73. (−)-*N*-[(Cyclopropyl)methyl]-3,4-dimethoxy-5-methylmorphinan-6-one, an Opioid Agonist with Preference for Kappa Opioid Receptors

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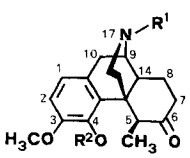
N-Allyl- and *N*-[(cyclopropyl)methyl]-3,4-dimethoxy-5-methylmorphinan-6-one (**9** and **10**, resp.) were synthesized from 5-methyldihydrothebainone (**1**). This essential intermediate was prepared from thebaine *via* 5-methylthebaine (**5**) employing a novel route. The pharmacological studies showed **9** and **10** to be potent opioid agonists. Compound **10** was found to have preference for kappa rather than mu opioid receptors.

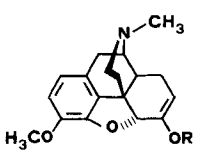
Introduction. – The opioid agonistic properties of *N*-methylmorphinan-6-ones are very much dependent on the substitution pattern at the aromatic ring [1]. Besides the 'natural' substitution pattern 3-hydroxy-4,5-epoxy, the 4-monomethoxy and the 3,4-dimethoxy substitution was found to be most effective. Among the compounds with high antinociceptive potency was 3,4-dimethoxy-5,17-dimethylmorphinan-6-one (**6**) [2], which prompted us to replace its N-CH_3 group by substituents known to introduce in most cases opioid antagonistic effects in morphinans. We chose the *N*-allyl and the *N*-(cyclopropyl)-methyl group which are present in the opioid antagonists naloxone and naltrexone, respectively.

Chemistry. – The essential intermediate, 5-methyldihydrothebainone (**1**), was synthesized by a route different from that described by *Small et al.* [3][4] who prepared **1** either from 8,14-dihydrothebaine (**2**) or 8,14-dihydrocodeinone enol acetate (**3**) with a *Grignard* reaction.

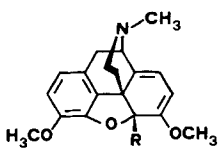
We introduced the 5-CH_3 group directly into thebaine (**4**) to obtain 5-methylthebaine (**5**). The procedure we followed was slightly different from that published by *Gates* and



1 $R^1 = CH_3$, $R^2 = H$
6 $R^1 = R^2 = CH_3$
7 $R^1 = CO_2CH_2CCl_3$, $R^2 = CH_3$
8 $R^1 = H$, $R^2 = CH_3$
9 $R^1 = CH_2{-}CH{=}CH_2$, $R^2 = CH_3$
10 $R^1 = CH_2CH(CH_2)_2$, $R^2 = CH_3$



2 $R = CH_3$
3 $R = COCH_3$



4 R = H (thebaine)
5 $R = CH_3$

coworkers [5]. We used dimethyl sulfate instead of methyl fluorosulfonate as alkylating agent. Catalytical hydrogenation over Pd/C in diluted AcOH, which was carried out similarly to the hydrogenation thebaine → dihydrothebainone [6][7], afforded **1** in 68% yield. This new synthesis of 5-methyldihydrothebainone (**1**) provides a route to this important intermediate which is much more efficient than the one developed by *Small* and coworkers [3][4].

O-Methylation of **1** was accomplished with phenyltrimethylammonium chloride using K_2CO_3 instead of NaH [2] as base. The dimethoxymorphinanone **6** was *N*-demethylated with 2,2,2-trichloroethyl chloroformate to give the carbamate **7**, which was cleaved reductively with Zn/NH_4Cl in MeOH to yield the *N*-normorphinanone **8**. Alkylation of **8** with allyl bromide and (cyclopropyl)methyl chloride in DMF afforded **9** and **10**, respectively.

Pharmacology. – In vivo *Studies*. Opioid antagonism was determined using the AcOH writhing antagonism test in mice *(Table 1)*. Compound **9** and **10** did not show antagonistic effects against morphine (selective mu agonist) and U-50.488 (selective kappa agonist).

