Opioid Agonist and Antagonist Activities of Morphindoles Related to Naltrindole

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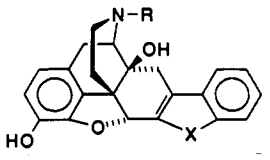
A series of naltrindole-related ligands (4–10) with an *N*-methyl, *N*-phenethyl, *N*-cinnamyl, or an unsubstituted basic nitrogen were synthesized and tested for opioid agonist and antagonist activity in smooth muscle preparations and in mice. The nor compounds (4 and 6) and the phenethyl derivatives (5 and 8) displayed full agonist activity (IC_{50} = 85–179 nM) in the mouse vas deferens preparation (MVD) while the other members of the series exhibited partial agonist or weak antagonist activity. In the guinea pig ileum preparation (GPI), all compounds except 8 were partial agonists. The ligands that were evaluated in mice were found to produce antinociception that was not selectively mediated via δ opioid receptors. However, two of these ligands (4 and 5) appeared to be δ-selective opioid receptor antagonists at subthreshold doses for antinociception. The finding that all of the compounds bind selectively to δ opioid receptors in guinea pig brain membranes together with the in vitro pharmacology and in vivo antagonist studies suggests that the lack of δ agonist selectivity in vivo may be due to a number of factors, including a basic difference between the δ receptor system in the MVD and in the mouse brain. Further, it is suggested that the constellation of message and address components in the morphindole nucleus may tend to stabilize δ receptors in the brain in an antagonist state.

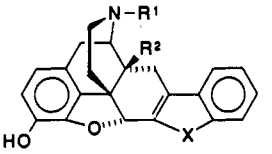
It is now well established that there are at least three major types of opioid receptors (μ, δ, and κ), although their physiologic roles are still not entirely understood.[1] Because the endogenous opioid peptides are not very selective for specific opioid receptor types, a variety of highly selective ligands have been developed as tools to sort out the effects mediated by these recognition sites. A number of δ-selective opioid agonists that are structurally related to enkephalin are employed routinely as probes,[2] but no nonpeptide ligands have been reported for this purpose. Such ligands would be useful, particularly when peripheral administration is desirable.

An initial effort to explore this objective involved the replacement of the cyclopropylmethyl group of naltrindole (NTI) 1 with a methyl group, as this substituent in opiate structures is usually associated with agonist activity. These *N*-methyl compounds (2 and 3) were reported to be devoid of antagonist activity in smooth muscle preparations and one of them, oxymorphindole (OMI) 2, was found to possess partial agonist activity and bind with high affinity and selectivity to δ sites.[3]



	R	X
1	$CH_2CH(CH_2)_2$	NH
2	CH_3	NH
3	CH_3	NCH_3

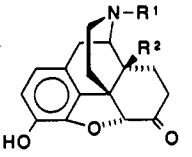
In light of these results we have investigated other N-substituted morphindoles 4–10 as an approach to developing full agonists that are δ selective both in vitro and in vivo. Here we present structure–activity studies directed toward this goal.



	R^1	R^2	X
4	H	OH	NH
5	$CH_2CH_2C_6H_5$	OH	NH
6	H	H	NH
7	CH_3	H	NH
8	$CH_2CH_2C_6H_5$	H	NH
9	$CH_2CH{=}CHC_6H_5$	H	NH
10	CH_3	OH	O

Chemistry

The indoles 4, 6, 7, and 9 were synthesized by reacting their corresponding ketones (11, 13–15) with phenylhydrazine under Fischer indolization conditions. The cinnamylnoroxymorphone 15 that was employed in the



