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Fentanyl analogues with a modified propanamido group as potential affinity labels: Synthesis and *in vivo* activity

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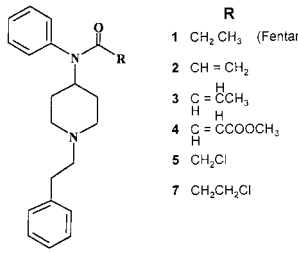
Ligands which form covalent bonds with a particular type or subtype of opioid receptors have provided useful tools for investigating opioid receptors and activities [1, 2]. This report describes the synthesis and analgesic activity (hot-plate test) of potential affinity labels of the μ-opioid fentanyl (**1**) [3]. In these derivatives (**2**–**5**), the propanamido group of fentanyl was modified to include an electrophilic center which may interact with a receptor nucleophile. Reaction of 4-anilino-1-(2-phenethyl)piperidine (**6**) [4] with 3-chloropropionyl chloride and TEA in CH_2Cl_2 for 48 h at RT afforded propenanilide **2**, apparently via the chloro derivative **7**. Shorter reaction time in absence of TEA provided **7** · HCl (m.p. 231–235 °C, acetone) Reaction of **6** with crotonyl chloride, (*E*)-4-chloro-4-oxo-2-butenic acid methyl ester [5] or chloroacetyl chloride gave **3**, **4** and **5**, respectively. Compounds **2**–**5** were isolated and purified as bases or HCl salts (Table). Mouse 55 °C hot-plate test was performed as reported [6]. Solutions of compounds **2**–**5** were prepared by adding equimolar amounts of citric acid to their base forms in water. These and reference solutions [fentanyl citrate (Janssen) and morphine sulfate (El-Kahira)] were administered i.p. (10 mL/kg). Naloxone HCl (Dupont) was used s.c.

Compound **2** showed a remarkable dose-related analgesic effect. When compared with fentanyl (at doses of less than 1 mg/kg), it was more potent and had a longer duration of action (Fig.). The analgesic effect of fentanyl (estimated at 0.1, 0.2 and 0.5 mg/kg) dropped significantly 60–70 min and became insignificant 90–100 min after administration. At comparable doses, **2** maintained considerable analgesia 90 and 120 min after administration. In its duration, the time-response profile of **2** resembled more closely that of morphine (20 mg/kg) than that of fentanyl. At higher doses, **2** showed a maximum possible effect (MPE; animals not responding to nociception within 60 s) that was retained for several hours without signs of opioid toxicity. Thus, at a dose of 6.8 mg/kg, 50% of the animals maintained a MPE unaltered for 4.5 h. At 17 mg/kg, 100% of the animals maintained a MPE for 3 h and 50% of these animals continued as such for 6 h. Motor activity was inhibited after a dose of 25 mg/kg, however, animals were not cataleptic and returned to contineous circuling behaviour 3.5 h after treatment. Convulsions developed 1 h after a dose of 50 mg/kg and 60% lethality was observed from apparent respiratory depression.

Naloxone (2 mg/kg) administered 30 min before **2** (0.85 mg/kg) blocked the effect of **2** for about 40 min, then the tested animals showed analgesia and other mor-

Table: Physical and spectral properties of fentanyl derivatives 2–5



Compd.	Yield (%)	m.p.[a] (°C) (solvent)	Formula[b]	Spectral data[c]
2[d]	67	101–103 (EtOAc)	$C_{22}H_{26}N_2O$	IR (KBr) 1615, 1655 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.6–7.0 (m, 10H, Ar*H*), 6.35 (d, J = 16.7 Hz, 1H, olefinic *H cis* to C=O), 5.81 (dd, J = 16.7 and 10.2 Hz, 1H, olefinic *H geminal* to C=O), 5.45 (d, J = 10.2 Hz, 1H, olefinic *H trans* to C=O), 4.74 (t, J = 12.4 Hz, W_H = 27.8 Hz, 1H, H-4), 3.25–1.0 (m, 12H); EIMS *m/z* 334 $(M)^+$ (0.1%), 243 $(M\text{-}CH_2Ph)^+$ (100%).
3 · HCl	21	202–204 (Me_2CO)	$C_{23}H_{28}N_2O \cdot HCl$	IR (KBr) 1625–1660 cm^{-1}; ^1H NMR (CD_3CN) δ 12.2 (br, 1H, N^+H), 7.5–6.8 (m, 10H, Ar*H*), 5.85 (m, 1H), 5.1–4.6 (m, 2H), 3.6–3.5 (m, 2H), 3.3–3.0 (m, 4H), 2.9–2.5 (m, 3H), 2.3–1.8 (m, 3H), 1.7 (d, 3H, CH_3); EIMS *m/z* 346 $(M\text{-}2)^+$ (0.9%), 257 $(M\text{-}CH_2Ph)^+$ (4.5%).
4	54	139–144 (CH_2Cl_2)	$C_{24}H_{28}N_2O_3$	IR (KBr) 1630, 1660, 1725 cm^{-1}; ^1H NMR ($CDCl_3$) δ 7.7–7.0 (m, 10H, Ar*H*), 6.83 (d, J = 15.2 Hz, 1H, C*H*COOMe), 6.57 (d, J = 15.2 Hz, 1H, C*H*CONPh), 4.71 (t, $J \approx$ 12 Hz, W_H = 30.5 Hz, 1H, H-4), 3.75 (s, 3H, OCH_3), 3.2–0.8 (m, 12H); EIMS *m/z* 392 $(M)^+$ (0.6%), 301 $(M\text{-}CH_2Ph)^+$ (100%).
5 · HCl[e]	57	250–253 (dec) (Me_2CO)	$C_{21}H_{25}ClN_2O \cdot HCl$[f]	IR (KBr) 1665 cm^{-1}; ^1H NMR ($CDCl_3$) δ 12.6 (N^+H), 7.6–7.0 (m, 10H, Ar*H*), 4.76 (t, J = 11.8 Hz, W_H = 27.9 Hz, 1H, H-4), 3.8–2.6 (m, 9H), 2.4–1.0 (m, 5H); EIMS *m/z* 265 $(M\text{-}CH_2Ph)^+$ (6.2%), 91 $(PhCH_2)^+$ (100%).

