Quantum Chemical Studies of N-Substituent Variation in the Oxymorphone Series of Opiate Narcotics

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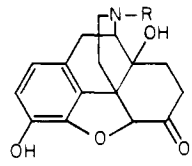
Quantum chemical calculations were performed on six N-derivatives of oxymorphone including *N*-methyl- (oxymorphone), *N*-allyl- (naloxone), *N*-dimethylallyl- (nalmexone), *N*-methylcyclopropyl- (naltrexone), *N*-methylcyclobutyl- (nalbuphone), and *N*-phenethylnoroxymorphone using the PCILO method. The object of the study was to identify conformational features of the N-substituents which might be responsible for the intrinsic observed pharmacological properties of opiate agonism and antagonism. Both axial and equatorial N-substituent conformers were considered, as well as possible interactions of the C_{14}-OH group with such substituents. Variations of agonist/antagonist potency ratios within this series could not be explained by differing relative energies of equatorial and axial conformations or by varying rates of interconversion between the two. Direct effects of the C_{14}-OH group on conformations of N-substituents also could not account for their relative agonist/antagonist potencies. Consistent with a previous hypothesis, the observed potencies and binding data could be explained most consistently by the availability of several low-energy equatorial conformations of N-substituents and their interactions with the C_{14}-OH group through a common anionic receptor site.

The oxymorphone series (**1a–f**) of potent opiate narcotic



1a, R = CH_3 (oxymorphone)
b, R = $CH_2CH{=}CH_2$ (naloxone)
c, R = $CH_2CH{=}C(CH_3)_2$ (nalmexone)
d, R = CH_2-c-C_3H_5 (naltrexone)
e, R = CH_2-c-C_4H_7 (nalbuphone)
f, R = CH_2CH_2Ph

agonists and antagonists is a good example of the complex role the N-substituent plays in controlling pharmacological behavior ranging from potent agonism (**1a** and **1f**) to mixed agonism–antagonism (**1c** and **1e**) to pure potent antagonism (**1b** and **1d**).[1-5] Although metabolism may have some effect on the relative agonist/antagonist potencies of some of these compounds (for example, the weak and incomplete analgesia of naltrexone has been attributed to a metabolite, β-naltrexol[6]), the data in Table I establish a fairly consistent trend. While there are three structural differences between the morphine and oxymorphone parent compounds, it is the C_{14}-OH group that appears to most drastically alter the morphine-like pharmacological profile for varying N-substituents.[7-9] In pure agonists, the C_{14}-OH appears to slightly decrease intrinsic in vitro potency[9] while increasing in vivo potency,[8] presumably by a favorable transport effect. In mixed agonist–antagonists the C_{14}-OH drastically diminishes agonism while increasing antagonism.[7] While these general observations apply to the few compounds studied, more experimental study is required to firmly establish these trends.

To explore the role of the C_{14}-OH group and various N-substituents in mediating agonism and antagonism, a systematic quantum chemical study has been made of **1a–f**. The purpose of this study was to identify structural properties which might explain the observed variations in relative agonist/antagonist potency. Calculaltions were performed to examine the conformational behavior of both axial and equatorial N-substituents, to estimate minimum energy differences between axial and equatorial conformations in the protonated and neutral species, and to estimate inversion barriers in the neutral molecule in which such inversions must occur.

Several authors have recently speculated on the roles of N-substituents and the axial C_{14}-OH group in conferring agonist or antagonist activity on the morphine-like fused-ring structures.[10-15] It has been suggested that axial C_{14}-OH groups might act by direct steric interference with certain equatorial N-substituent conformations.[10] It has also been suggested that agonist and antagonist activities are associated with axial and equatorial N-substituents, respectively.[15] Although there is evidence for rapid interconversion between axial and equatorial N-substituent orientations of unhindered piperidines in solution,[16] nitrogen inversion is unverified in the highly rigid three,[17] four, and five fused-ring opiates, and its correlation with relative agonist and antagonist potencies is unproven. In all crystal structures of neutral and protonated three, four, and five fused-ring opiates reported to date, N-substituents have been found to be equatorial.

We recently proposed that the dual agonist–antagonist behavior of nalorphine was due to the existence of two low-energy "types" of equatorial *N*-allyl conformations at the receptor. Differences in activity between agonists and antagonists could not be attributed to electronic effects.[11-13] Such differences in drug conformation, as well as in receptor conformation, could account for the differences between agonist and antagonist binding associated with the observed[3] Na^+ effect. We have also recently reported calculations which suggest that the C_{14}-OH group and various N-substituents cannot interact directly but might interact indirectly through a common anionic receptor site such as RSO_4^- or $R_1R_2PO_4^-$. In this study we use both of these hypotheses to help understand the variation of agonist/antagonist potency in the series of oxymorphone derivates selected.

