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N,N-Diethyl-4-[(3-hydroxyphenyl)(piperidin-4-yl)amino] benzamide derivatives: The development of diaryl amino piperidines as potent δ opioid receptor agonists with in vivo anti-nociceptive activity in rodent models

Paul Jones [a], Andrew M. Griffin [a,*], Lars Gawell [a], Rico Lavoie [a], Daniel Delorme [a], Edward Roberts [a], William Brown [a], Christopher Walpole [a], Wenhau Xiao [b], Jamie Boulet [b], Maryse Labarre [c], Martin Coupal [c], Joanne Butterworth [c], Stephane St-Onge [c], Lejla Hodzic [c], Dominic Salois [c]

[a] Department of Medicinal Chemistry, AstraZeneca R&D Montréal, 7171 Frédérick-Banting, Ville St. Laurent, Québec, Canada H4S 1Z9
[b] Department of Bioscience, AstraZeneca R&D Montréal, 7171 Frédérick-Banting, Ville St. Laurent, Québec, Canada H4S 1Z9
[c] Department of InVitro Biology & DMPK, AstraZeneca R&D Montréal, 7171 Frédérick-Banting, Ville St. Laurent, Québec, Canada H4S 1Z9

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ABSTRACT

We have investigated a series of phenolic diaryl amino piperidine delta opioid receptor agonists, establishing the importance of the phenol functional group and substitution on the piperdine nitrogen for delta agonist activity and selectivity versus the mu and kappa opioid receptors. This study uncovered compounds with improved agonist potency and selectivity compared to the standard, non-peptidic delta agonist SNC-80. In vivo anti-nociceptive activity of analog **8e** in two rodent models is discussed, demonstrating the potential of delta agonists to provide a novel mechanism for pain relief.

The discovery of the delta receptor as a member of the opioid family of receptors and its implication in pain pathways highlighted the potential importance of the delta receptor as a novel treatment for pain.[1,2] Early studies demonstrated delta receptor mediated analgesia using delta selective peptidic agonists,[2–4] with the potential to provide pain relief without the limiting side effects associated with the commonly used mu opioid receptor agonists such as morphine and fentanyl (respiratory depression, tolerance and physical dependence). More recently, Adolor published information on their development delta agonist, ADL5859, for pain management.[4b] Increasingly, alternative therapeutic areas have been identified where delta receptor agonists play a beneficial role.[5] They have been shown to possess immunostimulatory activity,[6] involved in modulating anxiety and depression,[7] may provide a means of cardioprotection following ischemia[8] and have been reported to be involved in the treatment of irritable bowel syndrome.[9]

As part of our earlier contributions in the development of selective, non-peptidic delta agonists we reported on two distinct series of compounds of general structure **1**, piperizines[10] and **2**, olefins[11] and we now report on a third class of compounds of general structure **3**, aminopiperidines (Fig. 1). Series **3** is related to piperazine series **1** via inversion of the benzylic carbon and piperazine nitrogen and was first reported by our group,[12] followed shortly by Carson and Podlogar.[13] We herein report on our SAR studies in this series, focussing on the effects of modifying substitution on the piperidine nitrogen.

A synthetic scheme was developed to allow access to both the methyl ether **7** and phenol **8** amino piperidine analogs (Scheme 1). Secondary amine **4** was synthesized under standard reductive amination conditions using 3-methoxyaniline and 1-benzylpiperidin-4-one. Palladium N-arylation of amine **4** was achieved using xantphos[14] as the ligand as it gave a superior yield (85%) of biaryl amine **5** compared to using BINAP (~60%) where the reaction could not be forced to completion, even after prolonged heating. The *N*-benzyl group was removed with 1-chloroethyl chloroformate to yield amine **6** and the southern group installed via reductive amination or via alkylation using the desired alkyl halide. Final cleavage of the methyl ether **7** to generate the phenol **8** was achieved using boron tribromide. All test compounds were purified by reverse-phase chromatography with a water/acetonitrile gradient containing 0.05% TFA v/v.[15]

The pharmacological profiles of the compounds were determined by radioligand binding assays. The binding affinities of all compounds were determined using cloned human δ, μ, κ receptors and the agonist potencies (EC_{50}) at the delta receptor were measured using a GTP [γ-^{35}S] binding assay.[16] In all experiments the known non-peptic delta agonist SNC-80[17] was included as a control and the efficacy of compounds in the [^{35}S] GTPγS binding

* Corresponding author. Tel.: +1 514 832 3200; fax: +1 514 832 3232.
E-mail address: andrew.griffin@astrazeneca.com (A.M. Griffin).