	R^1	R^2
11	CH_3	H
12	CH_3	OH
13	H	OH
14	H	H
15	$CH_2CH{=}CHC_6H_5$	H

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Table I. Opioid Agonist Activity of N-Substituted Morphindoles in Smooth Muscle Preparations

compd	MVD % max response[a]	MVD IC_{50}, nM[b]	MVD IC_{50}, nM (δ-enriched)[c]	GPI % max response[a]
2	65 (±5) (5)	–		12 (±4) (3)
3	23 (±2) (3)	–		9 (±4) (3)
4	–	124 (46–497) (3)	121 (82–178) (4)	41 (±4) (4)
5	–	171 (52–565) (4)	76 (24–241) (4)	15 (±6) (3)
6	–	85 (55–132) (4)		20 (±13) (3)
7	34 (±4) (4)	–		9 (±1) (3)
8	–	179 (64–497) (4)	174 (54–561) (3)	1900 (660–5460)[b]
9	19 (±5) (3)	–		2 (±1) (3)
10	41 (±11) (3)	–		23 (±4) (3)
DPDPE		2.8 (2.1–3.9) (9)		

[a] Partial agonist potency (±SE) at 1 μM unless otherwise indicated. [b] The IC_{50} (95% confidence limits) is the concentration of the agonist required for half-maximal response of the preparation. [c] Reference 5.

synthesis of **9** was prepared from noroxymorphone **13** by alkylation with cinnamyl bromide. The phenethyl compounds **5** and **8** were synthesized from indoles **4** and **6**, respectively, by reductive alkylation using phenylacetaldehyde and $NaCNBH_3$. The benzofuran **10** was prepared from O-phenylhydroxylamine and oxymorphone **12** under conditions similar to the Fischer indole synthesis.

Biological Results

Smooth Muscle Preparations. Compounds **4–10** were tested in the electrically stimulated guinea pig ileal longitudinal muscle (GPI) and mouse vas deferens (MVD) preparations as described previously.[4] For full agonists (Table I), 4–6 different concentrations of the test compounds were employed to construct concentration–response curves that were compared to those of standard agonists in the same preparation. When the test compound was found to be a partial agonist or weakly active, it was incubated with the smooth muscle preparation at a single concentration (1 μM) to determine its maximal response. Test compounds that were full agonists in the MVD were tested on a preparation that was enriched in δ receptors in order to evaluate the δ selectivity of the agonist effect.[5]

In the MVD, compounds **4–6** and **8** were found to produce a full agonist effect, while **2, 3, 7, 9,** and **10** behaved as partial agonists with effects that ranged from 19% (**9**) to 65% (**2**) at 1 μM (Table I). The most potent of the full agonists was normorphindole **6** (IC_{50} = 85 nM), whereas the least potent was the phenethyl compound **8** (IC_{50} = 179 nM). When tested on the δ-enriched preparation, only the phenethyl derivative **5** exhibited a significant difference in the IC_{50} which unexpectedly was lower than that in the untreated preparation. In the GPI, all compounds except **8** (IC_{50} = 1900 nM) exhibited partial agonist activity in the range of 2–41% inhibition at 1 μM concentration (Table I).

Antagonist potencies were evaluated by incubating the test compounds with the smooth muscle preparation for 15 min prior to testing with standard agonists (Table II). The ligands were tested at a concentration of 100 nM unless they possessed agonist activity. In such cases the concentrations were adjusted so they would not be greater than their ED_{20} values. Morphine (M), ethylketazocine (EK), and [D-Ala^2,D-Leu^5]enkephalin[6] (DADLE) were employed as μ-, κ-, and δ-selective agonists, respectively. Morphine and EK were employed in the GPI, and DADLE was used in the MVD. With the exception of **7** and **10**, which weakly antagonized DADLE, the compounds were ineffective as antagonists in the MVD and GPI. At a concentration of 3 nM, **5** appeared to potentiate the effect of DADLE (IC_{50} ratio = 0.26).

