Available online at www.sciencedirect.com
SCIENCE DIRECT
Bioorganic & Medicinal Chemistry Letters 16 (2006) 691–694
Bioorganic & Medicinal Chemistry Letters

Derivatives of tramadol for increased duration of effect

Liming Shao,* Craig Abolin, Michael C. Hewitt, Patrick Koch and Mark Varney

Sepracor Inc., 84 Waterford Drive, Marlborough, MA 01752, USA

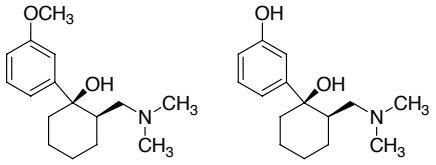
Received 23 August 2005; revised 6 October 2005; accepted 7 October 2005
Available online 27 October 2005

Abstract—Tramadol is a centrally acting opioid analgesic structurally related to codeine and morphine. Analogs of tramadol with deuterium-for-hydrogen replacement at metabolically active sites were prepared and evaluated in vitro and in vivo.
© 2005 Elsevier Ltd. All rights reserved.

Tramadol (±)-*cis* is used for the treatment of moderate to moderately severe pain.[1] Tramadol has a relatively short duration of analgesic effect due to extensive first pass metabolism and as such is dosed as frequently as 100 mg every 4–6 h.[2] The analgesic activity of tramadol is thought to be the result of a dual mechanism—the parent (**1**) acts as an inhibitor of norepinephrine and serotonin reuptake, and the major *O*-desmethyl metabolite (**M1**) is a potent agonist at μ-opioid receptors (Fig. 1).[3]

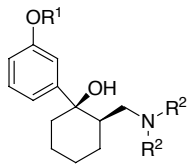
The opioid antagonist naloxone only partially inhibits the analgesic activity of tramadol in animal tests. Not unlike other opioids, tramadol causes a number of side effects including constipation, nausea, dizziness, and somnolence. A tramadol analog that had a longer half-life (and therefore required less-frequent dosing) and was devoid of opioid side effects would have therapeutic benefit. Herein, we wish to disclose our efforts to develop an improved tramadol analog with a longer half-life.

Our approach to tramadol analogs took advantage of the primary kinetic isotope effect[4] to slow CYP450-mediated metabolism (Fig. 2). Replacing hydrogen with deuterium at metabolically active sites can result in a slower metabolism due to the reduced rate of cleavage of a C–D bond relative to a C–H bond. This approach has been shown to be effective for a number of pharmacological agents including amphetamine,[5] butethal,[6] and morphine.[7] For tramadol, formation of metabolite **M1** is both the primary route of metabolism and is responsible for its opioid-like effects. By slowing down metabolism of tramadol, it was anticipated that the formation of **M1** would be reduced, providing a longer-acting drug with potentially fewer opioid-related side effects. Toward that end, the hydrogen atoms on the *O*-methyl and *N*-methyl groups were replaced by deuterium as shown in structures **D3**, **D6**, **D9**, and **D6 M1**.



1 **M1**

Figure 1. (±)-Tramadol **1** and metabolite (±)-**M1**.



D_3 $R^1 = CD_3$, $R^2 = CH_3$
D_6 $R^1 = CH_3$, $R^2 = CD_3$
D_6 M_1 $R^1 = H$, $R^2 = CD_3$
D_9 $R^1 = CD_3$, $R^2 = CD_3$

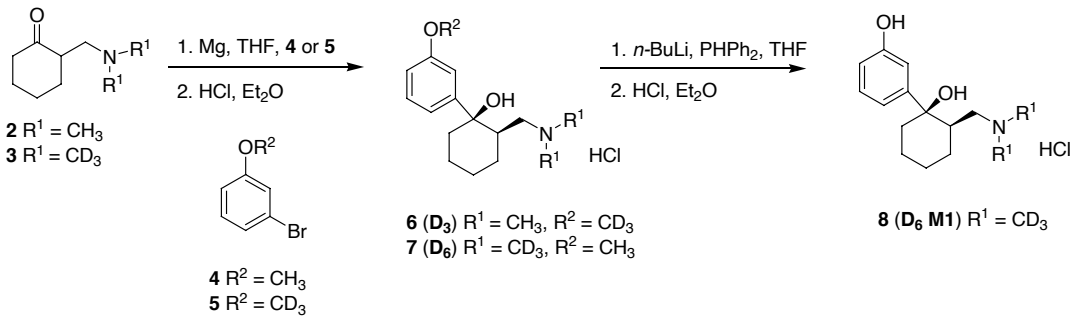
Dramadol analogs

Figure 2. D-tramadol (dramadol) analogs.

