotor activating effect. Given that compound **10d** was not the most potent triple reuptake inhibitor we disclosed in this paper, and yet was still orally active in vivo in a rodent model of depression, we feel the triple reuptake inhibition landscape is tremendously fertile with opportunities for development of antidepressants. Future publications will disclose our efforts along those lines.

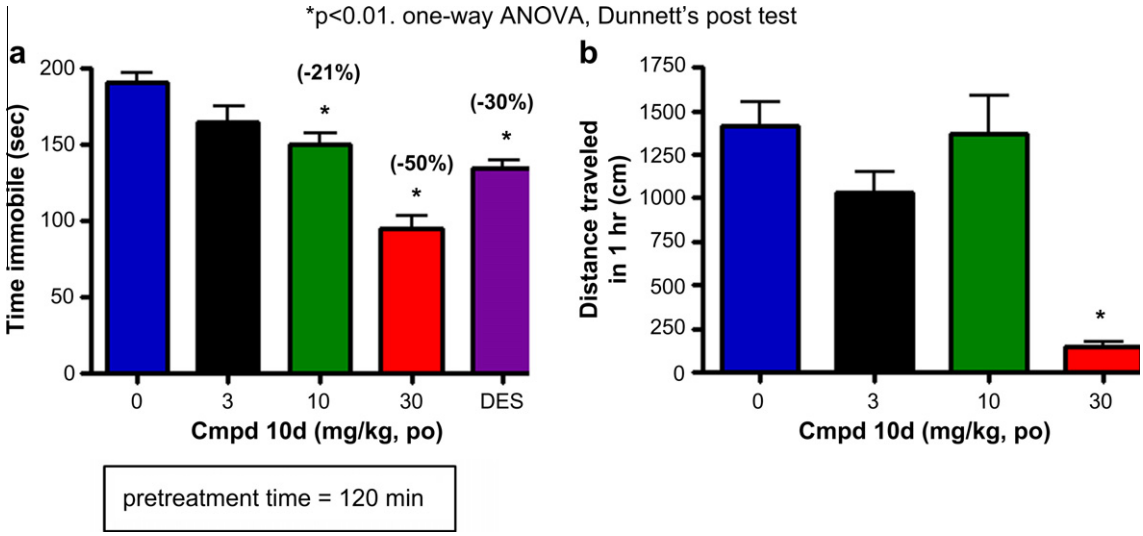


Figure 3. (a) Compound **10d** in the mouse TST at 3, 10 and 30 mg/kg po. (b) Compound **10d** in mouse locomotor activity assay at 5 min time point, at 3, 10, and 30 mg/kg po.

Table 3
Whole brain and plasma levels of compound **10d** from TST animals

Compound	Dose (mg/kg), po	Plasma levels (ng/mL)	Brain levels (ng/g)
10d	3	96	2830
	10	164	6021
	30	344	13,571

4. Experimental section

4.1. Chemistry

4.1.1. General statement

Reagents were commercial grade and were used as received unless otherwise noted. Reactions were run with anhydrous solvents under an atmosphere of nitrogen unless otherwise noted. Flash column chromatography was done using an Isco® purification system on silica gel columns. The structures of all new compounds were consistent with their ^{1}H, ^{13}C NMR and mass spectra and were judged to be >95% pure. Purified chiral compounds were all >95% enantiomeric excess (ee). Chiral chromatography was done using the designated Chiral Technologies Chiracel 20 micron analytical (0.46 cm ID × 25 cm L) column, a flow rate of 1 mL/min with the designated solvent, with detection by UV at 220 and 254 nM. LC–MS was performed on an Agilent 1100 Series system connected to a Micromass Platform LC. GC–MS was performed on a Hewlett Packard 6890 Series GC System with an HP1 column (30 m, 0.15 μ film thickness) coupled to a Hewlett Packard 5973 Series Mass Selective Detector. Accurate mass measurements were carried out on a Waters Q-TOF micro system.

4.1.1.1. Synthesis of 4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalen-1-ol. To a stirring mixture of α-tetralone 4-(3,4-dichloro-phenyl)-3,4-dihydro-2*H*-naphthalen-1-one **5** (53 g, l82 mmol) in methanol (400 mL) was added sodium borohydride (12 g) in portions. The mixture was stirred at ambient temperature for 3 h. Water was added and the volatile components were removed in vacuo. The aqueous remainder was extracted with ethyl acetate. The organic phase was separated, washed with water, dried (Na_2SO_4), and evaporated to dryness to yield the crude mixture of diastereomeric alcohol 4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydro-naphthalen-1-ol (53 g). ^{1}H NMR ($CDC1_3$) δ 7.58–6.8 (m, 7H), 4.84 (m, 1H), 4.15 (t) and 3.92 (m, total 1H), 2.38 (m) and 2.2–1.9 (m, total 3H), 1.8 (m, 1H). ^{13}C NMR ($CDC1_3$) δ 146.9, 146.8, 139.6, 138.9, 138.3, 137.7, 132.3, 132.2, 130.6, 130.5, 130.3, 130.2, 129.8, 129.7, 129.0, 128.2, 128.1, 128.1, 128.0, 127.9, 127.1, 127.0, 68.1, 67.7, 45.0, 44.4, 30.0, 29.9, 28.9, 28.1.

4.1.1.2. Synthesis of 1-(3,4-dichlorophenyl)-1,2-dihydronaphthalene (6). To a solution of the crude alcohol 4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydro-naphthalen-1-ol (53 g) in toluene (500 mL) was added silica gel coated with sulfuric acid (3%, 14 g). The mixture was heated to 100 °C and monitored by TLC (prod R_f (25% EA/hex) = 0.58). After 3 h, the mixture was filtered. The organic phase was washed with water and sodium bicarbonate solution, dried (Na_2SO_4), and evaporated to give the alkene **6** (42 g, 84%) as a pale-brown solid. GC–MS R_t = 13.55 min, m/z = 274 (M+). ^{1}H NMR ($CDCl_3$) δ 7.4–6.7 (m, 7H), 6.54 (d, J = 9.6 Hz, 1H), 6.0 (m, 1H), 4.08 (t, J = 8.0 Hz, 1H), 2.7 (m, 1H), 2.5 (m, 1H). ^{13}C NMR ($CDCl_3$) δ 143.6, 136.3, 133.8, 132.2, 130.2, 130.2, 128.1, 127.7, 127.2, 126.4, 129.7, 127.4, 42.8, 31.6.

4.1.1.3. Synthesis of 4-(3,4-dichlorophenyl)-3,4-dihydro-1*H*-naphthalen-2-one (8). To a stirring solution of the alkene **6** (3 g, 10.8 mmol) in acetone (40 mL) was added NMO (2 g, 1.6 equiv) and water (10 mL). After the NMO dissolved, osmium tetroxide (1.3 mL, 0.1 M in toluene, 5 mol %) was added and the solution was stirred at ambient temperature for 40 min. Sodium bisulfate (10 mL, 10% solution in water) was added and the mixture was stirred for an additional 30 min. After this time, the solvent was removed in vacuo and the resultant oily solid was partitioned between MTBE and, sequentially, water and brine. The organic solvent was evaporated to yield the crude diol (3.6 g) as a brown glass. TLC R_f (50% EA/hex) = 0.14. The crude diol (**7**) was sufficiently pure for the next step, and could be confirmed by the three diagnostic peaks that were discernable in the ^{1}H NMR (4.8, 4.4, 4.2 ppm).