The conformational calculations reported here were done on the protonated form, thought to be the active form,[18] and no solvent effects were included. Solvent effects on N-substituent conformations are not expected to be substantial for the lipophilic substituents studied. Solvent interactions were not found to affect inversion energies in unhindered piperidine compounds.[16] The possibility of induced conformational change at the receptor site is included in our study by considering many low-energy conformers, rather than a single global minimum, as receptor site candidates.

Experimental Section

The conformational calculations reported here were done on the isolated, protonated form of the opiates, thought to be the active form.[18] All calculations were performed using the program

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Table I. Agonist and Antagonist Potency Data of Methyl-, Allyl-, Dimethylallyl-, Methylcyclopropyl-, Methylcyclobutyl-, and Phenethylnoroxymorphones

	In vitro			In vivo[b]			
	Binding			Agonism		Antagonism	
Compd	Pig ileum, Ke,[a] nM	Rat brain homog,[e] EC_{50}, nM	Agonism, pig ileum, ID_{50},[a] nM	Rat writhing, ED_{50}, mg/kg	Mouse writhing, ED_{50}, mg/kg	Rat,[c] ED_{50}, mg/kg	Mouse,[d] ED_{50}, mg/kg	Agonism, mouse hot plate,[f] ED_{50}
1a	11.2 ± 2.5	1.0	12.1 ± 2.4	0.020 (0.013–0.030)	0.046 (0.027–0.078)			0.17
1b	1.22 ± 0.03	1.5	∞	>100	>100	0.051 (0.030–0.080)	0.14 (0.09–0.23)	
1c	43 ± 10		400	0.74 (0.57–0.96)	2.0 (1.1–3.6)	1.4 (0.74–2.7)	5.6 (4.2–7.5)	10.7
1d	0.38 ± 0.07	0.5	>850	0.14 (0.064–0.31)	>100	0.023 (0.014–0.037)	0.047 (0.031–0.071)	
1e	3 ± 0.45		60 ± 17	0.06 (0.038–0.096)	0.39 (0.19–0.82)	0.19 (0.12–0.30)	0.57 (0.36–0.92)	
1f								0.014
Morphine	87.5 ± 18.1	3.0	68.2 ± 15.0	0.26 (0.18–0.38)	0.8 (0.64–0.97)			1.17
Nalorphine	4.47 ± 0.59	1.5	24.3 ± 1.3	1.0[g]		0.90 (0.58–1.39)	1.45 (0.95–2.15)	

[a] See ref 1; for nalmexone see ref 4. [b] See ref 2. [c] Antagonist potency measured as counteraction of oxymorphone Straub tail effect in mouse (sc). [d] Antagonist potency measured as counteraction of oxymorphone narcosis in rat (sc). [e] See ref 3. [f] See ref 5. [g] Data from R. I. Taber, *Adv. Biochem. Psychopharmacol.*, 8, 191 (1974) (morphine = 0.5 in same study).

PCILO[19] which has been used to study a variety of biological systems.[11,20,21] The geometry of the oxymorphone fused-ring structure was taken from a recent crystal structure of naloxone[22] and is comparable to other recent oxymorphone derivative crystal structures.[23,24] Hydrogens were placed by combining standard bond lengths[25] with bond angles (∠CCH,∠HCH) equalized due to a lack of refinement in the crystal structure. Geometries of the N-substituents except around the nitrogen atom were also standardized[25] with verification for this assumption from crystal structure data for methyl,[23,26] allyl,[22] methylcyclopropyl,[27] and methylcyclobutyl[28] groups.

Conformational calculations on equatorial N-substituents were performed with a bonding geometry about the nitrogen atom taken from the crystal structure of naloxone.[22] For the minimum energy conformer, the bond angles $\angle C_9NH$,$\angle C_9NC_\alpha$ and the torsion angles $\tau(C_{10}C_9NC_\alpha)$ and $\tau(C_{10}C_9NH)$ were reoptimized to give a more accurate energy for the best equatorial conformer. For the axial N-substituent, the geometry about the nitrogen atom was initially minimized for a single conformation of each N-substituent. Energy–conformation tables of the axial N-substituents were obtained using these optimized nitrogen geometries. The global minimum obtained by this procedure for each compound was then reoptimized with respect to the nitrogen geometry to obtain a more accurate minimum energy for the axial conformation.