Table II. Evaluation of Opioid Antagonist Activity of Morphindoles in Smooth Muscle Preparations

compd	IC_{50} Ratio[a] MVD[b] morphine	MVD[b] DADLE	GPI[b] morphine	GPI[b] EK
2	0.9 ± 0.1	1.3 ± 0.3	0.9 ± 0.2	0.9 ± 0.3
3	1.0 ± 0.2	1.2 ± 0.3	0.7 ± 0.1	0.5 ± 0.1
4	0.7 ± 0.1[c]	0.9 ± 0.3[c]	0.7 ± 0.2	0.8 ± 0.2
5	0.9 ± 0.1[d]	0.6 ± 0.2[d]	0.8 ± 0.1	0.8 ± 0.2
6	1.0 ± 0.3[e]	0.8 ± 0.1[e]	0.6 ± 0.2	1.3 ± 0.3
7	0.9 ± 0.3	4.2 ± 2.0	1.0 ± 0.2	0.5 ± 0.4
8	0.9 ± 0.2[d]	0.6 ± 0.4[d]	0.8 ± 0.2	0.9 ± 0.3
9	1.1 ± 0.3	0.8 ± 0.2	0.8 ± 0.2	0.9 ± 0.2
10	0.8 ± 0.2	10.2 ± 2.1	0.5 ± 0.1	0.8 ± 0.1

[a] The ratio represents the IC_{50} of the standard agonists in the presence of ligands 2–10 divided by the control IC_{50} in the same preparation (n = 3). [b] Compounds were tested at 100 nM unless specified. [c] Tested at 20 nM. [d] Tested at 30 nM. [e] Tested at 10 nM.

Table III. Antinociceptive Potencies of Morphindoles

compd	assay[a]	ED_{50} (95% confidence limits), nmol/mouse, icv
1 (NTI)[b]	AS	0.04 (0.01–0.21)
2 (OMI)[b]	AS	0.019 (0.01–0.03)
3	AS	0.010 (0.010–0.011)
4	AS	1.1 (0.7–1.8)
	TF	10.9 (7.5–15.5)
5	AS	0.4 (0.2–0.8)
	TF	2.2 (1.6–3.3)
8	TF	6.3 (4.9–8.3)
DSLET	AS	0.0032 (0.0026–0.0039)
	TF	0.45 (0.24–0.78)
DPDPE	AS	0.9 (0.4–1.7)
	TF	4.0 (3.1–5.0)

[a] The abdominal stretch (AS) or tail flick (TF) assays were employed. [b] Reported in ref 18.

In Vivo Studies. The antinociceptive potencies of selected compounds (**1–5, 8**) were determined in mice using either the tail flick[7] (TF) or abdominal stretch[8] (AS) assays and the icv route (Table III). For compounds (**4, 5,** and **8**) tested using the TF procedure, the range of ED_{50} values

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Table IV. Effect of Selected Opioid Antagonists on the Antinociceptive Potencies of Morphindoles 4 and 5

antagonist pretreatment[a]	assay[b]	ED_{50} ratio[c] (95% confidence limits) 4	5
β-FNA, 20 μmol/kg, 24 h	AS	0.9 (0.4–1.7)	1.3 (0.3–1.0)
	TF	*d*	5.8 (3.5–9.0)
norBNI, 12 μmol/kg, 2 h	AS	1.6 (0.5–5.0)	5.5 (2.0–12.2)
	TF	*d*	21.6 (15.7–31.3)
NTI, 44 μmol/kg, 0.5 h	AS	1.2 (0.8–1.9)	4.0 (0.9–10.5)
	TF	1.6 (1.0–2.8)	3.2 (1.8–5.2)

[a] Administered sc at the dose and pretreatment time indicated. [b] The abdominal stretch (AS) or tail flick (TF) assays were employed in mice. [c] Compounds 4 and 5 were administered icv. The ED_{50} ratio represents the ED_{50} of the test compound in the pretreated animal divided by the control ED_{50}. [d] Data could not be obtained because either higher doses would not dissolve or caused severe toxic signs.