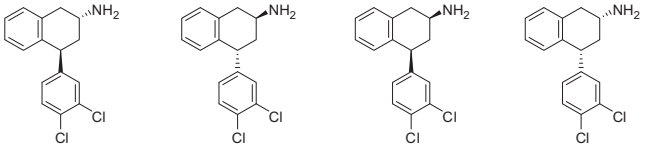
The diol **7** was dissolved in toluene (200 mL). Tosic acid (600 mg, 30 mol %) was added and the solution was heated to reflux in a Dean–Stark trap until the diol was consumed. After 3 h, the reaction mixture was cooled and most of the toluene was removed. The remaining liquid was partitioned between MTBE and, sequentially, 10% aqueous KOH, water, and brine. The organic layer was evaporated and the crude green oil was separated on silica gel to give the β-tetralone **8** (1.46 g, 46%) as a pale-yellow oil. GC–MS R_t = 13.54 min, m/z = 290 (M+). ^{1}H NMR ($CDCl_3$) δ 7.39 (d, J = 8.0 Hz, 1H), 7.3–7.2 (m, 4H), 7.0–6.9 (m, 2H), 4.42 (t,

J = 6.4 Hz, 1H), 3.62 (q, J = 20 Hz, 2H), 2.9 (m, 2H). ^{13}C NMR ($CDCl_3$) δ 208.2, 141.7, 137.7, 133.0, 132.8, 131.0, 130.6, 129.8, 128.7, 127.8, 127.5, 127.1, 45.4, 44.5, 43.8.

4.1.1.4. Synthesis of [4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalen-2-yl]-amine (9a–d). Ammonium chloride (643 mg, l0 equiv) was dissolved in methanol (24 mL) by heating to 50 °C. After it cooled, a solution of ketone **8** (350 mg, 1.202 mmol) in THF (18 mL) was added followed by sodium cyanoborohydride (6.0 mL, 5 equiv). The mixture was heated in a 50 °C oil bath overnight. The reaction was then cooled, quenched with aqueous sodium bicarbonate, and extracted with MTBE. The combined organic layer was washed with brine and evaporated to give a brown-green oil. The oil was separated on silica gel to give the primary amine **9** (145 mg, 41%) as a pale-green oil.

As isolated, the amine was a mixture of four stereoisomers which were separable using chiral columns. First, the mixture was separated on a Chiracel OD column (90:10:0.1 Hex/IPA/DEA) to give three fractions. Symchiral *trans* (compound **9a** at 11.9 min, racemic *cis* at 14.7 min, and symchiral *trans* (compound **9b**) at 22.3 min. The racemic *cis* was then resubmitted to the Chiracel AD column 95:2:3:0.1 Hex/MeOH/EtOH/DEA) to give the symchiral *cis* (Compound **9c**) at 11.1 min and symchiral *cis* (compound **9d**) at 13.9 min. Retention times are summarized in Table 4, below.

Absolute stereochemistries for compounds **9a–d** were determined using a combination of NMR techniques (determination of *cis*- and *trans*-configurations) and chiral HPLC analyses using authentic samples, which were prepared from commercial (*S*)-α-tetralone as described above. The resulting structures indicating absolute stereochemistries are shown below:



9a **9b** **9c** **9d**

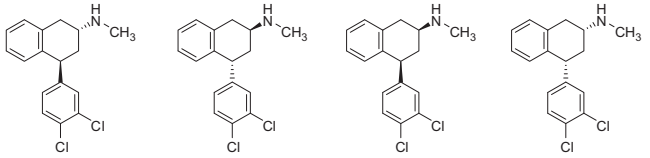
*4.1.1.4.1. trans-Isomers 9a and **9b**.* GC–MS R_t = 13.52 min, m/z = 291 (M+). ^{1}H NMR ($CDC1_3$) δ 7.4–6.8 (m, 7H), 4.32 (t, J = 5.4 Hz, 1H), 3.3 (m, 1H), 3.17 (dd, J = 4.9, 16.3 Hz, 1H), 2.7 (m, 1H), 2.1 (m, 2H). ^{13}C NMR ($CDC1_3$) δ 160.3, 147.2, 136.0, 135.2, 132.3, 130.5, 130.1, 130.0, 129.5, 128.0, 126.9, 126.5, 43.2, 42.9, 40.2, 38.2.

*4.1.1.4.2. cis-Isomers 9c and **9d**.* GC–MS R_t = 13.61 min, m/z = 291 (M+). ^{1}H NMR ($CDC1_3$) δ 7.4–6.7 (m, 7H), 4.11 (dd, J = 5.5, 12.1 Hz, 1H), 3.26 (ddt, J = 3.1, 4.9, 11.3 Hz, 1H), 3.07 (ddd, J = 2.2, 4.8, 15.9 Hz, 1H), 2.7 (m, 1H), 2.2 (m, 1H), 1.6 (m, 1H). ^{13}C NMR ($CDC1_3$) δ 146.7, 137.8, 135.9, 132.4, 130.6, 130.5, 129.1, 129.1, 128.1, 126.5, 126.2, 130.2, 47.6, 46.0, 44.4, 40.2.

4.1.1.5. Synthesis of 4-(3,4-dichlorophenyl)-*N*-methyl-1,2,3,4-tetrahydronaphthalen-2-amine (10a–d). To a solution of ketone **8** (350 mg, 1.202 mmol) in THF (18 mL) and methanol (24 mL) was added methylamine hydrochloride (980 mg, 10 equiv). After the solid dissolved, sodium cyanoborohydride (6.0 mL, 1 M in THF, 5 equiv) was added in one portion. The mixture was heated in a 50 °C oil bath overnight before being quenched with aqueous sodium bicarbonate and extracted with MTBE. The combined organic layer was washed with brine and evaporated to give a brown-green oil. The oil was dissolved in MTBE and extracted into 10% aqueous hydrochloric acid. The aqueous layer was basicified with KOH and extracted with MTBE. The volatile components were removed in vacuo and the crude green oil was separated on silica gel to give the methylamine (0.20 g, 54%) as a pale-green oil.

As isolated, the amine was a mixture of four stereoisomers which were separable on chiral columns. First, the mixture was separated on a Chiracel OD column (98:2:0.1 Hex/IPA/DEA) to give three fractions. Symchiral *trans* (compound **10a**) at 12.4 min, racemic *cis* at 15.8 and 17.6 min, and symchiral *trans* (compound 10b) at 29.7 min. The racemic *cis* was then resubmitted to a Chiracel AD column (98:2:0.1 Hex/IPA/DEA) to give the symchiral *cis* (Compound **10c**) at 20.2 min and symchiral *cis* (SME compound **10d**) at 27.7 min. Retention times are summarized in Table 5, below.

Absolute stereochemistries of compounds **10a–d** were determined using a combination of NMR techniques (determination of *cis*- and *trans*-configurations) and chiral HPLC analyses using authentic samples, which were prepared from commercial (*S*)-α-tetralone as described above. The resulting structures indicating absolute stereochemistries are shown below:



10a **10b** **10c** **10d**

*4.1.1.5.1. trans-Isomers 10a and **10b**.* HRMS [M+H]$^+$: calcd for (**10a**) $C_{17}H_{18}Cl_2N$ 306.0811, found 306.0832; LC–MS R_t = 8.3 min, m/z = 306 (M+1). GC–MS R_t = 13.64 min, m/z = 305 (M+). ^{1}H NMR ($CDC1_3$) δ 7.4–6.8 (m, 7H), 4.26 (t, J = 5.8 Hz, 1H), 3.15 (dd, J = 4.6, 16.2 Hz, 1H), 2.9 (m, 1H), 2.66 (dd, J = 7.8, 16.2 Hz, 1H), 2.43 (s, 3H), 2.0 (m, 2H. ^{13}C NMR ($CDC1_3$) δ 147.5, 136.8, 135.6, 132.2, 130.6, 130.1, 129.9, 129.8, 129.5, 128.1, 126.7, 126.2, 51.1, 42.5, 37.8, 36.0, 33.7.