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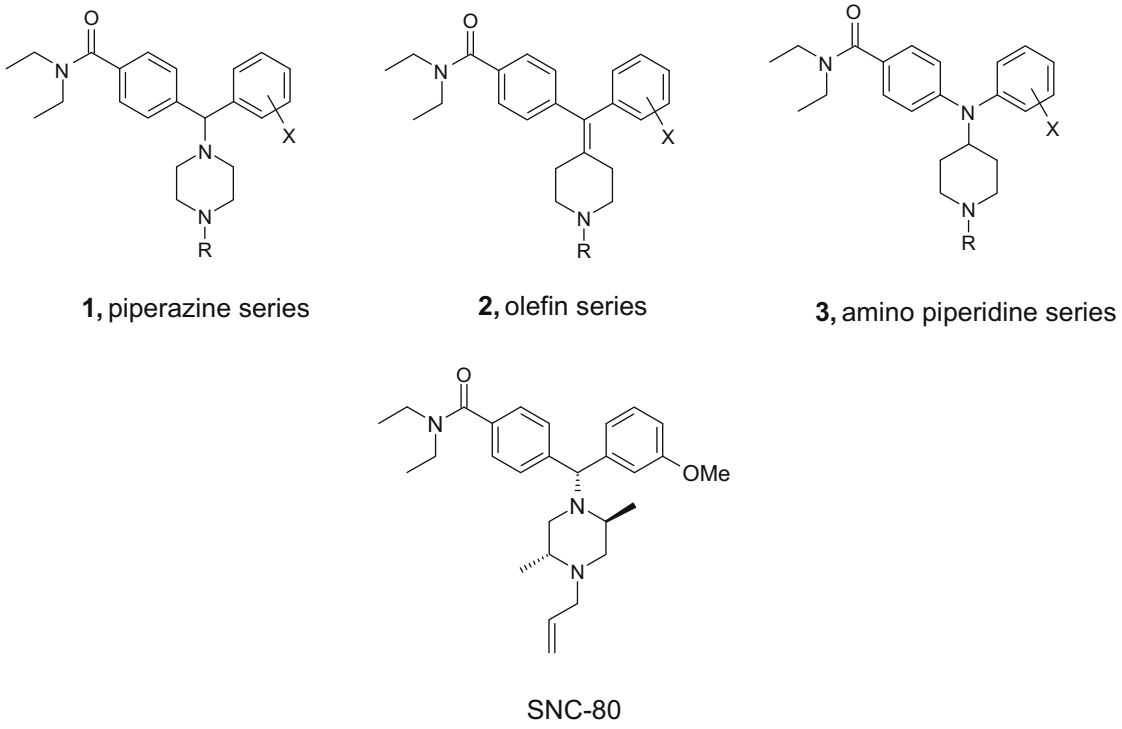
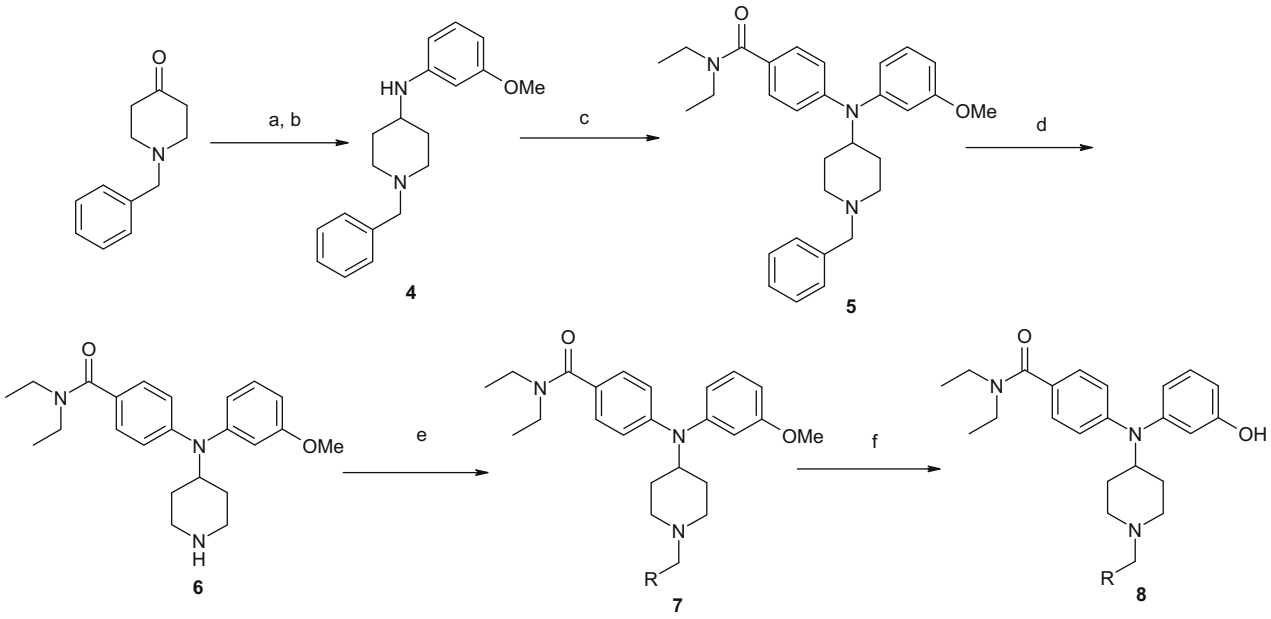