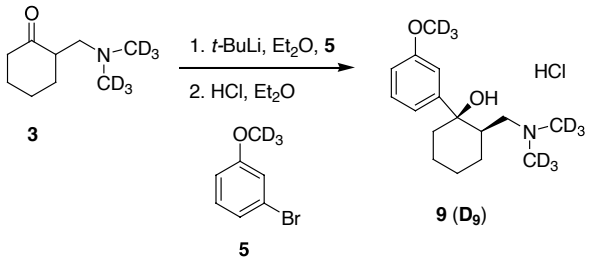
Keywords: Tramadol; Metabolic stability; Deuterium isotope effect; Opioids.
* Corresponding author. Tel.: +1 5083577467; fax: +1 5083577467; e-mail: liming.shao@sepracor.com

0960-894X/$ - see front matter © 2005 Elsevier Ltd. All rights reserved.

DOCKET ALARM
Find authenticated court documents without watermarks at docketalarm.com.



Scheme 1. Synthesis of **D3**, **D6**, and **D6 M1** metabolites.



Scheme 2. Synthesis of **9** (**D9**).

The syntheses of the deuterated tramadol analogs were relatively straightforward and began with Mannich base **2** or **3** (Scheme 1). Specifically, addition of the Grignard reagent **4** or **5** to ketones **2** or **3** provided crude **6** (**D3**) and **7** (**D6**) as mixtures of diastereomers (76% *cis* for **D6**). The abundance of the *cis* isomer could be improved to 93% after formation of the HCl salt and multiple recrystallizations from isopropanol.[8] Demethylation of **7** (**D6**) occurred without incident to provide phenol **8** (**D6 M1**).[9] Purification was effected via formation of the HCl salt and recrystallization from isopropanol/ethyl acetate to give the phenol salt as an analytically pure white powder. The reaction to form **D9** was attempted several times with starting materials **3** and **5**, with little product formation seen by LCMS and HPLC analysis. This led to an alternate strategy to synthesize **D9** that relied on a lithium–halogen exchange reaction that had been outlined previously in the literature,[10] and was successfully employed using Mannich base **3** and aromatic bromide **5** (Scheme 2).

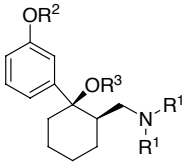
Crude **9** (**D9**) was also converted to its HCl salt and recrystallized several times in isopropanol to provide a 93% pure *cis* isomer.[11]

The receptor binding affinity and functional monoamine uptake inhibition by tramadol and the deuterated tramadol analogs are shown in Table 1.

As expected, the deuterated analog of M1 retained the μ-opioid receptor binding affinity present in metabolite M1. In general, the deuterated analogs retained in vitro activity comparable to their non-deuterated parent molecules, with a slightly diminished reuptake inhibition of norepinephrine compared to tramadol.

The in vitro metabolic stability results for the deuterated tramadol compounds are detailed in Table 2.[12,13] Based on disappearance of parent compound in incubation media, the most stable compound of the six tested in human liver microsomes and hepatocytes was clearly the **D9**-tramadol derivative **9**. Tramadol and **D6**-tramadol

Table 1. μ-Opioid binding, and 5-HT and NE reuptake inhibition of (±)-cis tramadol and deuterated derivatives



Compound	R^1	R^2	R^3	IC_{50} (nM)		
				μ	**5-HT**	NE
1 Tramadol	CH_3	CH_3	H	7600	4300	790
Metabolite M1	CH_3	H	H	47	4600	>10,000
6 (D3)	CH_3	CD_3	H	>10,000	1900	3600
7 (D6)	**CD_3**	CH_3	H	>10,000	3100	3200
8 (D6 M1)	CD_3	H	H	43	9900	6700
9 (D9)	CD_3	CD_3	H	5300	1100	2000

Note: IC_{50} is the concentration required to inhibit 50% binding to μ-opioid receptors or reuptake of 5-hydroxytryptamine (5-HT) and norepinephrine (NE).

DOCKET ALARM
Find authenticated court documents without watermarks at docketalarm.com.