*4.1.1.5.2. cis-Isomers 10c and **10d**.* HRMS [M+H]$^+$: calcd for (**10c**) $C_{17}H_{18}Cl_2N$ 306.0811, found 306.0822; LC–MS R_t = 8.5 min, m/z = 306 (M+1). GC–MS R_t = 13.82 min, m/z = 305 (M+). ^{1}H NMR ($CDC1_3$) δ 7.4–6.7 (m, 7H), 4.08 (dd, J = 5.4, 12.2 Hz,1H), 3.12 (ddd, J = 2.2, 4.7, 15.7 Hz, 1H), 2.93 (ddt, J = 2.9, 4.8, 11.2 Hz, 1H), 2.70 (dd, J = 11.1, 15.7 Hz, 1H), 2.52 (s, 3H), 2.3 (m, 1H), 1.6 (m, 1H). ^{13}C NMR ($CDC1_3$) δ 146.8, 138.2, 135.8, 132.4, 130.6, 130.5, 130.2, 129.2, 129.0, 128.1, 126.4, 126.1, 55.5, 45.8, 40.5, 37.4, 33.6.

4.1.1.6. Synthesis of [4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalen-2-yl]-dimethylamine (11a–d). The respective methylamine **10** (e.g., 28.4 mg, 0.0927 mmo1) was dissolved in 96% formic acid (0.5 mL) and 37% aqueous formaldehyde (0.5 mL) and heated at 100 °C for 2 h. After cooling, the solution was basicified (aq KOH) and extracted with MTBE. The organic phase was dried with sodium sulfate, filtered, and evaporated to give the dimethylamine (e.g., 27.1 mg, 93%) as a clear oil.

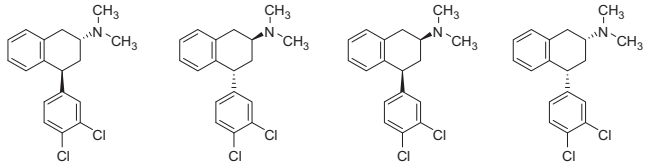
Absolute stereochemistries for compounds **11a–d** were determined and are shown below:

Table 4
Retention times for each diastereomer [min]

	9a *trans*	**9c** *cis*	**9d** *cis*	**9b** *trans*
HPLC R_t (Chiracel OD, 90:10:0.1 Hex/IPA/DEA)	11.9	14.7	14.7	22.3
HPLC R_t (Chiracel AD, 95:2:3:0.1 Hex/MeOH/EtOH/DEA)		11.1	13.9	

Table 5
Retention times for each diastereomer [min]

	10a *trans*	**10c** *cis*	**10d** *cis*	**10b** *trans*
HPLC R_t (Chiracel OD, 98:2:0.1 Hex/IPA/DEA)	12.4	15.8	17.6	29.7
HPLC R_t (Chiracel AD, 98:2:0.1 Hex/IPA/DEA)		20.2	27.7	



11a 11b 11c 11d

*4.1.1.6.1. trans-Isomers **11a** and **11b**.* LC–MS R_t = 9.0 min, m/z = 320 (M+l). ^{1}H NMR ($CDC1_3$) δ 7.4–6.8 (m, 7H), 4.32 (t, J = 5.4 Hz, 1H), 3.02 (dd, J = 4.8, 16.3 Hz, 1H), 2.84 (dd, J = 9.3, 16.3 Hz, 1H), 2.6 (m, 1H), 2.27 (s, 6H), 2.1 (m, 2H). ^{13}C NMR ($CDC1_3$) δ 147.3, 136.6, 136.3, 132.1, 130.5, 130.0, 129.8, 129.5, 128.1, 126.7, 126.2, 129.9, 56.0, 43.3, 41.9, 34.9, 32.1.

*4.1.1.6.2. cis-Isomers 11c and **11d**.* LC–MS R_t = 9.1 min, m/z = 320 (M+1). ^{1}H NMR ($CDC1_3$) δ 7.4–6.7 (m, 7H), 4.07 (dd, J = 5.3, 12.2 Hz, 1H), 3.1–2.9 (m, 2H), 2.80 (ddt, J = 2.5, 4.9, 11.4 Hz, 1H), 2.37 (s, 6H), 2.3 (m, 1H), 1.65 (q, J = 12.2 Hz, 1H). ^{13}C NMR ($CDC1_3$) δ 146.9, 138.0, 136.3, 132.4, 130.6, 130.5, 130.3, 129.5, 129.0, 128.1, 126.4, 126.1, 60.3, 46.4, 41.4, 36.8, 32.8.

4.1.1.7. Synthesis of 4-(3,4-dichlorophenyl)-1-oxo-1,2,3,4-tetrahydro-naphthalene-2-carboxylic acid ethyl ester (12). To a stirred suspension of NaH (60% dispersion in mineral oil, 1.69 g, 42 mmol) in THF (80 mL) under N_2 was added dropwise diethylcarbonate (4.85 mL, 40 mmol) at room temperature, followed by 4-(3′,4′-dichlorophenyl)-3,4-dihydro-l-(2*H*)-naphthalone **4** (5.82 g, 20 mmol) in THF (20 mL). The mixture was refluxed for 48 h, then cooled to 0 °C. Acetic acid (10 mL) was added dropwise, and the mixture was extracted with Et_2O. The Et_2O extracts were washed with saturated $NaHCO_3$ solution, brine, dried over $MgSO_4$, and evaporated. The residue was purified by chromatography, CombiFlash silica gel column (hexane/CH_2C1_2 = 50:50) to give compound **12** as a clear oil (5.81 g, 80%). ^{1}H NMR ($CDC1_3$) δ 1.30 (t, J = 7.2 Hz, 3H), 2.77 (dd, J = 16 Hz, 9.6 Hz, 1H), 2.91 (dd, J = 15.6 Hz, 6.4 Hz, 1H), 4.10 (dd, J = 12 Hz, 6.4 Hz, 1H), 4.19–4.30 (m, 2H), 6.87 (d, J = 6.8 Hz, 1H), 7.00 (dd, J = 8.4 Hz, 2.0 Hz, 1H), 7.27–7.36 (m, 5H), 7.89 (dd, J = 8.4 Hz, 2.0 Hz, 1H), 12.50 (s, 1H). ^{13}C NMR ($CDC1_3$) δ 14.5, 29.1, 43.4, 61.0, 95.6, 125.0, 127.6, 127.9, 128.1, 130.2, 130.6, 130.7, 131.0, 131.3, 132.8, 140.4, 143.9, 164.8, 172.6.

4.1.1.8. Synthesis of 4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydro-naphthalene-2-carboxylic acid ethyl ester (13). To a solution of **12** (2.81 g, 7.74 mmol) in TFA (30 mL) was added dropwise Et_3SiH (7.42 mL, 46.44 mmol) at 0 °C. Stirring was continued at 0 °C for 2 h. Then, the solvent was evaporated, and the residue was purified by chromatography, CombiFlash silica gel column, hexane/CH_2C1_2, CH_2C1_2 from 0% to 50%, to give compound **13** as a clear oil (mixture of *cis* and *trans* diastereomers, 2.63 g, 97%). ^{1}H NMR ($CDC1_3$) δ 1.18–1.34 (m, 3H), 1.88 (dd, J = 25.2 Hz, 12.4 Hz) and 2.14–2.19 (m, total 1H), 2.25–2.33 (m) and 2.43–2.55 (m, total 1H), 2.67–2.74 (m) and 2.82–2.92 (m, total 1H), 3.00–3.18 (m, 2H), 4.08–4.29 (m,, 3H), 6.72–7.42 (m, 7H).