Opioid agonism of **9** and **10** was studied in the AcOH writhing test and the hot-plate assay in mice *(Table 2)*. In addition to that, kappa agonism of **10** was evaluated in the diuresis test in rats *(Table 3)*.

Table 1. *Antagonistic Effects of* **9**, **10**, *and Reference Drugs*[a])

Compound	mg/kg sc.	% Reduction of analgesia induced by	
		morphine 10 mg/kg sc.	U-50.488 15 mg/kg sc.
9	30	0	0
10	30	0	0
Levallorphan	10	67	50
Naloxone	1	87	83

[a]) Determined by the AcOH writhing antagonism test in mice.

Table 2. *Antinociceptive Potencies of* **9**, **10**, *and Reference Drugs*

Compound	AcOH writhing test ED_{50} [mg/kg][a])	Hot-plate test, ED_{50} [mg/kg][a])
9	2.1 (0.82–5.2)	25 (18–34)
10	3.8 (1.5–9.4)	25 (10–60)
Morphine	0.52 (0.27–0.99)	2.9 (1.7–5.0)
Levallorphan	1.5 (0.24–9.3)	> 30
Naloxone	> 10	> 30

[a]) The ED_{50} values represent the effective dose at which 50% of the mice showed an analgesic response.

Table 3. *Effects of* **10** *and Reference Drugs on Diuresis in Rats*

Compound	mg/kg sc.	Excretion of urine, ml/kg/5 h (±SE)
10	10	40.7 (±1.7)
Morphine	10	20.8 (±1.5)
Levallorphan	1.0	26.6 (±1.3)
Naloxone	1.0	18.9 (±1.5)
U-50.488	1.0	45.1 (±3.6)
Control (NaCl)	–	18.6 (±1.1)

Compounds **9** and **10** showed relatively high antinociceptive potencies in the AcOH writhing test, whereas the potencies in the hot-plate assay were rather low. The effect of compound **10** on the diuresis of rats was significant, suggesting preference for kappa opioid receptors.

In vitro *Studies*. Compounds **9** and **10** were evaluated *in vitro* for opioid agonistic and antagonistic properties in opiod receptor binding assays and in the isolated guinea-pig ileal longitudinal muscle preparation. The ligands used in the binding assays were [^{3}H]naloxone (non-selective antagonist) and [^{3}H]tifluadom (kappa-selective agonist) *(Table 4)*. In guinea-pig ileum, the opioid agonistic effects were tested in the absence and presence of naloxone *(Table 5)*, the antagonistic effects against morphine (mu-selective agonist) and dynorphin A (kappa-selective agonist).

Table 4. *Effects of* **9**, **10**, *and Reference Drugs in Opioid Receptor Binding Assays*[a])

Compound	[^{3}H]Naloxone		[^{3}H]Tifluadom
	+NaCl	−NaCl	
9	550	600	80
10	170	120	17
Morphine	65.5	2.0	162
Levallorphan	1.1	0.54	0.67
Naloxone	2.0	2.0	9.0
U-50.488	12,000	2,150	5.0

[a]) The values are IC_{50} in nM. The unlabeled drugs were examined with at least 5 concentrations in duplicate in two independent determinations in the presence of [^{3}H]ligands.

Table 5. *Effects of* **9**, **10**, *and Reference Drugs in the Isolated Guinea-Pig Ileal Longitudinal Muscle Preparation*

Compound	Agonistic activity		
	EC_{50} [mol/l]	CR[a])	K_D [nmol/l][b])
9	2.1×10^{-7}	10.4	11.5
10	3.3×10^{-8}	8.1	14.4
Morphine	5.0×10^{-8}	74	1.4
Levallorphan	1.2×10^{-8}	7.8	14.6
U-50.488	5.5×10^{-10}	23.6	4.4

[a]) CR is the concentration ratio (EC_{50} of agonist in the presence of 10^{-7} mol/l of naloxone divided by EC_{50} of agonist alone).