Table 2. Summary of in vitro metabolic stability of tramadol and deuterated derivatives

Compound	Half-life[a] (min)/rank[b]	
	HLM[c]	Hepatocytes
9 (**D9**), (+/−)-*cis*	7210/1	3610/1
8 (**M1 D6**), (+/−)-*cis*	747/5	1650/2
Metabolite M1, (+/−)-*cis*	2820/2	1560/3
6 (**D3**), (+/−)-*cis*	744/4	1330/4
1 Tramadol, (+/−)-*cis*	741/3	968/5
7 (**D6**), (+/−)-*cis*	547/6	852/6

[a] Calculated from samples taken at 0, 180, and 360 min for hepatocytes and 0, 90, and 180 min for microsomes.
[b] Ranked from 1, the most stable to 6, the least stable.
[c] HLM = human liver microsomes.

7 were the least stable compounds in human liver microsomes and hepatocytes.

Metabolic stability was also evaluated based on the formation rate of metabolite **M1** from tramadol (**1**) and three tramadol deuterated derivatives capable of producing metabolite M1. The results are summarized for hepatocytes and microsomes in Figures 3 and 4, respectively. Consistent with a deuterium isotope effect on the metabolically labile *O*-methyl group, **6** (**D3**) and **9** (**D9**) slowed the formation of the primary metabolite **M1** by approximately 5-fold. The formation rate of metabolite **M1** from **7** (**D6**) was nearly indistinguishable from that of tramadol in human hepatocytes.

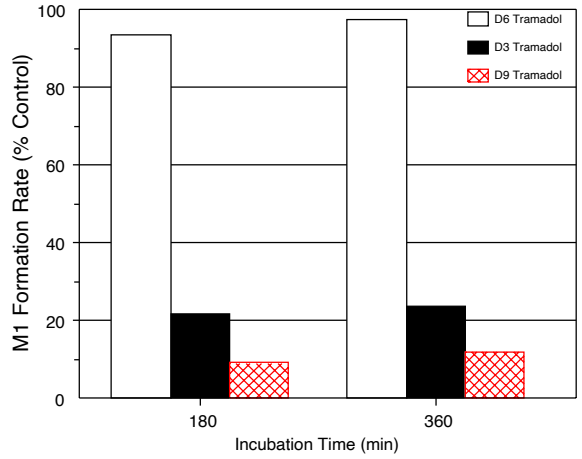
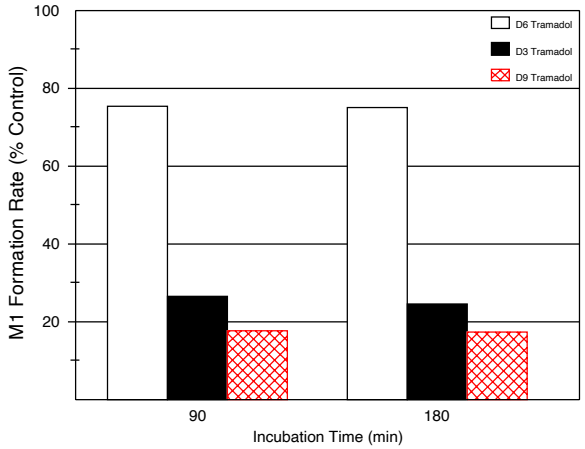


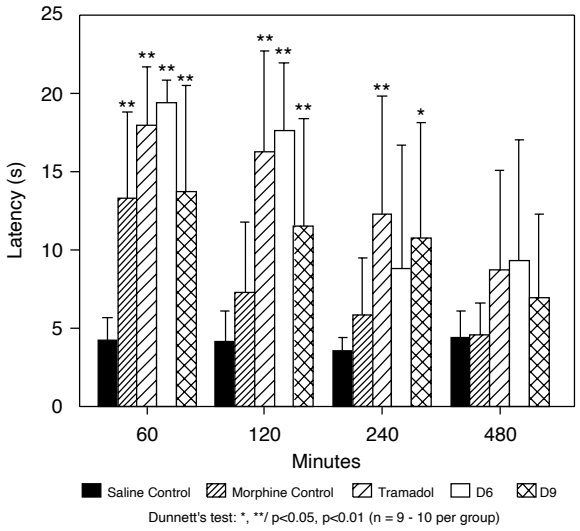
Figure 3. Stability of tramadol analogs in human hepatocytes.



The in vivo activities of tramadol (**1**), **7** (**D6**), and **9** (**D9**) were tested in the rat tail-flick model. Tramadol (**1**), **7** (**D6**), and **9** (**D9**) were administered at a dose of 50 mg/kg intraperitoneally. Intraperitoneal morphine (5 mg/kg) and saline were used as controls. Analgesic activity (tail-flick latency) was evaluated at 1, 2, 4, and 8 h after the dose. The results of efficacy (tail-flick latency) are shown in Figure 5.