4.1.1.9. Synthesis of [4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydro-naphthalen-2-yl]-methanol (14). A solution of **13** (2.55 g, 7.3 mmol) in THF (40 mL) was added dropwise to a stirring mixture of $LiAlH_4$ (0.304 g, 8.0 mmol) in THF (20 mL) at 0 °C. The resulting suspension was stirred at room temperature for 3 h, then, the mixture was cooled to 0 °C, and water (0.15 mL) was added dropwise to destroy the excess hydride. The mixture was filtered, and the solvent was evaporated in vacuo to give a colorless oil. The residue was purified by chromatography, CombiFlash silica gel column, MeOH/CH_2C1_2, MeOH from 0% to 3%, to give Compound **14** as a clear oil (mixture of *cis* and *trans* diastereomers, 1.80 g, 80%). ^{1}H NMR ($CDC1_3$) δ 1.31–1.54 (m, 1H), 1.92–1.98 (m, 1H), 2.10–2.26 (m, 1H), 2.54–2.71 (m, 1H), 2.92–3.03 (m, 1H), 3.53–3.75 (m, 2H), 4.07 (dd, J = 12 Hz, 5.2 Hz) and 4.25 (t, J = 3.6 Hz, total 1H), 6.72–7.38 (m, 7H). ^{13}C NMR ($CDC1_3$) δ 32.0, 32.5, 33.4, 34.6, 37.5, 37.6, 43.3, 46.3, 67.5, 67.9, 126.3, 126.4, 126.6, 127.0, 128.4, 128.5, 129.5, 129.7, 130.2, 130.5, 130.7, 130.9, 132.4, 132.7, 136.7, 136.9, 138.8, 147.5, 147.9.

4.1.1.10. Synthesis of 3-bromomethyl-l-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalene (15). To a solution of compound **14** (1.25 g, 4.07 mmol) and CBr_4 (2.33 G, 7.04 mmol) in CH_2C1_2 (15 mL) was added Ph_3P (1.82 g, 6.92 mmol) in CH_2C1_2 (15 mL) at 0 °C. The reaction was allowed to warm to room temperature overnight, then poured into water (40 mL), extracted with CH_2Cl_2 (75 mL), dried over Na_2SO_4, and the solvent was evaporated. The residue was purified by chromatography, CombiFlash silica gel column, EtOAC hexanes, EtOAc from 0% to 15%, to give compound **15** as a clear oil (mixture of *cis* and *trans* diastereomers, 1.50 g, 99%). ^{1}H NMR: ($CDC1_3$) δ 1.52–1.62 (m) and 1.97–2.15 (m, total 2H), 2.25–2.30 (m, 1H), 2.64–2.77 (m, 1H), 3.02–3.12 (m, 1H), 3.34–3.47 (m, 2H), 4.08 (dd, J = 12 Hz, 5.2 Hz) and 4.26 (t, J = 3.6 Hz, total 1H), 6.72–7.39 (m, 7H). ^{13}C NMR ($CDC1_3$) δ 31.8, 34.6, 35.6, 36.8, 37.2, 39.2, 39.3, 39.4, 43.3, 46.4, 126.5, 126.8, 126.9, 127.2, 128.3, 128.4, 128.6, 129.0, 129.4, 129.5, 129.7, 130.2, 130.5, 130.7, 130.9, 132.5, 132.8, 136.1, 136.3, 138.3, 147.0, 147.5.

4.1.1.11. Synthesis of 3-azidomethyl-l-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalene (16). A mixture of compound **15** (0.293 g, 0.79 mmol) and sodium azide (0.154 g, 2.38 mmol) in DMF (5 mL) was stirred at 60 °C for 24 h. The reaction mixture was filtered and evaporated in vacuo. The residue was partitioned between water and EtOAc. The organic layer was separated, washed with water, dried over Na_2SO_4, and evaporated to give compound **16** as a pale-yellow oil (mixture of *cis* and *trans* diastereomers, ratio = 1:1.1, 0.18 g, 68%). The diastereomers were separated using a preparative chiral HPLC procedure (ChiralPak OD column; hexanes/MeOH = 98:2; μ = 8 mL/min; and λ = 225 nm) to give compounds **16a**–**d** (retention times: 9.8 min, 12.0 min, 14.5 min and 20.1 min, respectively).

*4.1.1.11.1. cis-Isomers 16a and **16b**.* ^{1}H NMR ($CDC1_3$) δ 1.92–2.09 (m, 3H), 2.61 (dd, J = 16.4 Hz, 9.8 Hz, 1H), 3.00 (dd, J = 16.8 Hz, 4.8 Hz, 1H), 3.29 (d, J = 6.0 Hz, 2H), 4.25 (t, J = 4.8 Hz, 1H), 6.81–6.92 (m, 2H), 7.08–7.15 (m, 2H), 7.18–7.21 (m, 2H), 7.32 (d, J = 6.0 Hz, 1H). ^{13}C NMR ($CDC1_3$) δ 30.2, 33.4, 35.5, 43.2, 56.8, 126.6, 127.2, 128.3, 128.9, 129.6, 130.4, 130.5, 130.8, 132.5, 136.2, 136.6, 147.5.

*4.1.1.11.2. trans-Isomers 16c and **16d**.* ^{1}H NMR ($CDCl_3$) δ 1.53 (dd, J = 24.8 Hz, 12.4 Hz, 1H), 2.13–2.25 (m, 2H), 2.67–2.74 (m, 1H), 2.94–3.00 (m, 1H), 3.32–3.41 (m, 2H), 4.08 (dd, J = 12 Hz, 5.2 Hz, 1H), 6.74 (d, J = 12.4 Hz, 1H), 7.00–7.08 (m, 2H), 7.14–7.18 (m, 2H), 7.27 (d, J = 7.8 Hz, 1H), 7.38 (d, J = 12.4 Hz, 1H). ^{13}C NMR ($CDC1_3$) δ 34.3, 35.4, 38.3, 46.2, 57.3, 126.5, 126.8, 128.4, 129.4, 129.6, 130.6, 130.7, 130.8, 132.7, 136.0, 138.4, 147.1.

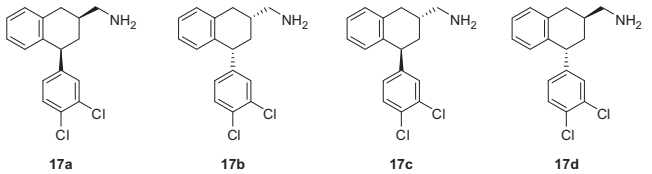
4.1.1.12. Synthesis of (4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalen-2-yl)methanamine (17a–d). To a solution of compound **16a** (36 mg, 0.108 mmol), in EtOH (5 mL) was added Pd/C (10%, 13 mg). A hydrogen balloon was attached and the reaction mixture was stirred at room temperature for 15 min. The mixture was filtered and concentrated in vacuo. The residue was purified by HPLC, AD column, hexanes/IPA/DEA = 90:10:0.05. Compound **17a** was obtained as a clear oil (23 mg, 70%).

Compound **17b** was prepared from compound **16b** (32 mg, 0.096 mmol) according to the procedure outlined above and was obtained as a clear oil (19 mg, 63%). LRMS m/z 306.2.

Compound **17c** was prepared from compound **16c** (33 mg, 0.099 mmol) following the procedure outlined above and was obtained as a clear oil (26 mg, 86%).

Compound **17d** was prepared from **16d** (32 mg, 0.096 mmol) following the procedure outlined above and was obtained as a clear oil (20 mg, 70%). LRMS *m*/*z* 306.2.