Table V. Evaluation of Opioid Antagonist Potency and Selectivity of Morphindoles 4 and 5 in Mice

compd[b]	ED_{50} ratio (95% confidence limits)[a] DSLET[c]	DPDPE[c]	morphine[d]	U50488H[d]
4	5.4 (2.9–9.8)	2.5 (1.2–6.5)	1.1 (0.5–3.0)	0.7 (0.3–1.4)
5	2.8 (1.0–7.5)	1.4 (0.5–14.1)	0.4 (0.1–1.1)	0.6 (0.4–0.9)

[a] The abdominal stretch (AS) assay was employed. [b] Compounds 4 and 5 were administered icv at doses (0.031 and 0.0078 nmol, respectively) that did not produce agonism and at a time that permitted the peak antagonist response (10 min) to correspond to the time of the peak antinociception of the agonist. [c] The peak times for antinociception after icv administration were 10 (DSLET) and 20 min (DPDPE). [d] The peak times for antinociception were 15 (U50488H) and 30 min (morphine).

was 2–11 nmol/mouse. Where both assays were run on the same compounds (4 and 5), the TF ED_{50} values were 5–10-fold greater than those obtained from the AS assay. Interestingly, NTI 1, OMI 2, and its related N-methyl compound 3 were found to be 10–100-fold more potent than 4 and 5.

The selectivities of the nor (4) and phenethyl (5) compounds were evaluated using the selective antagonists, naltrindole[9] (NTI, δ), norbinaltorphimine[10] (norBNI, κ), and β-funaltrexamine[11] (β-FNA, μ) (Table IV). Using the TF assay, all three antagonists antagonized the antinociceptive effect of 5, with norBNI producing the greatest antagonism. In the AS assay, antinociception was antagonized significantly to about the same degree by NTI and norBNI, with shifts in the range of 4–6. On the other hand, β-FNA did not significantly change the dose ratio. In contrast to 5, the nor compound 4 was not antagonized by any of the selective antagonists.

Both 4 and 5 were evaluated for opioid antagonist activity at doses that did not produce antinociception using the AS assay (Table V). The nor compound 4 was effective in antagonizing the antinociceptive activity of both [D-Ser^2,Leu^5]enkephalin-Thr^6 [12] (DSLET) and [D-Pen^2,D-Pen^5]enkephalin[13] (DPDPE), but not morphine or U50488. The phenethyl 5 antagonized the antinociceptive effect of DSLET only.

Binding Studies. Opioid receptor binding assays using guinea pig brain membranes were carried out on compounds 4–6, 8, and 9 by competition with selective radioligands using a modification of the procedure of Werling et al.[14] Binding to δ, μ, and κ sites was determined by displacement of tritiated DPDPE, [D-Ala^2,MePhe^4,Gly-ol^5]enkephalin[15] (DAMGO), and the benzeneacetamide, U69593.[16]

All of the ligands tested were found to be δ selective (Table VI). They possessed lower affinity for δ sites relative to that of NTI 1 and with the exception of 9 they had greater affinity than the standard δ ligand, (allyl)$_2$Tyr-Aib-Aib-Phe-Leu[17] (ICI 174864).

Discussion

The replacement of a cyclopropylmethyl with methyl, phenethyl, or H at the basic nitrogen of the morphindole pharmacophore afforded compounds that were either weak or inactive as antagonists at ≤100 nM in the smooth muscle preparations (Tables I and II). Moreover, it appears that the phenethyl analogues (5 and 8) and the secondary amines (4 and 6) are full agonists with moderate potency in the MVD preparation. The fact that there was no significant decrease in potency when compounds 4, 5, and 8 were tested on the δ-enriched MVD preparation suggests that they mediate their full agonist effect through interaction with δ receptors. This is supported by the binding data (Table VI) which show the ligands to be δ selective.

The in vivo evaluation of the ligands (4, 5, and 8) that possess full agonist activity in vitro revealed that they share ED_{50} values in the same dose range as the δ-selective agonist, DPDPE (Table III). Interestingly, the *N*-methyl compound 3, which exhibited relatively weak partial agonist activity in the MVD and GPI preparations, was 40–100-fold more potent than the nor (4) and phenethyl (5) analogues. Thus, it appears that there is no correlation between the in vitro and in vivo agonist potencies.