Figure 1.



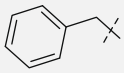
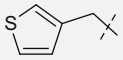
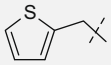
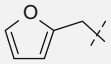
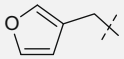
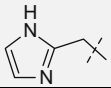
Scheme 1. Reagents and conditions: (a) 3-methoxyaniline, $Ti(O^iPr)_4$; (b) $NaBH_4$, ethanol; (c) 4-bromo-*N*,*N*-diethylbenzamide, $Pd_2(dba)_3$, xanthphos, NaO^tBu, toluene, 110 °C; (d) (1) $CH_3CHClOCOCl$, $Cl(CH_2)_2Cl$; (2) MeOH, reflux; (e) RCH_2Br/RCH_2Cl, triethylamine, CH_2Cl_2 or RCHO, MeOH, $NaBH_3CN$, AcOH; (f) 1 N BBr_3 in CH_2Cl_2.

assay calculated in relation to the response from SNC-80 (E_{max} 100%).

Representative data for methyl ethers **7** is shown in Table 1, along with data for the reference delta agonist SNC-80 (see Fig. 1 for structure). All compounds showed reduced binding affinity and agonist activity at the delta receptor compared to SNC-80 and although compounds evaluated in the GTP [γ-^{35}S] binding assay were all full agonists, the most potent analog 2-thiophene **7c** was five times less potent than SNC-80. It is of interest to note the loss of delta activity when compared to SNC-80 given that SNC-80 is also a methyl ether, although from the piperazine series of delta agonists. All analogs demonstrated selective binding to the delta receptor over mu and kappa receptors.