Optimized global minima were used to obtain energy differences between the best axial and equatorial conformations in the protonated form. Using the same global minima, the nitrogen bond angles were reoptimized for both axial and equatorial conformers to obtain energy differences in the neutral species. Estimates were also obtained for the neutral molecule inversion barriers between axial and equatorial conformers. The transition state was assumed to be a roughly planar conformation about the nitrogen, and a geometry optimized energy for such a state, i.e., when $\tau(C_{10}C_9NC_\alpha)$ was at a maximum and other angles at their minimum energy value, was calculated. Energies for the planar, transition states were calculated with sp^2 hybridization of the nitrogen atom, an option available in the PCILO program, while axial and equatorial N-substituent orientations were calculated using sp^3 hybridization appropriate for tetrahedral nitrogen orientations. In order to calibrate such estimates, calculations for the inversion barrier were also made for ammonia and aniline for which experimental values of barrier heights are known.

All torsion angles used in this work are defined by the convention: τ_i(ABCD) = clockwise rotation of atom A into D while looking along the B–C axis from B to C. τ_1 for all compounds defines the twist angle C_9–N–C_α–C_β while τ_2 defines N–C_α–C_β–C_γ. N-Substituent geometries are defined as pairs of values of τ_1 and τ_2 in parentheses, i.e., for τ_1 = 300° and τ_2 = 60° the designation is (300°,60°). For the methycyclopropyl derivative, τ_2 = 0° when $\tau_{H(C_\beta)}$ = 214°, while for the methylcyclobutyl derivative τ_2 = 0° when $\tau_{H(C_\beta)}$ = 130°.

N-Substituent rotations for all compounds were performed with the OH group at its position in the crystal structure of naloxone (τ_{OH} = C_9–C_{14}–O–H = 270°). For the axial orientation in oxymorphone, the OH group was rotated in 30° steps with the NCH_3 both and staggered and eclipsed. The minimum energy position was not substantially different from the crystal structure. Conformations with the hydroxyl proton pointing toward the N-substituent were high-energy forms. Thus, for all other N-substituent rotations, the unhindered crystal structure orientation τ_{OH} = 270° was used. Coupling of the equatorial N-substituent position to the OH twist angle was explored in naloxone by rotating the OH group with the allyl group fixed at positions alternately favoring (300°,0°) or not favoring (180°,180°) C_{14}-OH allyl interaction. All rotations reported here were performed in 30° intervals unless otherwise noted.

For the nalmexone derivative, the two C_γ-methyl group conformations were determined by energy minimization at three equatorial local minima of the allyl substituent in naloxone, namely, (180°,240°), (90°,240°), and (300°,120°). These minimizations all gave the same results. The methyl group cis to C_α has hydrogen atoms staggered with respect to C_β and the *trans*-methyl group has one hydrogen cis to C_β.

For the phenethyl derivative full rotations of the three rotation axes were not feasible. Instead, a rotation in 30° steps was performed for τ_3 (with τ_1 = τ_2 = 180°) where τ_3 was defined by

Table II. Axial–Equatorial Energy Differences in the Protonated and Neutral Species and Neutral Molecule Inversion Energies

Energy	Compd							
	Morphine	Nalorphine	Oxymorphone	Naloxone	Nalmexone	Naltrexone	Nalbuphone	*N*-Phenylnoroxymorphone
ΔE^a (protonated)	0.4	1.2	1.8	1.9	1.0	2.0	1.9	2.2
ΔE^b (neutral)	2.9	2.1	5.1	5.4	4.5	4.0	3.5	3.9
ΔE^{*c} (inversion)	14.9	12.1	15.0	13.2	13.4	11.8	12.3	15.2

[a] Energy differences in kcal/mol between axial and equatorial minimum energy protonated conformations using optimized N bond angles as shown in Table IIIB (supplementary material) ($\Delta E = E_{ax} - E_{eq}$). [b] Energy differences in kcal/mol between axial and equatorial minimum energy neutral molecule conformations using optimized bond angles as shown in Table IIIA (supplementary material) ($\Delta E = E_{ax} - E_{eq}$). [c] Energy differences in kcal/mol between planar and equatorial minimum energy neutral molecule conformations using optimized geometries as shown in Table IIIA (supplementary material). These energies correspond to gas-phase inversion energies of activation.

the positions of atoms C_α–C_β–C_γ–C_{Ph}. Three local minima were found at $\tau_3 = 0°$, 60°, and 300° and were each used in full rotations of τ_1 and τ_2 for the equatorial conformation. For the axial rotations, τ_3 was held at 0°.

Results

Neutral molecule inversion barriers (ΔE^*) are shown in Table II, together with energy differences between the best axial and equatorial N-substituent conformations of both neutral and protonated opiates. These energy differences provide an indication of the relative proportion of each conformer present.

For all compounds studied, the equatorial conformer is the lowest energy form. This result is consistent with crystal structure data as are the optimized values of equilibrium bond angles about the nitrogen. Morphine and nalorphine have comparable axial–equatorial N-substituent energy differences, as do oxymorphone and naloxone. Some effect of the C_{14}-OH can be seen by larger energy differences in the oxymorphone series over those in morphine and nalorphine but the effect on the pure agonist and pure antagonist N-substituents is comparable.