Absolute stereochemistries for compounds **14a–d** were determined using a combination of NMR techniques (determination of *cis*- and *trans*-configurations) and chiral HPLC analyses using authentic samples, which were prepared from commercial (*S*)-α-tetralone as described in the text. The resulting structures indicating absolute stereochemistries are shown below:



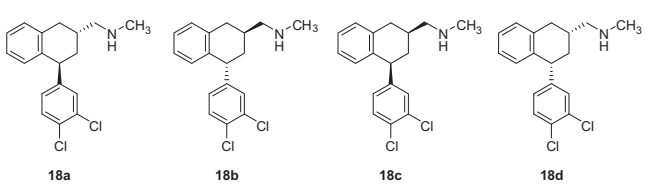
17a **17b** **17c** **17d**

*4.1.1.12.1. cis-Isomers **17a** and **17b**.* ^{1}H NMR ($CDC1_3$) δ 1.75–1.97 (m, 3H), 2.51 (dd, *J* = 16.8 Hz, 9.8 Hz, 1H), 2.61–2.70 (m, 2H), 3.02 (dd, *J* = 16.8 Hz, 5.2 Hz, 1H), 4.24 (t, *J* = 3.6 Hz, 1H), 6.81-6.91 (m, 2H), 7.07–7.12 (m, 2H), 7.16–7.20 (m, 2H), 7.29 (d, *J* = 6.0 Hz, 1H). ^{13}C NMR ($CDC1_3$) δ 32.5, 33.8, 35.8, 43.5, 47.8, 126.4, 127.0, 128.4, 129.6, 130.1, 13 0.2, 130 5, 130.9 132.4, 137.1, 148.1. LRMS *m*/*z* 306.2.

*4.1.1.12.2. trans-Isomers 17c and **17d**.* ^{1}H NMR ($CDC1_3$) δ 1.45 (dd, *J* = 24.9 Hz, 12.3 Hz, 1H), 1.92–2.00 (m, 1H), 2.18–2.24 (m, 1H), 2.58–2.62 (m, 1H), 2.69–2.80 (m, 2H), 2.94–3.01 (m, 1H), 4.06 (dd, *J* = 12 Hz, 5.2 Hz, 1H), 6.72 (d, *J* = 12.4 Hz, 1H), 6.99–7.05 (m, 2H), 7.13–7.18 (m, 2H), 7.27 (d, *J* = 7.8 Hz, 1H), 7.36 (d, *J* = 12.4 Hz.: 1H). ^{13}C NMR ($CDC1_3$) δ 34.7, 38.2, 38.7, 46.5, 48.2, 126.3, 126.6, 128.4, 129.4, 129.5, 130.4, 130.7, 130.8, 132.7, 136.9, 138.9, 147.6. LRMS *m*/*z* 306.2.

4.1.1.13. Synthesis of [4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydro-naphthalen-2-ylmethyl]-methylamine (18a–d). A mixture of compound **15** (0.342 g, 0.92 mmol) and methylamine (2.0 M in THF, 4.6 mL, 9.24 mmol) in a sealed tube was heated to 100 °C for 5 h. The reaction mixture was evaporated in vacuo. The residue was purified by chromatography, CombiFlash silica gel column, MeOH/CH_2C1_2, MeOH from 0% to 5%, to give Compound **18** as a clear oil (mixture of *cis* and *trans* diastereomers, ratio = 1:1.2, 0.201 g, 68%). Compounds **18a**, **18b**, **18c**, and **18d** were separated using a preparative chiral HPLC procedure (ChiralPak OD column; hexanes/IPA/DEA = 96:10:0.05; μ = 8 mL/min; and λ = 225 nm) to give **18a–d** (retention times: 11.2, 14.7, 16.3, and 21.2 min, respectively).

Absolute stereochemistries of compounds **18a–d** were determined using a combination of NMR techniques (determination of *cis*- and *trans*-configurations) and chiral HPLC analyses using authentic samples, which were prepared from commercial (*S*)-α-tetralone as described in the text. The resulting structures indicating absolute stereochemistries are shown below:



18a **18b** **18c** **18d**

*4.1.1.13.1. trans-Diastereomers 18a and **18b**.* HRMS $[M+H]^+$: calcd for (**18a**) $C_{18}H_{20}Cl_2N$ 320.0967, found 320.0975, calculated for (**18b**) $C_{18}H_{20}Cl_2N$ 320.0967, found 320.0995; LC–MS ^{1}H NMR ($CDC1_3$) δ 1.04 (br s, 1H), 1.88–1.97 (m, 3H), 2.39 (s, 3H), 2.47–2.56 (m, 3H), 3.21 (dd. *J* = 12.6 Hz, 2.7 Hz, 1H), 4.23 (t, *J* = 3.6 Hz, 1H), 6.81–6.91 (m, 2H), 7.07–7.12 (m, 2H), 7.16–7.20 (m, 2H), 7.29 (d, *J* = 6.0 Hz, 1H). ^{13}C NMR ($CDC1_3$) δ 29.6, 34.5, 36.4, 37.1, 43.5, 58.0, 126.4, 126.9, 127.3, 128.4, 129.6, 130.0, 130.2, 130.5, 132.4, 137.1, 137.2, 148.0. LRMS *m*/*z* 320.3.

*4.1.1.13.2. cis-Diastereomers 18c and **18d**.* HRMS $[M+H]^+$: calcd for (**18c**) $C_{18}H_{20}Cl_2N$ 320.0967, found 320.0994, calculated for (**18d**) $C_{18}H_{20}Cl_2N$ 320.0967, found 320.0976; ^{1}H NMR ($CDC1_3$) δ 1.35 (br s, 1H), 1.47 (dd, *J* = 24.9 Hz, 12.3 Hz, 1H), 2.03–2.13 (m, 1H), 2.17–2.24 (m, 1H), 2.48 (s, 3H), 2.58–2.67 (m, 2H), 2.94–3.11 (m, 1H), 4.06 (dd, *J* = 12 Hz, 5.2 Hz, 1H), 6.73 (d, *J* = 12.4 Hz, 1H), 6.97–7.05 (m, 2H), 7.09–7.18 (m, 2H), 7.26 (d, *J* = 7.8 Hz, 1H), 7.35 (d, *J* = 12.4 Hz, 1H). ^{13}C NMR ($CDC1_3$) δ 35.2, 35.3, 37.1, 39.3, 46.5, 58.5, 126.3, 126.6, 128.5, 129.5, 129.6, 130.3, 130.7, 130.9, 132.6, 137.2, 139.0, 147.6. LRMS *m*/*z* 320.3.

4.1.1.14. Synthesis of [4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydro-naphthalen-2-ylmethyl]-dimethylamine (19a, 19b). A mixture of compound **15** (0.40 g, 1.08 mmol) and dimethylamine (2.0 M in THF, 5.4 mL, 10.8 mmol) in a scaled tube was heated to 100 °C for 5 h. The reaction mixture was evaporated in vacuo. The residue was purified by chromatography, CombiFlash silica gel column, MeOH/CH_2C1_2, MeOH from 0% to 5%, to give **16** as a clear oil (mixture of *cis* and *trans* diastereomers, ratio = 1:1.2, 0.253 g, 70%). *cis*- and *trans*-Diastereomers were separated using a preparative HPLC procedure (ChiralPak OD column; hexanes/EtOH/MeOH/DEA = 96:2:2:0.05; μ = 8 mL/min; and λ = 225 nm) to give a mixture of *cis*-enantiomers (**19a**) and a mixture of *trans*-enantiomers (**19b**).

*4.1.1.14.1. cis-Diastereomers **19a**.* ^{1}H NMR ($CDC1_3$) δ 1.42 (dd, *J* = 24.9 Hz, 12.3 Hz, 1H), 2.03–2.13 (m, 1H), 2.15–2.22 (m, 3H), 2.23 (s, 6H), 2.51–2.61 (m, 1H), 2.94–3.10 (m, 1H), 4.06 (dd, *J* = 12 Hz, 5.2 Hz, 1H), 6.73 (d, *J* = 12.4 Hz, 1H), 6.97–7.05 (m, 2H), 7.09–7.18 (m, 2H), 7.26 (d, *J* = 7.8 Hz, 1H), 7.35 (d, *J* = 12.4 Hz, 1H). ^{13}C NMR ($CDC1_3$) δ 33.3, 35.6, 39.7, 46.3, 46.6, 66.6, 126.3, 126.6, 128.5, 129.5, 129.6, 130.3, 130.7, 130.9, 132.6, 137.2, 139.0, 147.7. LRMS *m*/*z* 334.3.