Turning our attention to phenol derivatives **8**, we initially investigated the effect of introducing alkyl groups onto the southern piperidine nitrogen as shown in Table 2. Secondary amine **8a**, while selective for the delta receptor, showed reduced agonist potency compared to the alkyl derivatives **8b, 8c** and **8d**, demonstrating the need for substitution on the piperidine nitrogen. Introduction of the allyl group, **8b**, and the cyclopropyl group **8c**, gave

Table 1
Binding affinity and δ agonist activity of selected methyl ethers **7**

Compound	R	Binding affinities (K_i nM)			δ Agonist activity	
		δ	μ	κ	EC_{50} (nM)	E_{Max} (%)
	SNC-80	1.29 ± 0.13	352 ± 43	4169 ± 829	2.99 ± 0.06	101 ± 1
7a		49.6 ± 30.0	2106 ± 315	3606 ± 876	141 ± 18	101 ± 5
7b		18.2 ± 0.9	1596 ± 22	2549 ± 635		
7c		11.1 ± 1.2	1963 ± 582	6405 ± 1071	17.4 ± 4.0	173 ± 62
7d		50.2 ± 8.1	2533 ± 214	7081 ± 1790		
7e		14.0 ± 0.8	1299 ± 62	4985 ± 998	86.5 ± 22.9	115 ± 11
7f		22.8 ± 1.5	4445 ± 287	>10,000	125 ± 44	193 ± 82

similar profiles while the cyclohexyl derivative showed a drop in delta binding and activation. None of these derivatives showed improvements in activity at the delta receptor in comparison to SNC-80.

In our previous work in the piperazine series **1**[10] and olefin series **2**[11] we did not investigate the effects of a southern aromatic or heteroaromatic group on delta activation and selectivity. Such modifications were introduced in the current series and data reported in Table 3. Benzyl derivative **8e** shows enhanced agonist potency versus the corresponding methyl ether **7a** (0.56 nM vs 141 nM), highlighting the importance of the phenol functionality for good agonist activity at the delta receptor. Furthermore, when comparing the potency of southern benzyl substituted **8e** to the alkyl derivatives in Table 2, a positive effect of the southern aromatic group on delta binding and activation is seen. Compound **8e** also showed improved delta agonist potency compared to SNC-80, although selectivity over mu and kappa opioid receptors was decreased. SAR around **8e** was expanded to investigate effects of substituents on the southern aromatic ring (agonist activity and selectivity over mu and kappa) and also to determine if heterocycles were tolerated in this position (see Table 3). The incorporation of heterocycles would enable us to modulate the physical chemical properties of the molecules (aqueous solubility, lipophilicity).

Substitution at the benzylic position with a methyl group, **8f**, was not tolerated and showed a sevenfold drop in agonist potency compared to **8e**. Analogs **8g** to **8i** demonstrated that substitution on the aromatic ring effected binding at the mu receptor: *p*-Me **8g** showed enhanced mu binding (62 nM) whereas *o*-F **8h** had decreased binding at mu (162 nM) as well as *m*-F **8i** (472 nM). Both **8h** and **8i** had improved delta/mu selectivity compared to unsubstituted phenyl derivative **8e**, demonstrating that the introduction of a small group on the ring can decrease mu binding while maintaining activity at the delta receptor.

Heterocyclic derivatives were more extensively studied and they generally had improved selectivity for delta/mu as well as gave having improved physical chemical properties (data not shown). For thiophene and furan substitution the 3-isomer is preferred over the 2-isomer and the 2-imidazole **8n** 12 times more potent that regiomeric imidazole **8p**. Methylation of 2-imidazole **8n** to give **8r** proved to be detrimental, causing a 12-fold drop in agonist potency. Overall most of the heterocyclic analogs had improved delta agonist potency compared to SNC-80 (see Table 1). Of all the heterocyclic derivatives investigated, only the 3-furyl **8j**, 3-thiophenyl **8k** and 2-pyridinyl **8o** were more potent delta agonists than benzyl substituted **8e**.