Tramadol and two of its deuterated derivatives (**7** (**D6**) and **9** (**D9**)) significantly lengthened tail-flick latency when tested at 1 and 2 h after intraperitoneal administration of 50 mg/kg. Tramadol and **9** (**D9**) were also effective when tested 4 h after dosing. By 8 h the effects of the test compounds were no longer statistically significant. As expected, morphine increased tail-flick latency at the 1 h time point, but at later times the effect was not significantly greater than that of vehicle. The results indicate that the test compounds effectively reduced acute nociception for 2–4 h following a single acute intraperitoneal dose.

In conclusion, the substitution of deuterium for hydrogen in the methyl groups of tramadol did not adversely affect the in vitro binding affinity. In vitro testing in human microsomes confirmed that replacing hydrogen with deuterium in the metabolically labile *O*-methyl position of **1** slowed the formation of the primary metabolite **M1** by approximately 5-fold. The deuterated derivatives **7** (**D6**) and **9** (**D9**) were active analgesics in the rat tail-flick model, but were not superior to tramadol in terms of potency or duration of effect. Deuterium



DOCKET ALARM
Find authenticated court documents without watermarks at docketalarm.com.

for hydrogen replacement at metabolically active sites had no obvious deleterious effects in vivo but did not result in a longer duration of effect. In this case, deuteration at metabolically active sites produced a pharmacological agent equipotent in vivo with tramadol.