4.1.1.14.2. trans-Diastereomers 19b. ^{1}H NMR ($CDC1_3$) δ 1.82–2.05 (m, 3H), 2.13 (s, 6H), 2.20–2.25 (m, 2H), 2.50 (dd, *J* = 12 Hz, 5.2 Hz, 1H), 2.95–3.04 (m, 2H), 4.22 (t, *J* = 3.6 Hz, 1H), 6.8–1- 6.91 (m, 2H), 7.07–7.12 (m, 2H), 7.16–7.20 (m, 1H), 7.3 (d, *J* = 6.0 Hz, 1H). ^{13}C NMR ($CDC1_3$) δ 27.3, 34.6, 36.2, 43.3, 46.0, 65.4, 126.4, 126.9, 127.3, 128.4, 129.6, 130.0, 130.2, 130.5, 132.4, 137.1, 137.2, 148.1. LRMS *m*/*z* 334.3.

4.1.1.15. Synthesis of 2-(-1-(3,4-dichlorophenyl)-1,2-dihydronaphthalen-3-yl)acetonitrile (20). To a stirred solution of diethyl cyanomethyl phosphonate EtO_2POCH_2CN (0.324 mL, 2 equiv) in THF (2 mL) was added sodium hydride (60 mg, 60% in oil) in portions. After 30 min, 4-(3,4-dichlorophenyl)-3,4-dihydronaphthalen-2(1*H*)-one (β-tetralone) **8** (291 mg, 1 mmol) was added as a solution in THF (3 mL). After the mixture was stirred for 2 h at 0 °C, the reaction was quenched with ammonium chloride solution, extracted with MTBE, dried over sodium sulfate and evaporated. The residue was separated on silica to give the unsaturated nitrile (0.24 g, 77%) as a pale-green oil. GC–MS R_t = 14.59 min, *m*/*z* = 313 (M+). ^{1}H NMR ($CDCl_3$) δ 7.37 (d, *J* = 8.3 Hz, 1H), 7.3 (m, 2H), 7.2 (m, 2H), 7.00 (dd, *J* = 2.1, 8.3 Hz, 1H), 6.84 (d, *J* = 7.4 Hz, 1H), 6.64 (bs, 1H), 4.17 (t, *J* = 8.1 Hz, 1H), 3.21 (br s, 2H), 2.69 (dd, *J* = 6.9, 17.3 Hz, 1H), 2.52 (dd, *J* = 8.4, 17.2 Hz, 1H). ^{13}C NMR ($CDCl_3$) δ 143.9, 135.2, 132.9, 132.5, 130.7, 130.5, 130.0, 128.2, 127.7, 127.5, 126.8, 126.4, 116.5, 43.1, 35.1, 25.1.

4.1.1.16. Synthesis of 2-(4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalen-2-yl)acetonitrile. To a solution of the unsaturated nitrile **20** (210 mg, 0.6683 mmol) in 1% wet methanol (28 mL) was added 5% Pd/C (21 mg). The atmosphere was

evacuated under vacuum and refilled with hydrogen from a balloon. The reaction was monitored by HPLC and was stopped after 220 min. The catalyst was removed by filtration (Celite) and the solvent removed in vacuo. The residue was diluted with DCM and filtered through an aminopropyl cartridge. The solvent was stripped to give the intermediate (201 mg, 95%) as a pale-yellow oil. ^{1}H NMR ($CDCl_3$) δ 7.40 (d, J = 8.3 Hz, 1H), 7.3 (m, 1H), 7.2 (m, 2H), 7.1 (m, 1H), 7.02 (dd, J = 2.1, 8.2 Hz, 1H), 6.76 (d, J = 7.7 Hz, 1H), 4.12 (dd, J = 5.4, 12.1 Hz, 1H), 3.06 (ddd, J = 2.4, 4.3, 16.2 Hz, 1H), 2.80 (dd, J = 12.4, 15.6 Hz, 1H), 2.48 (dd, J = 2.6, 6.5 Hz, 2H), 2.3 (m, 2H), 1.66 (q, J = 12.6 Hz, 1H). ^{13}C NMR ($CDCl_3$) δ 146.3, 137.6, 135.1, 132.6, 130.6, 130.6, 129.3, 129.0, 128.1, 126.7, 126.6, 118.1, 45.9, 39.6, 35.8, 31.9, 24.2.

4.1.1.17. Synthesis of 2-(4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalen-2-yl)ethanamine (21). To a stirring solution of the nitrile (200 mg, 0.6324 mmol) and THF (8 mL) at ambient temperature was added borane-THF (4 mL, 6 equiv) dropwise. After heating in the microwave (maximum temperature 130 °C) for 5 min, the reaction was cooled, quenched with 6 N HCl, and washed with MTBE. The aqueous layer was chilled, basicified with KOH, and extracted with MTBE. The organic layer was evaporated, diluted with DCM, dried over sodium sulfate, filtered through an aminopropyl cartridge and evaporated to give amine **21** (101 mg, 50%) as a pale-yellow oil. LC–MS R_t = 9.41 min, m/z = 320 (M+1). ^{1}H NMR ($CDCl_3$) δ 7.36 (d, J = 8.2 Hz, 1H), 7.26 (s, 1H), 7.1 (m, 2H), 7.0 (m, 2H), 6.72 (d, J = 7.7 Hz, 1H), 4.05 (dd, J = 5.3, 12.0 Hz, 1H), 2.92 (dd, J = 2.4, 16.4 Hz, 1H), 2.83 (t, J = 7.3 Hz, 2H), 2.60 (m, 1H), 2.1 (m, 1H), 2.0 (M, 1H), 1.6–1.4 (m, 3H). ^{13}C NMR ($CDCl_3$) δ 147.4, 138.5, 137.0, 132.3, 130.6, 130.4, 130.1, 129.3, 129.0, 128.1, 126.2, 125.9, 46.5, 41.0, 40.8, 39.6, 36.9, 32.3.

4.1.1.18. Enantiomeric separation of 21; synthesis of *tert*-butyl 2-(4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalen-2-yl)ethylcarbamate. To a solution of the primary amine (100 mg, 0.3122 mmol) in ether (3 mL) was added 10% KOH (1 mL) and BOC anhydride (136 mg, 2 equiv). After 2 h at ambient temperature, the solution was extracted with MTBE. The organic phase was separated and the volatiles removed in vacuo to give the crude carbamate (208 mg) as a 1:1 mixture with excess BOC anhydride. Most of the anhydride was removed by washing an MBTE solution of the crude product with 1 M HCl. This material was separated on a Chiracel OD semiprep column (90:10:0.1 Hex/IPA/DEA) to give the fast moving enantiomer (56.2 mg, 50%) and the slow-moving enantiomer (55.7 mg, 50%). NMR analysis indicated that the formed enantiomers had *cis*-configuration. ^{1}H NMR ($CDCl_3$) δ 7.36 (d, J = 8.2 Hz, 1H), 7.3 (m, 1H), 7.1 (m, 2H), 7.0 (m, 2H), 6.72 (d, J = 7.7 Hz, 1H), 4.56 (br s, 1H), 4.04 (dd, J = 5.4, 12.0 Hz, 1H), 3.2 (m, 2H), 2.93 (dd, J = 2.6, 16.3 Hz, 1H), 2.60 (dd, J = 12.0, 16.1 Hz, 1H), 2.2 (m, 1H), 1.9 (m, 1H), 1.57 (q, J = 7.1 Hz, 2H), 1.44 (s, 9H). ^{13}C NMR ($CDCl_3$) δ 155.9, 147.3, 138.4, 136.7, 132.4, 130.6, 130.4, 130.1, 129.3, 129.0, 128.1, 126.3, 126.0, 46.4, 40.8, 38.1, 37.0, 36.7, 32.3, 28.4.