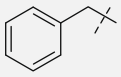
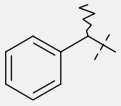
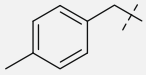
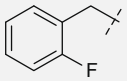
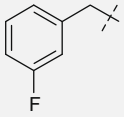
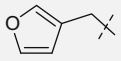
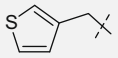
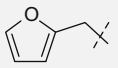
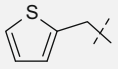
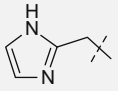
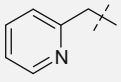
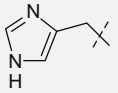
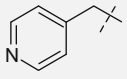
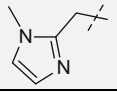
Within our program, phenol **8e** was identified as a candidate of interest and was assessed in two rodent models of nociception. Compound **8e** demonstrated excellent binding affinity, agonist potency and good selectivity for the delta receptor, as well as having a moderate pharmacokinetic profile the in rat (half-life 0.8 h; bioavailability, 25%).

Phenol **8e** was evaluated in vivo in both the mouse abdominal constriction antinociception model for acute pain and the rat carrageenan model for acute inflammatory hyperalgesia. In the mouse acetic acid induced abdominal constriction model, **8e** produced a dose dependent response with an ED_{50} of 21.5 ± 1.5 μmol/kg iv and a maximum inhibition of writhing of 57 ± 9% at 25 μmol/kg iv.[18] The pre-administration of the delta selective antagonist naltrindole blocked the observed anti-nociceptive response confirming the analgesic effect is mediated through activation of the delta receptor. In the rat carrageenan model, **8e** produced a dose dependent response with an ED_{50} of 17.5 μmol/kg iv and a maxi-

Table 2
Binding affinity and δ agonist activity of phenols **8** with *N*-alkyl substitution

Compound	R	Binding affinities (K_i nM)			Selectivity ratios		δ Agonist activity	
		δ	μ	κ	μ/δ	k/δ	EC_{50} (nM)	E_{Max} (%)
8a	H	0.99 ± 0.05	87 ± 9	901 ± 67	88	910	25.6 ± 3.2	105 ± 5
8b	Allyl	1.03 ± 0.07	130 ± 23	98 ± 9	127	96	4.13 ± 0.92	77 ± 2
8c	CH_2-cyclohexyl	2.30 ± 0.23	289 ± 60	726 ± 26	125	315	18.9 ± 2.9	106 ± 8
8d	CH_2-cyclopropyl	0.97 ± 0.05	126 ± 24	508 ± 91	129	522	3.40 ± 0.19	80 ± 10

Table 3
Binding affinity and δ agonist activity of phenols **8** with *N*-benzyl and *N*-CH_2heterocycle substitution

Compound	R	Binding affinities (K_i nM)			Selectivity ratios		δ Agonist activity	
		δ	μ	K	μ/δ	κ/δ	EC_{50} (nM)	E_{Max} (%)
8e		0.42 ± 0.03	88 ± 11	744 ± 127	210	1771	0.56 ± 0.07	100 ± 2
8f		1.40 ± 0.28	648 ± 122	2191 ± 506	463	1564	3.79 ± 0.26	101 ± 14
8g		0.64 ± 0.02	62 ± 9	726 ± 63	97	1133	1.14 ± 0.10	105 ± 5
8h		0.76 ± 0.05	162 ± 41	967 ± 55	342	1266	1.13 ± 0.38	111 ± 2
8i		0.70 ± 0.05	472 ± 33	1358 ± 90	670	1929	2.63 ± 0.48	92 ± 2
8j		0.22 ± 0.01	177 ± 21	121 ± 15	802	550	0.22 ± 0.02	105 ± 13
8k		0.27 ± 0.01	127 ± 15	495 ± 165	470	1825	0.22 ± 0.02	97 ± 3
8l		0.42 ± 0.02	115 ± 17	263 ± 23	276	632	1.29 ± 0.28	101 ± 5
8m		0.65 ± 0.10	181 ± 28	453 ± 71	278	696	1.41 ± 0.43	104 ± 5
8n		0.30 ± 0.02	1251 ± 110	1189 ± 204	4168	3959	1.17 ± 0.59	118 ± 14
8o		0.24 ± 0.02	205 ± 26	2043 ± 220	854	8523	0.25 ± 0.03	98 ± 4
8p		1.61 ± 0.08	309 ± 55	1451 ± 124	192	901	14.1 ± 1.6	105 ± 8
8q		1.02 ± 0.03	1549 ± 210	4568 ± 460	1514	4465	5.87 ± 2.05	111 ± 17
8r		1.70 ± 0.11	772 ± 197	4595 ± 405	452	2691	14.5±3.7	92 ± 6