Another notable difference between the in vitro and in vivo pharmacology is the selectivity of the agonist effect, as compounds 4 and 5 are not δ-selective opioid agonists in mice (Table IV). It was found that the agonist effect of 4 was not antagonized significantly by selective opioid antagonists, while that of 5 appeared to be blocked more potently by the κ antagonist, norBNI. While it is not known why the agonist effect of 4 was not antagonized, these studies suggest that the phenethyl compound 5 is a relatively κ-selective agonist. In this regard, it has been reported recently that other morphindoles such as NTI 1

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Table VI. Binding of Morphindoles to Guinea Pig Brain Membranes

compd	K_i,[a] nM			K_i ratio	
	μ	δ	κ	μ/δ	κ/δ
1 (NTI)[b]	3.8 (0.6–22)	0.03 (0.001–0.17)[c]	332 (267–413)[d]	127	11066
2 (OMI)[b]	468 (258–853)	0.7 (0.08–7)[c]	4467 (1954–10190)[d]	669	6381
4	628 (342–1154)	2.8 (1.1–7.5)	>1000	224	>357
5	>1000	11.0 (5–23)	>1000	>90	>90
6	375 (60–2355)	5.3 (1.7–17)	935 (832–1050)	71	176
8	173 (10–2904)	21.9 (6.0–80.9)	2618 (940–7295)	8	120
9	>1000	105.7 (16–698)	>1000	>9	>9
ICI174864[b]	>1000	35 (19–65)	>1000	>29	>29

[a] The geometric mean (95% confidence limits) of K_i values for [^{3}H]DAMGO, [^{3}H]DPDPE, and [^{3}H]U69593, representing μ, δ, and κ receptor binding, respectively. [b] Taken from ref 3. [c] Using [^{3}H]DADLE (in the presence of 1 μM DAMGO) as displaced ligand. [d] Using [^{3}H]ethylketazocine (in presence of 1 μM DAMGO) as displaced ligand.

and OMI 2 also exhibit κ-selective agonist activity at ED_{50} values that are greater than the antagonist dose.[18]

Because the κ agonist effect of 5 occurs at an icv ED_{50} value that is 50-fold higher than the antagonist dose, it is conceivable that the κ-binding component may be small relative to binding to δ sites. This could account for the observed δ-binding selectivity of 5 (Table VI) despite its κ-selective agonist effect in vivo. However, this does not explain the relatively high binding selectivity of NTI 1 and OMI 2 for δ sites in view of the low agonist/antagonist dose ratios for these ligands.[18]

Another possible explanation for the observed κ agonist selectivity in vivo and its discrepancy with the in vitro data is that an initial interaction with δ opioid receptors may trigger the release of dynorphin which could mediate antinociception via κ receptors. This explanation implies that the δ receptors in the MVD may be different from those in the CNS and that the efficacy of the morphindole nucleus is too low to produce a significant direct δ-receptor-mediated antinociceptive effect in the latter. The reported difference between brain (rat, guinea pig) and MVD δ opioid receptors supports this idea.[19,20] In this context it is conceivable that the constellation of message and address elements embedded in the morphindole portion of compounds in this series may tend to stabilize the δ receptor in an antagonist state in vivo. Under such circumstances, changing the N substituent from cyclopropylmethyl to phenethyl or methyl would not convert an antagonist into an agonist. It appears that the indolic benzene moiety, which is coplanar with the C ring of the morphinan nucleus, may contribute to such stabilization, as we have recently discovered that an out-of-plane phenyl group (a δ address mimic) affords compounds that are δ-selective agonists in vivo.[21]

It is conceivable that the binding site on the δ receptor which recognizes NTI and related morphindoles is not identical to the peptidergic site that is involved in signal transduction, even though such ligands compete efficiently with δ-selective radioligands. In such a case, the morphindole could bind to a site that is allosterically coupled to the radioligand peptidergic binding site involved in signal transduction. This would also explain the inability to transform a δ antagonist (e.g., naltrindole) into an agonist by replacement of its N substituent in the present study.