4.1.1.19. Synthesis of *cis* and *trans*-2-(1-(3,4-dichlorophenyl)-1,2,3,4-tetrahydronaphthalen-3-yl)acetonitrile (21a and 21b). To a solution of carbamate (20 mg, 0.05585 mmol) in $CDCl_3$ was added HCl (1 mL, 4 M in dioxane). After 1 h, the mixture was chilled, quenched with KOH (1 mL, 5 M in H_2O), extracted with MTBE and evaporated. The crude oil was diluted in DCM, filtered through an aminopropylcartridge and evaporated to give the pure primary amine **21a** (11.5 mg, 64%) as a clear oil.

The second enantiomer was prepared from the enantiomeric carbamate using the procedure described above to give the enantiomeric amine **21b** (11.1 mg, 62%) as a clear oil.

4.1.1.20. Synthesis of 1-(3,4-dichlorophenyl)-3,4-dihydronaphthalen-2(1*H*)-one (23). To a stirring solution of β-tetralone (**22**) (1.00 g, 6.84 mmol) and $Pd(dba)_2$ (39 mg, 1 mol %) in toluene was added t-Bu_3P (228uL, 10 wt % in hexanes, 1.1%). The solution was chilled (dry-ice bath) before adding LiHMDS (7.5 mL, 1 M in hexanes, 1.1 equiv) followed by 1-bromo-3,4-dichlorobenzene (1 mL, 1.1 equiv). The solution was then allowed to warm to ambient temperature and heated under microwave radiation for 5 min (maximum temperature 140 °C). After cooling, the reaction was quenched with aqueous ammonium chloride and extracted with MTBE. The organic layer was dried with sodium sulfate, filtered through Celite, and evaporated. The crude oil was separated on silic gel to give the title compound (1.45 g, 73%) as a slight brown oil. This material was assayed as 90% pure. GC–MS R_t = 13.21 min m/z = 290 (M+). ^{1}H NMR ($CDCl_3$) δ 7.37 (d, J = 8.3 Hz, 1H), 7.3–7.2 (m, 3H), 7.17 (d, J = 2.1 Hz, 1H), 6.9 (m, 2H), 4.68 (s, 1H), 3.1 (m, 2H), 2.7 (m, 2H). ^{13}C NMR ($CDCl_3$) δ 208.4, 137.7, 136.7, 135.3, 132.7, 131.5, 130.7, 130.5, 129.2, 128.2, 128.1, 127.7, 127.3, 58.6, 37.1, 28.0.

4.1.1.21. Synthesis of 1-(3,4-dichlorophenyl)-*N*-methyl-1,2,3,4-tetrahydronaphthalen-2-amine (24a–d). To a solution of tetralone **23** (400 mg, 1.374 mmol) in THF (10 mL) and methanol (15 mL) was added methylamine hydrochloride (1.12 g, 10 equiv). The resultant mixture was stirred at 50 °C. After dissolution (10 min), sodium cyanoborohydride (6.9 mL, 1 M in THF, 5 equiv) was added in a single portion. After 20 h, the organic layer was evaporated, filtered through silica and an aminopropyl cartridge. The crude oil was then diluted with sodium bicarbonate solution and extracted with MTBE to give the amine (280 mg, 66%) as a mixture of four stereoisomers (1:1:1:1).

These amines were separated using a Chiracel OD (98:2:0.1 Hex/IPA/DEA) column to give three fractions. The first was pure E1 (**24a**); the second was a mixture of E2 and E3; and the third was pure E4 (**24d**). The mixture was further separated using a Chiracel OD (2:3:95:0.1 MeOH/EtOHHex/ DEA) column. The order of elution of the middle fractions changes between these columns and was defined based on the OD 98:2:0.1 conditions. Retention times are summarized in Table 6, below. The relative (*cis*/*trans*) configuration of the compounds was assigned based on ^{1}H NMR coupling constants; the absolute stereochemistry was assigned arbitrarily.

*4.1.1.21.1. cis-Enantiomers 24b (E2) and **24d** (E4).* LC–MS R_t = 8.83 min m/z = 306 (M+1). ^{1}H NMR ($CDCl_3$) δ 7.31 (d, J = 8.3 Hz, 1H), 7.2–7.1 (m, 3H), 7.1–7.0 (m, 1H), 6.9 (m, 2H), 4.32 (d, J = 5.1 Hz, 1H), 3.1–2.8 (m, 3H), 2.50 (s, 3H), 1.9 (m, 1H), 1.6 (m, 1H). ^{13}C NMR ($CDCl_3$) δ 142.5, 137.4, 136.4, 132.0, 131.9, 130.5, 129.7, 129.6, 128.8, 126.7, 126.0, 58.5, 48.3, 33.9, 28.1, 23.7.

*4.1.1.21.2. trans-Enantiomers 24a (E1) and **24c** (E3).* LC–MS R_t = 9.12 min m/z = 306 (M+1). ^{1}H NMR ($CDCl_3$) δ 7.37 (d, J = 8.2 Hz, 1H), 7.2 (m, 1H), 7.1 (m, 2H), 7.0 (m,1H), 6.97 (dd, J = 2.0, 8.2 Hz, 1H), 6.69 (d, J = 7.8 Hz, 1H), 3.91 (d, J = 7.7 Hz, 1H), 2.94 (t, J = 6.5 Hz, 2H), 7.51 (td, J = 1.4, 7.5 Hz, 1H), 2.42 (s, 3H), 2.2 (m, 1H), 1.7 (m, 1H). ^{13}C NMR ($CDCl_3$) δ 145.1, 137.0, 136.4, 132.5, 131.4, 131.4, 130.8, 130.0, 129.9, 128.4, 126.7, 126.0, 62.3, 51.4, 33.7, 27.1, 25.5.

Table 6
Retention times for each isomer [min]

	24a E1 *trans*	**24b** E2 *cis*	**24c** E3 *trans*	**24d** E4 *cis*
HPLC R_t (Chiracel OD, 98:2:0.1 Hex/IPA/DEA)	6.0	6.7	7.9	13.7
HPLC R_t (Chiracel OD, 2:3:95:0.1 MeOH/EtOH/ Hex/DEA)	5.5	6.2	7.0	10.5

4.2. In vitro pharmacology

The novel compounds described above were tested for their inhibition of functional uptake of 5-HT,[14] NE,[15] or DA,[16] using recombinant transporters expressed in HEK-293, MDCK or CHO-K1 cells as described in the literature. Compounds were tested initially at 10 μM in duplicate, and if ⩾50% inhibition of uptake was observed, they were tested further at 10 different concentrations in duplicate in order to obtain full inhibition curves. IC_{50} values (concentration inhibiting control activity by 50%) were then determined by nonlinear regression analysis of the inhibition curves. Assay performance was monitored by the use of SERT reference compound fluoxetine (IC_{50} = 5.2 ± 0.6 nM), NET reference compound nisoxetine (IC_{50} = 8.3 ± 1.0 nM) and DAT reference compound nomifensine (IC_{50} = 30.2 ± 2.2 nM). Replicate studies done independently on a set of compounds allowed the determination of values for minimum significant ratios (MSR). MSR is a statistical measure for how many fold different IC_{50} values for two compounds (determined only once) must be for those two IC_{50} values to be considered significantly different with 95% confidence limits.[20] From replicate studies of the effects of 13 reuptake inhibitors assayed with the human transporters at MDS Pharma on two separate occasions, MSR values were 2, 2, and 3, respectively, for SERT, NET and DAT. Similar analysis of the assays performed internally at Sepracor provided MSR values of 4, 3, and 2, respectively, for SERT, NET, and DAT. These corresponded to average SD values (on pIC_{50}s) of 0.2–0.3.