mum anti-hyperalgesic effect at 30 μmol/kg iv with 60 ± 12% reversal.[19] It is of interest to note that no convulsant activity was observed with **8e** at doses administered in the abdominal constriction model and carrageenan model. This is contrary to reports for SNC-80 and BW373U86, where convulsant activity was seen at similar doses producing an anti-nociceptive effect.[20] Phenol **8e** did however produce hyperactivity at doses above 30 μmol/kg iv.

In summary, we have continued our earlier work in the development of non-peptic delta receptor agonists and now report on SAR studies in the amino piperidine series **3** (Fig. 1). This work has shown that within this series of agonists, the phenol is crucial for potent agonist activity and that the agonist activity is also modulated by substitution on the piperidine nitrogen. The incorporation of pendent aromatic and heteroaromatic groups has been found to give analogs that have improved potency at the delta receptor and improved selectivity over the mu and kappa receptors compared to the known standard delta agonist, SNC-80. We also describe in vivo data showing the anti-nociceptive effect of **8e** in two rodent models, the mouse abdominal constriction model and in the rat carrageenan, demonstrating the potential of delta agonists to provide a novel mechanism for pain relief.

Further studies from our group within this series of delta agonists, focussing on modifications of the phenol group are reported in the following paper.

References and notes

1. Heyman, J. S.; Vaught, J. L.; Raffa, R. B.; Porreca, F. *Trends Pharmacol. Sci.* **1988**, *9*, 134.
2. Rapaka, R. S.; Porreca, F. *Pharm. Res.* **1991**, *8*, 1.
3. Moulin, D. E.; Max, M. B.; Kaiko, R. F.; Inturrisi, C. E.; Maggard, J.; Yaksh, T. L.; Foley, K. M. *Pain* **1985**, *23*, 213.
4. (a) Galligan, J. J.; Mosberg, H. I.; Hurst, R.; Hruby, V. J.; Burks, T. F. *J. Pharmacol. Exp. Ther.* **1984**, *229*, 641; (b) Le Bourdonnec, B.; Windh, R. T.; Ajello, C. W.; Leister, L. K.; Gu, M.; Chu, G.-H.; Tuthill, P. A.; Barker, W. M.; Koblish, M.; Wiant, D. D.; Graczyk, T. M.; Belanger, S.; Cassel, J. A.; Feschenko, M. S.; Brogdon, B. L.; Smith, S. A.; Christ, D. D.; Derelanko, M. J.; Kutz, S.; Little, P. J.; DeHaven, R. N.; DeHaven-Hudkins, D. L.; Dolle, R. E. *J. Med. Chem.* **2008**, *51*, 5893.
5. Coop, A.; Rice, K. C. *Drug News Perspect.* **2000**, *13*, 481.
6. Bhargava, H. N.; House, R. V.; Thomas, P. T. *Analgesia* **1995**, *1*, 302.
7. (a) Saitoh, A.; Kimura, Y.; Suzuki, T.; Kawai, K.; Nagase, H.; Kamei, J. *J. Pharmacol. Sci.* **2004**, *95*, 374; (b) Broom, D. C.; Jutkiewicz, E. M.; Folk, J. E.; Traynor, J. R.; Rice, K. C.; Woods, J. H. *Neuropsychopharmacology* **2002**, *26*, 744; (c) Tejedor-Real, P.; Micó, J. A.; Smadja, C.; Maldonado, R.; Roques, B. P.; Gibert-Rahola, J. *Eur. J. Pharmacol.* **1998**, *354*, 1.