[b]) K_D is the dissociation constant of the agonist, calculated from the equation K_D = [antagonist]/CR-1.

In the binding assays, compounds **9** and **10** exhibited low potencies in displacing [^{3}H]naloxone in the presence and in the absence of NaCl. In displacing [^{3}H]tifluadom from its binding sites, particularly compound **10** was quite effective, indicating preference for kappa over mu opioid receptors.

In the guinea-pig ileum, both **9** and **10** were potent opioid agonists. They were virtually inactive in antagonizing the effects of morphine and dynorphin A.

In conclusion, it was found in the AcOH writhing antagonism test and in the guinea-pig ileum that the compounds **9** and **10** do not possess opioid antagonistic properties. In the AcOH writhing test and in the guinea-pig ileum, **9** and **10** were found to have considerable agonistic properties. In addition to that, it was shown that compound **10**

exhibits kappa rather than mu opioid agonism. In the guinea-pig ileum, levallorphan and compound **9** also appeared to have kappa (rather than mu) agonistic activity, as judged by the inhibitory constant value of naloxone.

Experimental Part

Chemistry. – *General.* CC: basic alumina (70–230 mesh ASTM) from *Merck*. M.p.: *Kofler* melting-point microscope; uncorrected. Optical rotations: concentration in g/100 ml *Perkin Elmer* 141 polarimeter. IR spectra (in cm^{-1}): *Beckman Accu Lab 2* apparatus. ^{1}H-NMR spectra: *Jeol-JNM-PMX-60* spectrometer; δ in ppm relative to tetramethylsilane as internal reference, J (apparent coupling constant) in Hz. EI-MS: *Finnigan MAT 44S* apparatus.

5-Methylthebaine (**5**) [5]. BuLi (1.7M soln. in hexane, 30 ml) was added dropwise within 10 min to a soln. of thebaine (**4**; 10.0 g, 32.1 mmol) in 800 ml of anh. THF at −78° under N_2 while stirring (→ deep red soln.). Dimethyl sulfate (4.6 ml, 48.6 mmol) was added after 15 min at −78° at once. Then the soln. was allowed to come to r.t. over night (→ orange-red). After addition of 10 ml of H_2O, the soln. was evaporated, the oily residue dissolved in 250 ml of $CHCl_3$, washed with H_2O (2 × 150 ml) and brine (1 × 50 ml), dried, and evaporated to give a yellow-brown, crystalline residue (12.3 g). Recrystallization from 10 ml of EtOH gave 6.25 g of **5**. M.p. 160–162° ([5]: 158–159° (AcOEt)). The mother liquor was evaporated and chromatographed on basic alumina (grade II) with CH_2Cl_2 to give another 1.76 g. M.p. 154–157°. Total yield of **5**, 8.01 g (77%). ^{1}H-NMR ($CDCl_3$): 6.55 (*s*, 2 arom. H); 5.48 (*d*, J = 7, H–C(4)); 4.88 (*d*, J = 7, H–C(8)); 3.80 (*s*, CH_3O–C(3)); 3.52 (*s*, CH_3O–C(6)); 2.43 (*s*, CH_3N);.1.72 (*s*, CH_3–C(5)).

(−)-4-Hydroxy-3-methoxy-5,17-dimethylmorphinan-6-one (*= 5-Methyldihydrothebainone;* **1**) [3][4]. A mixture of **5** (5.0 g, 15.4 mmol), 10% Pd/C (400 mg), and 120 ml of 25% AcOH/H_2O was hydrogenated at 40 psi and r.t. for 20 h. The catalyst was filtered off and the filtrate cooled and basified with conc. NH_4OH. After extraction with CH_2Cl_2 (3 × 40 ml), the org. layer was dried and evaporated to yield 4.82 g of a slightly purple semi-crystalline residue which was recrystallized from EtOH to give 3.31 g (68%) of **1**. M.p. 188–191°. A portion of this material was recrystallized from EtOH to afford colorless needles. M.p. 190–192° ([3]: 192–193°). $[\alpha]_D^{20} = -23.4$ (c = 1.02, EtOH; [3]: $[\alpha]_D^{25} = -20.5$ (c = 1.026, EtOH)).