In any case, the clear lack of correlation between the mouse vas deferens preparation and the antinociceptive assays in the present study underscores a basic difference between the δ receptor system in the smooth muscle and in the mouse brain. Inasmuch as there are at least two δ opioid receptor subtypes (δ_1 and δ_2) in the brains of mice, rats, and guinea pigs, as suggested from pharmacologic[20,22-25] and binding data,[20,26,27] it appears that caution sould be exercised in employing the MVD to screen for δ opioid receptor ligands.

Experimental Section

Melting points were determined in open capillary tubes with a Thomas-Hoover melting point apparatus and are uncorrected. Microanalyses were performed by M-H-W Laboratories, Phoenix, AZ, and were within ±0.4% of calculated values. Mass spectra were obtained on an AEIMS-30, Finnigan 4000, or VG 7070E-HF spectrometer. NMR spectra were recorded on an IBM Bruker AC-200, IBM Bruker AC-300, or Nicolet 300 MHz spectrometer using DMSO-d_6 as solvent. TLC data were obtained on 0.25-mm silica gel sheets plastic-backed (Polygram Sil G/UV, Macherey-Nagel) using chloroform/methanol/ammonium hydroxide (C/M/A) as the development solvent. Column chromatography was performed on silica gel (E. Merck, 230–400 mesh) using dry column technique and C/M/N as the development and elution solvent either isocratically or as a methanol component gradient. HPLC separations were performed on a semipreparative silica column (Analtech, 10 μm, 10 mm i.d. × 25 cm) using a Beckman 110A pump and detection at 254 nm. All reagents and solvents employed were reagent or HPLC grade and were used without purification. Hydromorphone, oxymorphone, and noroxymorphone were supplied by Mallinckrodt.

6,7-Dehydro-4,5α-epoxy-3,14-dihydroxy-6,7-2′,3′-indolomorphinan Hydrochloride (4·HCl). A mixture of noroxymorphone base 13 (1.0 g, 3.5 mmol) suspended in MeOH (20 mL)

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and phenylhydrazine hydrochloride (0.56 g, 3.9 mmol) dissolved in methanolic 1 M HCl was refluxed in a closed atmosphere for 16 h. The solvent was removed in vacuo, and the residue was digested in warm H_2O (30 mL) to afford a precipitate, 1.4 g (98%), of 4·HCl: mp >250 °C; R_f 0.22 (C/M/A, 75:25:1, v/v); ^{1}H NMR (DMSO-d_6) δ 9.24 (s, 1 H, ArOH), 7.38, 7.34 (dd, 2 H, H-4′ and H-7′), 7.12 (t, 1 H, J = 7.4 Hz, H-6′), 6.97 (t, 1 H, J = 7.5 Hz, H-5′), 6.66, 6.60 (dd, 2 H, H-1 and H-2), 5.66 (s, 1 H, H-5); EIMS m/z 360 (M^+). Anal. ($C_{22}H_{20}N_2O_3$·HCl·H_2O) C, H, N.

17-(2-Phenylethyl)-6,7-dehydro-4,5α-epoxy-3,14-dihydroxy-6,7-2′,3′-indolomorphinan Hydrochloride (5·HCl). Noroxymorphindole hydrochloride (4) (500 mg, 1.3 mmol) was suspended in KOAc–HOAc/MeOH buffer (20 mL, 1 M, pH 6) stored over 3-Å molecular sieves. Phenylacetaldehyde (315 mg, 2.6 mmol), $NaCNBH_3$ (411 mg, 6.6 mmol), and 3-Å sieves were added, and the mixture was stirred at room temperature for 16 h. The mixture was filtered, and the filtrate was basified with 20 mL of 1 N HCl. The methanol portion of the solvent was removed in vacuo, the residual aqueous solution was made basic with excess ammonia and extracted with 4 volumes (80 mL) of EtOAc, and the combined EtOAc extracts were dried and the solvent removed. The crude product was taken up in $CHCl_3$ and chromatographed through a column of silica gel using an eluant gradient [$CHCl_3$/MeOH (0–5%)/NH_4OH (1%)] to give **5** as a solid base (471 mg, 77%), which was converted in EtOAc/MeOH to the hydrochloride and phosphate salts with 1 N HCl or 1 N H_3PO_4 in MeOH. **5**·HCl: mp >250 °C; R_f 0.53 (C/M/A, 95:5:1, v/v); ^{1}H NMR (DMSO-d_6) δ 9.30 (s, 1 H, ArOH), 7.36 (m, 7 H, phenyl H, H-4′ and H-7′), 7.10 (t, 1 H, J = 7.1 Hz, H-6′), 6.97 (t, 1 H, J = 7.6 Hz, H-5′), 6.67, 6.60 (dd, 2 H, J = 8.1 Hz, H-1 and H-2), 6.43 (s, 1 H, C-14 OH), 5.70 (s, 1 H, H-5); FABMS m/z 465 (M^+ + 1). Anal. ($C_{30}H_{28}N_2O_3$·HCl·0.5H_2O) C, H, N.