8. (a) Bell, S. P.; Sack, M. N.; Patel, A.; Opie, L. H.; Yellon, D. M. *J. Am. Colloid. Cardiol.* **2000**, *36*, 2296; b Schultz, J. L.; Garrett, G. US Patent. 6,103,722.
9. Middleton, D. S.; Maw, G. N.; Challenger, C.; Jessiman, A.; Johnson, P. S.; Million, W. A.; Nichols, C. L.; Price, J. A.; Trevethick, M. *Bioorg. Med. Chem. Lett.* **2006**, *16*, 905.
10. Plobeck, N.; Delorme, D.; Wei, Z.-Y.; Yang, H.; Zhou, F.; Schwarz, P.; Gawell, L.; Gagnon, H.; Pelcman, B.; Schmidt, R.; Yue, S.-Y.; Walpole, C.; Brown, W.; Zhou, E.; Labarre, M.; Payza, K.; St-Onge, S.; Kamassah, A.; Morin, P.-E.; Projean, D.; Ducharme, J.; Roberts, E. *J. Med. Chem.* **2000**, *43*, 3878.
11. Wei, Z.-Y.; Brown, W.; Takasaki, B.; Plobeck, N.; Delorme, D.; Zhou, F.; Yang, H.; Jones, P.; Gawell, L.; Gagnon, H.; Schmidt, R.; Yue, S.-Y.; Walpole, C.; Payza, K.; St-Onge, S.; Labarre, M.; Godbout, C.; Jakob, A.; Butterworth, J.; Kamassah, A.; Morin, P.-E.; Projean, D.; Ducharme, J.; Roberts, E. *J. Med. Chem.* **2000**, *43*, 3895.
12. Roberts, E; Pelcman, B. WO 98/28270.
13. (a) Carson, J. R.; Carmosin, R. J.; Fitzpatrick, L. J.; Reitz, A. B.; Jetter, M. C. WO 99/33806.; (b) Podlogar, B. L.; Poda, G. I.; Demeter, D. A.; Zhang, S.-P.; Carson, J. R.; Neilson, L. A.; Reitz, A. B.; Ferguson, D. M. *Drug Des. Disc.* **2000**, *17*, 34.
14. Guari, Y.; van Es, D. S.; Reek, J. N. H.; Kamer, P. C. J.; van Leeuwen, P. W. N. M. *Tetrahedron Lett.* **1999**, *40*, 3789.
15. All products gave satisfactory analytical characterization showing purity >95% as determined by HPLC using a Zorbax C-18 column. ^{1}H NMR spectra were obtained from a 400 MHz Varian Unity Plus spectrometer. Mass spectra were obtained on a Micromass Quattro micro API or Agilent 1100 Series LC/MSD instrument using loop injection. Selected analytical characterizations; *compound* **8e**: ^{1}H NMR (CD_3OD) δ_H 1.16 (br s, 6H), 1.69–1.73 (m, 2H), 2.22 (d, J = 14 Hz, 2H), 3.27 (t, J = 8 Hz, 2H), 3.35–3.51 (m, 6H), 4.27 (s, 2H), 4.28–4.35 (m, 1H), 6.69 (d, J = 9 Hz, 3H), 7.20–7.23 (m, 4H), 7.46 (s, 6H). MS (ESI) m/z 458.23 (MH^+); *compound* **8o**: ^{1}H NMR (400 MHz, DMSO-d_6) δ_H 1.07 (t, 6H), 1.66–1.75 (m, 2H), 2.01–2.05 (m, 2H), 3.21–3.46 (m, 8H), 4.20–4.23 (m, 1H), 4.39 (s, 2H), 6.37–39 (m, 1H), 6.45–6.48 (m, 1H), 6.64 (d, J = 8.4 Hz, 2H), 6.66–6.68 (m, 1H), 7.18 (d, J = 8.4 Hz, 2H), 7.21–7.24 (m, 1H), 7.43–7.51 (m, 2H), 7.88–7.92 (s, 1H), 8.62–8.63 (m, 1H), 9.53 (br s, 1H), 9.69 (br s, 1H). MS. (ESI) m/z 459.23 (MH^+).