References and notes

1. (a) Raffa, R. B.; Friderichs, E.; Reimann, W.; Shank, R. P.; Good, E. E.; Vaught, J. L. *J. Pharmacol. Exp. Ther.* **1992**, *260*, 275; (b) Gibson, T. P. *Am. J. Med.* **1996**, *101*(Suppl 1A), 47S; (c) Radbruch, L.; Grond, S.; Lehmann, K. *Drug Safety* **1996**, *15*, 8; (d) Raffa, R. B. *Am. J. Med.* **1996**, *101*(Suppl 1A), 41S; (e) Garrido, M.; Valle, M.; Campanero, M. A.; Calvo, R.; Troconiz, I. F. *J. Pharmacol. Exp. Ther.* **2000**, *295*, 352; (f) Subrahmanyam, V.; Renwick, A. B.; Walters, D. G.; Young, P. J.; Price, R. J.; Tonelli, A. P.; Lake, B. G. *Drug Metab. Dispos.* **2001**, *29*, 1146; (g) Wu, W. N.; McKown, L. A.; Liao, S. *Xenobiotica* **2002**, *32*, 411; (h) Leppert, W.; Luczak, J. *Support Care Cancer* **2005**, *13*, 5; (i) Duhmke, R. M.; Cornblath, D. D.; Hollingshead, J. R. *Cochrane Database Syst. Rev.* **2004**, *2*, CD003726; (j) Grond, S.; Sablotzki, A. *Clin. Pharmacol. Ther.* **2004**, *43*, 879; (k) Klotz, U. *Arzneim.-Forsch.* **2003**, *53*, 681.
2. PDR Electronic Library, Thomson Scientific, http://www.thomsonhc.com/pdrel/librarian, reference for Ultram® tablets (Ortho-McNeil).
3. Raffa, R. B.; Nayak, R. K.; Liao, S.; Minn, F. L. *Rev. Contemp. Pharmacother.* **1995**, *6*, 485.
4. (a) Wilberg, K. B. *Chem. Rev.* **1955**, *55*, 713; (b) Westheimer, F. H. *Chem. Rev.* **1961**, *61*, 265.
5. Parli, C. J.; McMahon, R. E. *Drug Metab. Dispos.* **1973**, *1*, 337.
6. Tanabe, M.; Yasuda, D.; LeValley, S.; Mitoma, C. *Life Sci.* **1969**, *8*, 1123.
7. Elison, C.; Rapoport, H.; Laursen, R.; Elliott, H. W. *Science* **1961**, *134*, 1078.
8. NMR data for **16** (**D3**): ^{1}H NMR (400 MHz, CD_3OD) 7.36–7.31 (m, 1H), 7.16–7.11 (m, 2H), 6.85–6.83 (m, 1H), 3.04–2.97 (m, 1H), 2.74–2.64 (m, 7H), 2.34–2.26 (m, 1H), 1.97–1.60 (m, 8H); ^{13}C NMR (100 MHz, CD_3OD) 161.3, 150.5, 130.6, 118.2, 113.0, 112.2, 75.8, 61.8, 46.2, 42.8, 42.6, 41.6, 27.3, 26.0, 22.4; mp: 180–184 °C; TLC (10% MeOH in CH_2Cl_2) R_f = 0.48. NMR data for **17** (**D6**): ^{1}H NMR (400 MHz, CD_3OD) 7.30–7.27 (m, 1H), 7.10–7.05 (m, 2H), 6.83–6.80 (m, 1H), 3.79 (s, 3H), 2.98–2.93 (m, 1H), 2.66–2.62 (m, 1H), 2.25–2.20 (m, 1H), 1.94–1.51 (m, 8H); ^{13}C NMR (100 MHz, CD_3OD) 162.3, 151.4, 131.6, 119.1, 113.8, 113.2, 76.7, 62.6, 56.5, 43.8, 42.4, 27.9, 27.0, 23.3; mp: 175–180 °C; TLC (10% MeOH in CH_2Cl_2) R_f = 0.43.
9. NMR data for **18** (**D6 M1**): ^{1}H NMR (400 MHz, CD_3OD) 7.14–7.11 (m, 1H), 6.90–6.88 (m, 2H), 6.62–6.60 (m, 1H), 2.91–2.85 (m, 1H), 2.61 (dd, J = 2.57, 13.2 Hz, 1H), 2.13–2.07 (m, 1H), 1.94–1.43 (m, 8H); ^{13}C NMR (100 MHz, CD_3OD) 158.8, 150.5, 130.6, 117.1, 114.7, 113.2, 75.8, 61.8, 43.1, 41.5, 27.1, 26.2, 22.4; mp: 221–226 °C; TLC (20% MeOH in CH_2Cl_2) R_f = 0.23.
10. Draper, R. W.; Hou, D.; Iyer, R.; Lee, G. M.; Liang, J. T.; Mas, J. L.; Vater, E. J. *Org. Process Res. Dev.* **1998**, *2*, 186.
11. NMR data for **19** (**D9**): ^{1}H NMR (400 MHz, CD_3OD) 7.27–7.24 (m, 1H), 7.09–7.04 (m, 2H), 6.80 (d, J = 8.07 Hz, 1H), 2.94 (dd, J = 9.16, 13.2 Hz, 1H), 2.21–2.16 (m, 1H), 1.95–1.50 (m, 8H); ^{13}C NMR (100 MHz, CD_3OD) 161.0, 150.1, 130.5, 117.9, 112.7, 112.0, 75.6, 61.5, 42.8, 41.3, 27.0, 25.9, 22.1; mp: 176–182 °C; TLC (10% MeOH in CH_2Cl_2) R_f = 0.45.
12. General procedure for microsomal stability assay in Human liver microsomes: The test compounds at 23 μM were incubated separately with human liver microsomes (4 mg protein/mL) at 37 ± 1 °C in 3-mL incubation mixtures containing potassium phosphate buffer (50 mM, pH 7.4), $MgCl_2$ (3 mM), and EDTA (1 mM). Reactions were started by the addition of the NADPH-generating system. At designated times (0, 90, and 180 min), a 500-μL aliquot was removed from the incubation and added to 500 μL acetonitrile to terminate the reaction. The amount of unchanged test compound was quantified by LC/MS/MS. Where appropriate, the amount of *O*-desmethyl metabolite formed was also assessed by LC/MS/MS.
13. General procedure for in vitro stability assay in human hepatocytes. Test compounds (23 μM) were incubated separately with a pool (n = 2) of cryopreserved human hepatocytes (2 million cells/mL) in Waymouth's+ (Waymouth's medium [without phenol red] supplemented with FBS (4.5%), insulin (5.6 μg/mL), glutamine (3.6 mM), sodium pyruvate (4.5 mM), and dexamethasone (0.9 μM)) at the final concentrations indicated. Each test article was added to incubations in 2.5 μL (1%) of methanol. Reactions were started when placed in the incubator. At designated times (0, 180, and 360 min), a 250-μL aliquot was removed from the incubation and added to 250 μL acetonitrile to terminate the reaction. Precipitated protein was removed by centrifugation (920g for 10 min at 10 °C). Amount of unchanged parent compound was quantified as follows; an aliquot (20 μL) of the supernatant fraction was transferred to 1 mL of internal standard (50 ng/mL dextrorphan in 1:1 methanol/water, 51-fold dilution) and analyzed by LC/MS/MS. Additional incubations were performed with hepatocytes in which the test article was replaced with 7-ethoxycoumarin (500 μM, marker substrate) to determine if the hepatocytes were metabolically competent.

DOCKET ALARM
Find authenticated court documents without watermarks at docketalarm.com.