(−)-3,4-Dimethoxy-5,17-dimethylmorphinan-6-one Hydrobromide (**6**·HBr) [2]. A mixture of **1** (3.1 g, 9.83 mmol), K_2CO_3 (3.0 g, 21.7 mmol), phenyltrimethylammonium chloride (2.53 g, 14.7 mmol), and 20 ml of anh. DMF was stirred at 80° (bath temp.) under N_2 for 6 h. Then the mixture was poured on 500 ml of H_2O, extracted with CH_2Cl_2 (2 × 50 ml), the org. layer washed with H_2O (3 × 50 ml), dried, and evaporated to give 4.07 g of a slightly brown oil which was dissolved in *ca.* 5 ml of MeOH. After addition of 48% HBr soln. and *ca.* 2 ml of Et_2O, **6**·HBr (3.08 g, 76%) crystallized. M.p. 253–256° ([2]: 252–256°). This material was identical by mixed m.p., TLC, IR, and ^{1}H-NMR with an authentic sample.

(−)-3,4-Dimethoxy-5-methylmorphinan-6-one Hydrobromide (**8**·HBr). At r.t., 2,2,2-trichloroethyl chloroformate (7.74 ml, 54 mmol) was added dropwise to a mixture of **6**·HBr (4.0 g, 9.74 mmol), $KHCO_3$ (20 g, 0.19 mol), and 150 ml of EtOH-free $CHCl_3$ within 5 min. This mixture was stirred under reflux for 2 h, then filtered, and the filtrate was evaporated at 95° (bath temp.)/1 Torr. The resulting oily residue (6 g of **7**) was used for the next step without further characterization and purification. This oil was dissolved in 100 ml of boiling MeOH. After addition of NH_4Cl (12 g, 0.22 mol), activated Zn (12 g, 0.18 mol) was added in small portions within 5 min to the vigorously stirred mixture. After refluxing and stirring for additional 30 min, the mixture was filtered and the filtrate evaporated. The residue was partitioned between dil. NH_4OH and CH_2Cl_2, the org. layer dried and evaporated. To the resulting oily residue in MeOH, 48% HBr soln. and Et_2O were added to give 2.28 g (59%) of **8**·HBr. M.p. 250–253° (dec.). $[\alpha]_D^{20} = -11.9$ (c = 0.96, H_2O). IR (KBr): 3400 (NH_2^+), 1710 (CO). ^{1}H-NMR ($CDCl_3$): 9.20 (br. *s*, NH_2^+); 6.75 (*s*, 2 arom. H); 3.94, 3.78 (2*s*, 2 CH_3O); 1.36 (*d*, J = 7, CH_3–C(5)). EI-MS: 315 ($M^{+\cdot}$). Anal. calc. for $C_{19}H_{25}NO_3$·HBr (396.32): C 57.57, H 6.58, N 3.53; found: C 57.24, H 6.75, N 3.75.