6,7-Dehydro-4,5α-epoxy-3-hydroxy-6,7,2′,3′-indolomorphinan Hydrochloride (6·HCl). A mixture of 14·HCl[28] (200 mg, 0.65 mmol) and phenylhydrazine hydrochloride (113 mg, 0.78 mmol) in HOAc (5 mL) was stirred at 90 °C for 16 h and then filtered. The product then was washed sequentially with cold HOAc and acetone to yield 240 mg (98%) of 6·HCl: mp >250 °C; R_f 0.37 (C/M/A, 80:20:1, v/v); ^{1}H NMR (DMSO-d_6) δ 9.17 (s, 1 H, ArOH), 7.34 (m, 2 H, H-4′ and H-7′), 7.09 (t, 1 H, J = 7.5 Hz, H-6′), 6.95 (t, 1 H, J = 7.5 Hz, H-5′), 6.64, 6.59 (dd, 2 H, J = 8.1 Hz, H-1 and H-2), 5.76 (s, 1 H, H-5); EIMS m/z 344 (M^+). Anal. ($C_{22}H_{20}N_2O_2$·HCl·1.25H_2O) C, H, N.

17-Methyl-6,7-dehydro-4,5α-epoxy-3-hydroxy-6,7-2′,3′-indolomorphinan Hydrochloride (7·HCl). A mixture of 11 (321 mg, 1 mmol) and phenylhydrazine hydrochloride (216 mg, 1.5 mmol) was dissolved in EtOH (10 mL) and HCl (0.5 mL). The resulting solution was refluxed under nitrogen for 3 h; at the end of this period the mixture was cooled and solid precipitated. The precipitate was collected by filtration, washed with Et_2O (50 mL), dried, and crystallized from MeOH/EtOH (1:2) to afford 186 mg (52%) of pure 7·HCl salt: mp 270 °C dec; R_f 0.4 (C/M/A, 9:1:0.1, v/v); ^{1}H NMR (DMSO-d_6) δ 7.37 (dd, 1 H, H-4′ and H-7′), 7.02 (t, 1 H, H-5′), 6.90 (t, 1 H, H-6′), 6.68 (dd, 2 H, H-1 and H2), 5.8 (s, 1 H, H-5), 3.36 (s, 1 H, NCH_3); FABMS m/z 359 (M^+ + 1). Anal. ($C_{23}H_{22}N_2O_2$·HCl·H_2O) C, H, N.