16. *Receptor binding assays*. Membranes were combined with test compounds and approximately 0.07 nM of the appropriate radioligand [^{125}I]-[D-Ala2]-deltorphin II (K_d = 0.93 nM at delta), [^{125}I]-FK33824 (K_d = 1.14 nM at mu), and [^{125}I]-D-Pro10-Dynorphin A[1-11] (K_d = 0.16 nM at kappa) in 50 mM Tris, 3 mM $MgCl_2$, 1 mg/ml BSA, pH 7.4. The amounts of bound radioactivity were determined at equilibrium by filtration. The nonspecific (NS) binding was defined in the presence of 10 μM naloxone. The IC_{50} values of test compounds were determined from 2-parameter logistic curve fits of percent specific binding versus log (molar ligand), solving for IC_{50} and hill slope.*GTP[γ]^{35}S binding assays*. Membranes expressing 10 pmol of hDOR per mg protein were combined with test compounds and approximately 0.2 nM GTP[γ]^{35}S in 50 mM Hepes, 20 mM NaOH, pH 7.4, 5 mM MgCl2, 100 mM NaCl, 1 mM EDTA, 1 mM DTT, 0.1% BSA, 15 μM GDP. The bound radioactivity was determined after 1 h by filtration. Control and stimulated binding were determined in the absence and presence of 3 μM SNC-80 respectively. Values of EC_{50} and E_{max} for ligands were obtained from 3-parameter logistic curve fits of percent stimulated GTP[γ]^{35}S binding versus log (molar ligand), solving for EC_{50} and hill slope, and % E_{max}.
17. (a) Calderon, S. N.; Rothman, R. B.; Porreca, F.; Flippen-Anderson, J. L.; McNutt, R. W.; Xu, H.; Smith, L. E.; Bilsky, E. J.; Davis, P.; Rice, K. C. *J. Med. Chem.* **1994**, *37*, 2125; (b) Calderon, S. N.; Rice, K. C.; Rothman, R. B.; Porreca, F.; Flippen-Anderson, J. L.; Kayakiri, H.; Xu, H.; Becketts, K.; Smith, L. E.; Bilsky, E. J.; Davis, P.; Horvath, R. *J. Med. Chem.* **1997**, *40*, 695; (c) Bilsky, E. J.; Calderon, S. N.; Wang, T.; Bernstein, R. N.; Davis, P.; Hruby, V. J.; McNutt, R. W.; Rothman, R. B.; Rice, K. C.; Porreca, F. *J. Pharmacol. Exp. Ther.* **1995**, *273*, 359; (d) DoCarmo, G. P.; Folk, J. E.; Rice, K. C.; Chartoff, E.; Carlezon; William, A.; Negus, S. S. *Eur. J. Pharmacol.* **2009**, *604*, 58; (e) Bosse, K. E.; Jutkiewicz, E. M.; Gnegy, M. E.; Traynor, J. R. *Neuropharmacology* **2008**, *55*, 755.
18. Compound **8e** was dosed iv in CD1 mice at doses ranging from 4 to 25 μmol/kg. Measurements of antinoceptive activity were taken 20 min post iv dose. For comparison the gold standard mu agonist morphine displayed an ED_{50} of 1.5 μmol/kg.
19. Compound **8e** was dosed iv in Sprague-Dawley rats at doses ranging from 3 to 30 μmol/kg. Measurements of antinoceptive activity were taken 30 min post iv dose. For comparison the gold standard mu agonist morphine displayed an ED_{50} of 3.1 μmol/kg.
20. (a) Hong, E. J.; Rice, K. C.; Calderon, S.; Woods, J. H.; Traynor, J. R. *Analgesia* **1998**, *3*, 269; (b) Broom, D. C.; Nitsche, J. F.; Pintar, J. E.; Rice, K. C.; Woods, J. H.; Traynor, J. R. *J. Pharmacol. Exp. Ther.* **2002**, *303*, . 723.