(−)-17-Allyl-3,4-dimethoxy-5-methylmorphinan-6-one Hydrobromide (**9**·HBr) [2]. A mixture of **8** (800 mg, 2.02 mmol), allyl bromide (0.19 ml, 2.21 mmol), K_2CO_3 (1.0 g, 7.2 mmol), and 15 ml of anh. DMF was stirred under N_2 at 90° (bath temp.) for 45 min. The inorg. material was filtered off, the filtrate evaporated, and the residue partitioned between CH_2Cl_2 and H_2O. The org. layer was dried and evaporated to give a slightly brown oil which was converted into the HBr salt with 48% HBr soln.: 690 mg (78%) of **9**·HBr. M.p. 232–233° (dec.; acetone/Et_2O). $[\alpha]_D^{20} = -7.4$ (c = 1.35, $CHCl_3$). IR (KBr): 3440 (NH^+), 1705 (CO). ^{1}H-NMR 11.20 (br. *s*, NH^+); 6.74 (*s*, 2 arom. H); 5.40 (*m*, 3 olef. H); 3.90, 3.76 (2 *s*, 2 CH_3O); 1.45 (*d*, J = 7, CH_3–C(5)). EI-MS: 355 ($M^{+\cdot}$). Anal. calc. for $C_{22}H_{29}NO_3$·HBr (436.39): C 60.55, H 6.93, N 3.21; found: C 60.58, H 6.96, N 3.36.

(−)-17-[(Cyclopropyl)methyl]-3,4-dimethoxy-5-methylmorphinan-6-one Hydrobromide (**10**·HBr). A mixture of **8** (800 g, 2.02 mmol), (cyclopropyl)methyl chloride (0.20 ml, 2.4 mmol), K_2CO_3 (1.0 g, 7.2 mmol), and 10 ml of anh. DMF was stirred under N_2 at 100° (bath temp.) for 18 h. The inorg. material was filtered off and the filtrate evaporated. The residue was dissolved in CH_2Cl_2, washed with H_2O and brine, dried, and evaporated to give a slightly brown oil which was converted into the HBr salt in the usual way: 490 mg (54%) of **10**·HBr. M.p. 225–227° (dec.; acetone). $[\alpha]_D^{20} = -10.4$ (c = 1.0, $CHCl_3$). IR (KBr): 3400 (NH^+), 1700 (CO). ^{1}H-NMR ($CDCl_3$): 11.20 (br. *s*, NH^+); 6.75 (*s*, 2 arom. H); 3.92, 3.76 (2 *s*, 2 CH_3O); 1.44 (*d*, CH_3–C(5)): EI-MS: 369 ($M^{+\cdot}$). Anal. calc. for $C_{23}H_{31}NO_3$·HBr (450.41): C 61.32, H 7.16, N 3.11; found: C 61.43, H 7.21, N 3.36.

Pharmacology. – *Materials and Methods. Drugs Used.* Compounds **9** and **10** were used as HBr salts. Other compounds and their sources included: dynorphin A *(Bachem)*, levallorphan tartrate, U-50.488, and [^{3}H]tifluadom *(Roche)*, morphine·HCl *(Sandoz)*, naloxone·HCl *(Endo)*, [^{3}H]naloxone *(New England Nuclear)*, and naltrexone·HCl *(Endo)*. For *in vitro* experiments, the compounds were dissolved in the vehicles indicated in the respective methods. In *in vivo* experiments, they were dissolved in saline for parenteral injection and tap water for oral *(p.o.)* administration; sometimes, 1N HCl had to be added in order to obtain a soln. The volume of injection was 10 ml/kg in mice and 1.5 ml/kg in rabbits.

AcOH Writhing Test. This test was performed as described by *Witkin et al.* [8].

AcOH Writhing Antagonism Test. The test procedure corresponded to the AcOH writhing test [8], except that 20 min after the opioid agonist (morphine or U-50.488) was administered, the test compound was given.

Hot-Plate Test. This test was performed as described by *Woolfe et al.* [9].

Diuresis Test. Five to fifteen rats per dose were used. Immediately after sc. injection of the test compound or the vehicle, the animals were hydrated with NaCl (20 ml/kg *p.o.*) and placed into individual metabolism cages which permitted the collection of urine free from faecal contamination. Urine was collected in graduated glass cylinders over a period of 5 h. Thereafter, the urinary bladder was emptied. The average of urine volume per group was calculated and the mean value of the controls was taken as 100%. The results obtained with the test compounds are presented as % of the control group.