17-(2-Phenylethyl)-6,7-dehydro-4,5α-epoxy-3-hydroxy-6,7-2′,3′-indolomorphinan Phosphate (8·H_3PO_4). Compound 6·HCl (200 mg, 0.53 mmol) was suspended in 6 mL of KOAc/HOAc in MeOH buffer (1 M, pH 6) stored over 3-Å molecular sieves. To the mixture was added phenylacetaldehyde (126 mg, 1.05 mmol), $NaCNBH_3$ (160 mg, 2.65 mmol), and 3-Å molecular sieves. The mixture was stirred at ambient temperature overnight and filtered, and the filtrate was evaporated to dryness. The oily residue was partitioned between NH_4OH/H_2O and $CHCl_3$, and the combined $CHCl_3$ extracts were washed once with brine and dried ($NaSO_4$). The dried extract was concentrated and chromatographed on a silica gel column using C/M/N (95:5:1 v/v), isocratically. Fractions containing purified product were combined, concentrated, and acidified with 1 N H_3PO_4 in EtOH to give 26 mg (10%) of acid phosphate salt: mp >250 °C; R_f 0.55 (C/M/N, 95:5:1, v/v); ^{1}H NMR (DMSO-d_6) δ 9.13 (s, 1 H, ArOH), 7.32 (m, 7 H, phenyl H, H-4′ and H-7′), 7.10 (t, 1 H, J = 7.5 Hz, H-6′), 6.96 (t, 1 H, J = 7.5 Hz, H-5′), 6.62, 6.58 (dd, 2 H, H-1 and H-2), 5.65 (s, 1 H, H-5); CIMS m/z 448 (M^+). Anal. ($C_{30}H_{28}N_2O_2$·2H_3PO_4·H_2O) C, H, N.

17-Cinnamyl-6,7-dehydro-4,5α-epoxy-3-hydroxy-6,7-2′,3′-indolomorphinan Hydrochloride (9·HCl). A suspension of 6·HCl (65 mg, 0.21 mmol) in DMF (2 mL) was mixed with cinnamyl bromide (45 mg, 0.23 mmol) and stirred at 90 °C overnight in the presence of Na_2CO_3. The reaction mixture was filtered, and the filtrate was evaporated to dryness. The residue was purified on silica gel using C/M/N with a gradient of 0–5% MeOH to give **15** which was converted to the hydrochloride salt and crystallized from Et_2O/MeOH [72 mg; 87%; EIMS 387 (M^+)].

A mixture of 15·HCl (20 mg, 0.047 mmol) and phenylhydrazine hydrochloride (14 mg, 0.094 mmol) in 1 mL of HOAc was stirred at 100 °C for 3 h. The HOAc was removed in vacuo, and the residue was chromatographed using C/M (0–5%)/N. A subsequent purification by HPLC (C/M/N, 97:3:1 v/v isocratically at 2 mL/min) furnished **9** base (10 mg, 46%; t_R = 29 min), which was converted to 9·HCl: mp > 250 °C; R_f 0.51 (C/M/N, 95:5:1, v/v); ^{1}H NMR (DMSO-d_6) δ 9.91 (s, 1 H, ArOH), 7.46 (m, 7 H, H-4′, H-7′, and phenyl H), 7.30 (m, 2 H), 7.09 (t, 1 H, J = 7.6 Hz, H-6′), 6.91 (m, 2 H, H-5′ and vinyl), 6.7–6.4 (m, 3 H, H-1 and H-2, and vinyl), 5.82 (s, 1 H, H-5); EIMS m/z 460 (M^+).

17-Methyl-6,7-dehydro-4,5α-epoxy-3,14-dihydroxy-6,7-2′,3′-benzofuranomorphinan Methanesulfonate (10·CH_3SO_3H). A solution of 12 (150 mg, 0.50 mmol), *O*-phenylhydroxylamine hydrochloride[29] (80 mg, 0.55 mmol), and methanesulfonic acid (0.1 mL, 1.04 mmol) in EtOH (10 mL) was refluxed for 24 h, and product was then collected by filtration and washed with EtOH (10 mL). The solid was dried and crystallized from EtOH/MeOH (2:1) to afford 126 mg (33%) of methanesulfonate salt: mp 230 °C dec; R_f 0.60 (C/M/N, 9:1:0.1, v/v); ^{1}H NMR (DMSO-d_6) δ 7.61 (δ, 1 H, H-7′), 7.45 (d, 1 H, H-4′), 7.2 (t, 1 H, H-5′), 7.40 (t, 1 H, H-6′), 6.52 (dd, 2 H, H-1 and H-2), 5.65 (s, 1 H, H-5), 2.92 (s, 3 H, N-CH_3); FABMS m/z 376 (M^+ + 1). Anal. ($C_{23}H_{21}NO_4$·CH_3SO_3H·H_2O) C, H, N.

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