[a] Uncorrected, Griffin m.p. apparatus. [b] Analysed for C, H, N (±0.4% of theoretical values). [c] IR: Shimadzu 435; ^1H NMR: Bruker 200 (ppm from internal TMS); MS: Hewlett Packerd 5988. [d] HCl salt: m.p. 191–194 °C. [e] Base: m.p. 90–94 °C. [f] H: calculated 6.66%; Found 5.9%.

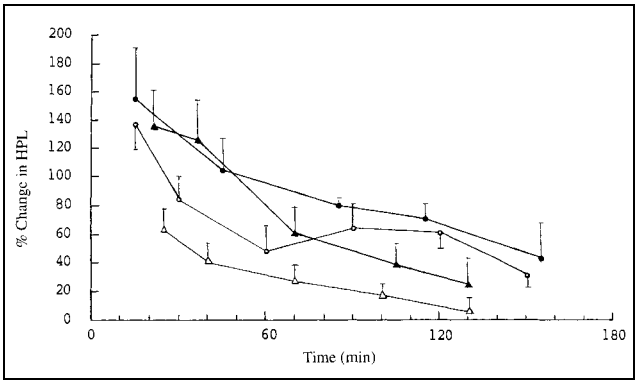


Fig.: % Changes in hot-plate latency (HPL) at several time intervals after i.p. administration at zero time of compound 2 (● 0.25, ○ 0.045 mg/kg) and of fentanyl (▲ 0.5, △ 0.1 mg/kg). Values are the mean ± the standard error of the mean. A cut off latency time of 30 s was used.

phine-like effects for about 50 min. Also, naloxone (2 mg/kg) administered 40 min after treatment of **2** (0.85 mg/kg) reversed its effects for 70 min, then the effects of **2** returned nearly to the same level (80% of MPE) as before naloxone-administration.

Compound **3** was clearly less potent than **2** or fentanyl, however, it also showed an extented duration of analgesia that was transiently reversed by naloxone. Compounds **2** and **3** were not tested for antagonistic activity. Methyl fumarate ester **4** produced no analgesia at any time after injection. A lethality of 50% (preceded by convulsions) was observed at a dose of 80 mg/kg. Also, **4** (at 20, 40 or 80 mg/kg) exhibited no inhibition of analgesia induced by morphine (20 mg/kg) as tested 2 and 24 h after administration of **4**. In addition, **4** (40 mg/kg) produced no inhibition of fentanyl-induced analgesia (0.25 mg/kg) as tested 2 h after its administration, Chloroacetanilide (**5**) showed only a weak and transient analgesia at 17 mg/kg.

Interpretation of the above results can be difficult because of pharmacokinetic and dispotional factors *in vivo*. It is conceivable, however, that the temporary reversal of the effect of **2** by naloxone was due to noncompetitive binding of naloxone to a regulatory site which allosterically inhibited the pharmacological response of covalently bound **2** until naloxone was eliminated [7, 8]. Because naloxone could not reverse the sustained effect of the opiate-derived irreversible agonist β-chloroymorphamine (β-COA) [9], it is likely that competetive antagonism in that case was essential. This implies that **2** has a mode of interaction with μ-receptors different from morphine [10]. Binding and *in vitro* studies should delineate this and other possibilities.

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References

1 Takemori, A. E.; Portoghese, P. S.: Annu. Rev. Pharmacol. Toxicol. **25**, 193 (1985)
2 Korlipara, V. L.; Takemori, A. E.; Portoghese, P. S.: J. Med. Chem. **38**, 1337 (1995)
3 Zhu, J.; Yin, J.; Law, P.-Y.; Claude, P. A.; Rice, K. C.; Evans, C. J.; Chen, C.; Yu, L.; Liu-Chen, L.-Y.: J. Biol. Chem. **271**, 1430 (1996)
4 Janssen, P. A. J.: Br. J. Anaesth. **34**, 260 (1962)
5 Acheson, R. M.; Feinberg, R. S.; Hands, A. R.: J. Chem. Soc. 526 (1964)
6 Eddy, N. B.; Leimbach, D.: J. Pharmacol. Exp. Ther. **107**, 385 (1953)
7 Cowi, A. L.; Kosterlitz, H. W.; Watt, A. J.; Nature (London) **220**, 1040 (1968)
8 Sayer, L. M.; Larson, D. L.; Takemori, A. E.; Portoghese, P. S.: J. Med. Chem. **27**, 1325 (1984)
9 Caruso, T. P.; Takemori, A. E.; Larson, D. L., Portoghese, P. S.; Science **204**, 316 (1979)
10 Portoghese, P. S.: J. Med. Chem. **8**, 609 (1965)

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