In vitro microsomal stability, hERG inhibition and CYP inhibition assays were performed at Cyprotex (www.cyprotex.com), Macclesfield UK, using their standard assay protocols.

4.3. In vivo pharmacology

4.3.1. Mouse tail suspension test

The method, which detects antidepressant activity, follows that described by Stéru et al. (*Psychopharmacology*, **1985**, *85*, 367–370). Rodents, suspended by the tail, rapidly become immobile. Antidepressants decrease the duration of immobility. The behavior of the animal was recorded automatically for 5 min using a computerized device (Med-Associates Inc.) similar to that developed by Stéru et al. (*Prog.* Neuropsychopharmacol. Exp. Psychiatry, **1987**, *11*, 659–671). Mice (10–12) were tested in each group. The test was performed blind. Compounds were typically evaluated at 3 doses (1–30 mg/kg), administered orally one time: 30–120 min before the test, and compared with a vehicle control group. Desipramine (100 mg/kg), administered under the same experimental conditions, was used as the positive reference substance.

Data were analyzed by one way analysis of variance (ANOVA) followed by post-hoc comparisons where appropriate. An effect was considered significant if $p < 0.05$.

4.3.2. Locomotor activity

In order to ensure effects of the compounds on immobility time were not related to a general stimulant effect on baseline motor activity, locomotor activity was assessed using photocell monitored cages (Med-Associates Inc.). Each test chamber was equipped with infrared photocell beams to measure movement of the animals. Horizontal and vertical activities were measured.

Rats or mice were pretreated with vehicle or test compounds and placed back in home cage, following which they were individually placed in locomotor cages and activity was monitored in 5 min intervals for up to 60 min.

Data were analyzed by one way analysis of variance (ANOVA) followed by post-hoc comparisons where appropriate. An effect was considered significant if $p < 0.05$.

Acknowledgements

The authors thank Zhi-Dong Jiang and Margaret E. Kearney for their technical support.

References and notes

1. British Association for Psychopharmacology. Evidence Based Guidelines for Treating Depressive Disorders with Antidepressants: A Revision of the 2000 British Association for Psychopharmacology Guidelines [Online]. Available from URL: http://www.bap.org.uk.
2. Paul, I. A. *Pharm. News* **2001**, *8*, 33.
3. Corrigan, M. H.; Denahan, A. Q.; Wright, C. E.; Ragual, R. J.; Evans, D. L. *Depress. Anxiety* **2000**, *11*, 58.
4. Sporn, J.; Ghaemi, S. N.; Sambur, M. R.; Rankin, M. A.; Recht, J.; Sachs, G. S.; Rosenbaum, J. F.; Fava, M. *Ann. Clin. Psych.* **2000**, *12*, 137.
5. D'Aquila, P. S.; Collu, M.; Gessa, G. L.; Serra, G. *Eur. J. Pharmacol.* **2000**, *405*, 365.
6. (a) Welch, W. M.; Kraska, A. R.; Sarges, R.; Koe, K. *J. Med. Chem.* **1984**, *27*, 1508; (b) Welch, W. M.; Harbert, C. A.; Koe, K, B.; Kraska, A. R. U.S. Patent 4556,676, 1980.; (c) MacQueen, G.; Born, L.; Steiner, M. C. N. S. *Drug Rev.* **2001**, *7*, 1; (d) McRae, A. L.; Brady, K. T. *Exp. Opin. Pharmacother.* **2001**, *2*, 883; (e) Taber, G. P.; Pfisterer, D. M.; Colberg, J. C. *Org. Process Res. Dev.* **2004**, *8*, 385.
7. Skolnick, P.; Popik, P.; Janowsky, A.; Beer, B.; Lippa, A. S. *Eur. J. Pharm.* **2003**, *461*, 99.
8. (a) Beer, B.; Stark, J.; Krieter, P.; Czobor, P.; Beer, G.; Lippa, A.; Skolnick, P. *J. Clin. Pharmacol.* **2004**, *44*, 1360; (b) Skolnick, P.; Popik, P.; Janowsky, A.; Beer, B.; Lippa, A. S. *Life Sci.* **2003**, *73*, 3175.
9. DOV Pharmaceutical Press Release September 27, 2005.
10. Graff, O.; Learned, S.; Roychowdhury, S. M.; Moate, R.; Morrison, D.; Lavergne, A; Krishnan, K. R.; Archer, G.; Modell, J.; Alexander, R.; Ratti, E. ACNP Poster, 2009.
11. Sepracor Inc. Press Release, July 1st, 2009.
12. *Eschweiler–Clarke Reductive Alkylation of Amines. Name Reactions for Functional Group Transformations*; John Wiley & Sons: Hoboken, N. J, 2007; pp 86–111.
13. (a) Wyrick, S. D.; Booth, R. G.; Myers, A. M.; Owens, C. E.; Kula, N. S.; Baldessarini, R. J.; McPhail, A. T.; Mailman, R. B. *J. Med. Chem.* **1993**, *36*, 2542; (b) Wyrick, S. D.; Booth, R. G.; Myers, A. M.; Owens, C. E.; Bucholtz, E. C.; Hooper, P. C.; Kula, N. S.; Baldessarini, R. J.; Mailman, R. B. *J. Med. Chem.* **1995**, *38*, 3857.
14. Gu, H.; Wall, S.; Rudnick, G. *J. Biol. Chem.* **1994**, *269*, 7124.
15. Galli, A.; DeFelice, L. J.; Duke, B. J.; Moore, K. R.; Blakely, R. D. *J. Exp. Biol.* **1995**, *198*, 2197.
16. Pristupa, Z. B.; Wilson, J. M.; Hoffman, B. J.; Kish, S. J.; Niznik, H. B. *Mol. Pharmacol.* **1994**, *45*, 125.
17. (a) Ozdemir, V.; Naranjo, C. A.; Herrman, N.; Shulman, R. W.; Sellers, E. M.; Reed, K.; Kalow, W. *J. Clin. Psychopharmacol.* **1998**, *18*, 55; (b) Sproule, B. A.; Otton, S. V.; Cheung, S. W.; Zhong, X. H.; Romach, M. K.; Sellers, E. M. *J. Clin. Psychopharmacol.* **1997**, *17*, 102.
18. Steru, L.; Chermat, R.; Thierry, B.; Simon, P. *Psychopharmacology* **1985**, *85*, 367.
19. Liu, X.; Smith, B. J.; Chen, C.; Callegari, E.; Becker, S. L.; Chen, X.; Cianfrogna, J.; Doran, A. C.; Doran, S. D.; Gibbs, J. P.; Hosea, N.; Liu, J.; Nelson, F. R.; Szewc, M. A.; Deusen, J. V. *Drug Metab. Dispos.* **2006**, *34*, 1443.
20. Eastwood, B. J.; Farmen, M. W.; Iversen, P. W.; Craft, T. J.; Smallwood, J. K.; Garbison, K. E.; Delapp, N. W.; Smith, G. F. *J. Biomol. Screening* **2006**, *11*, 253.
21. Middleton, D. S.; Andrews, M.; Glossop, P.; Gymer, G.; Jessiman, A.; Johnson, P. S.; MacKenney, M.; Pitcher, M. J.; Rooker, T.; Stobie, A.; Tang, K.; Morgan, P. *Bioorg. Med. Chem. Lett.* **2006**, *16*, 1434.