Opioid Receptor Binding Assays. [^{3}H]Naloxone binding was performed essentially as described by *Pert* and *Snyder* [10]. [^{3}H]Tifluadom binding was performed in homogenates of guinea-pig cerebellum as previously described by *Burkard et al.* [11] and *Gillan et al.* [12], respectively. IC_{50} values were determined graphically or by computer program assisted least squares fit of sigmoid curves. All experiments, performed in duplicate, were replicated at least once with similar results.

Isolated Guinea-Pig Ileal Longitudinal Muscle Preparation. The ileum of adult guinea pigs was removed and strips of longitudinal muscle were prepared from pieces of tissue 10 cm proximal to the ileo-caecal valve. The strips were suspended in a 10-ml organ bath containing *Krebs-Henseleit* soln. of the following composition (nmol/l): NaCl 115, KCl 4.7, $MgSO_4$ 1.2, $NaHCO_3$ 25, KH_2PO_4 1.2, glucose 10, and $CaCl_2$ 2.5. The soln. (pH 7.3) was aerated with 95% O_2/5% CO_2 (temp. 37°). The resting tension was adjusted to approximately 450 mg (4.4 mN) and isotonic contractions were recorded with *Shinko* (UL) transducer and displayed on a chart recorder (*Hellige*, Freiburg i. Br., FRG). Contractile responses were elicited by transmural electrical stimulation (0.1 Hz, 0.5 ms, 20 V). Prior to beginning an experiment, 10^{-6} mol/l of normophine were added to the bath to determine tissue reactivity. Any strip not responding with at least 60% reduction in twitch responses to electrical stimulation was discarded. *Ca.* 30–60 min was allowed, with frequent changes of the bathing soln., before addition of the compounds under study. Cumulative addition of three-fold increasing concentrations of test drug was used to determine concentration-response relationships. In experiments designed to test antagonistic effects of drugs, a 20 min incubation period preceded determination of agonist concentration-response curves.

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REFERENCES

[1] H. Schmidhammer, A. E. Jacobson, A. Brossi, *Med. Res. Rev.* **1983**, *3*, 1.
[2] A. Brossi, L. Atwell, A. E. Jacobson, M. D. Rozwadowska, H. Schmidhammer, J. L. Flippen-Anderson, R. Gilardi, *Helv. Chim. Acta* **1982**, *65*, 2394.
[3] L. Small, H. M. Fitch, W. E. Smith, *J. Am. Chem. Soc.* **1936**, *58*, 1457.
[4] L. Small, S. G. Turnbull, H. M. Fitch, *J. Org. Chem.* **1938**, *3*, 204.
[5] R. M. Boden, M. Gates, S. Peng Ho, P. Sundararaman, *J. Org. Chem.* **1982**, *47*, 1347.
[6] M. Freund, E. Speyer, *Chem. Ber.* **1920**, *53*, 2250.
[7] A. Skita, F. F. Nord, J. Reichert, P. Stukart, *Chem. Ber.* **1921**, *54*, 1560.
[8] L. B. Witkin, C. F. Heubner, F. Galdi, E. O'Keefe, P. Spitalett, A. J. Plummer, *J. Pharmacol. Exp. Ther.* **1961**, *133*, 400.
[9] G. Woolfe, A. D. McDonald, *J. Pharmacol. Exp. Ther.* **1944**, *80*, 300.
[10] C. B. Pert, S. H. Snyder, *Mol. Pharmacol.* **1974**, *10*, 868.
[11] W. P. Burkard, P. M. Müller, N. Flück, *J. Receptor Res.* **1984**, *4*, 165.
[12] M. G. C. Gillan, H. W. Kosterlitz, S. J. Paterson, *Br. J. Pharmacol.* **1980**, *70*, 481.