In the protonated form of all species, the equatorial–axial energy differences are smaller, reflecting the simultaneous presence of N ligands at both positions.

The inversion barriers correspond to the calculated differences in energy between the lowest energy (equatorial) and planar structures. They are relatively independent of N-substituent and the presence or absence of C_{14}-OH.

Calculated barrier heights for ammonia and aniline were 12 and 7 kcal/mol, respectively, compared to known values of 6 and 2. Thus, while the PCILO method overestimates these quantities, their relative values are reasonably reliable.

Table III gives the optimized geometries of axial, equatorial, and transition states for all the opiates studied in their neutral and protonated form. Tables IV–XVI give the complete energy–torsion angle variations obtained for axial and equatorial N-substituents for all compounds studied. (For Tables III–XVI, see paragraph at end of paper regarding supplementary material.) Figure 1 (A–K) shows low-energy equatorial N-substituent conformations for all compounds studied.

For oxymorphone, staggered and eclipsed equatorial and axial conformers were investigated. In the protonated equatorial form, the staggered conformation [Figure 1 (A)] was favored by 1.7 kcal/mol while in the axial conformer it was favored by 1.2 kcal/mol.

The rotational behavior of the C_{14}-OH group with an equatorial NCH_3 substituent has been discussed elsewhere[29] and is identical with that of equatorial naloxone, shown in Figure 2 (a). The rotational behavior of

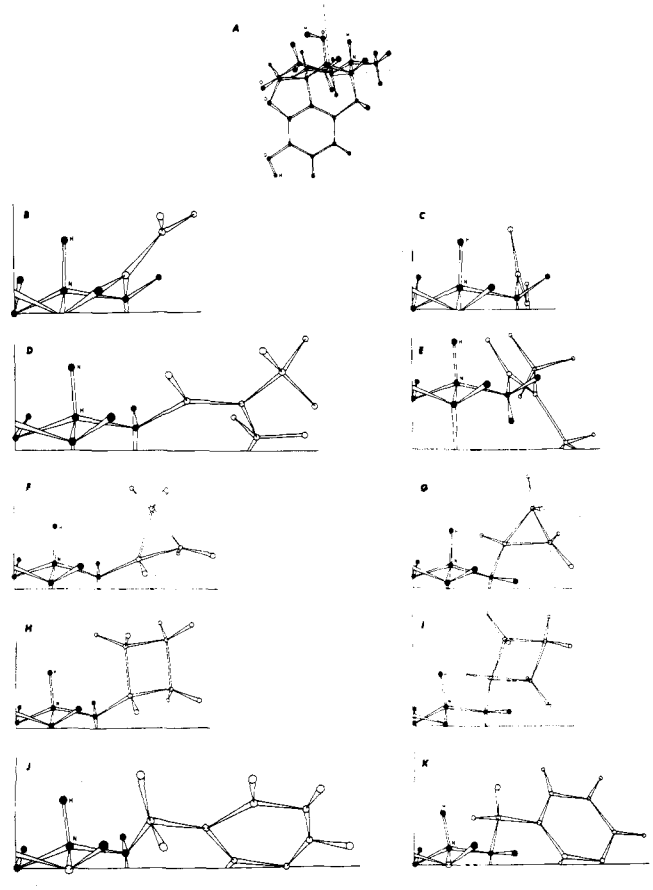

Figure 1. Two types of low-energy conformers for equatorial N-substituents of oxymphone derivatives: (A) oxymorphone (staggered); (B) naloxone (300°,240°); (C) naloxone (180°,240°); (D) nalmexone (300°,180°); (E) nalmexone (180°,210°); (F) naltrexone (300°,210°); (G) naltrexone (210°,90°); (H) nalbuphone (300°,300°); (I) nalbuphone (210°,180°); (J) phenethyl (300°,180°,0°); (K) phenethyl (210°,180°,0°).

the C_{14}-OH group with an axial NCH_3 substituent is shown in Figure 2 (b). All conformers in which the C_{14}-OH points toward the NCH_3 are high-energy forms. The low-energy region of the C_{14}-OH group is very similar to that for the equatorial N-substituent. With the C_{14}-OH in such positions, low-energy conformers of equatorial and axial N-substituents are not affected by the presence of the C_{14}-OH group.

Figure 2 (a) also shows no direct interaction between the C_{14}-OH and the allyl group in its equatorial conformation. This curve was reproduced to within 0.5 kcal/mol for two different conformers (τ_1,τ_2) of the allyl group chosen because they represented possible interacting (300°,0°) and noninteracting (180°,240°) conformations with the C_{14}-OH. The minimum distance possible between the hydrogen of

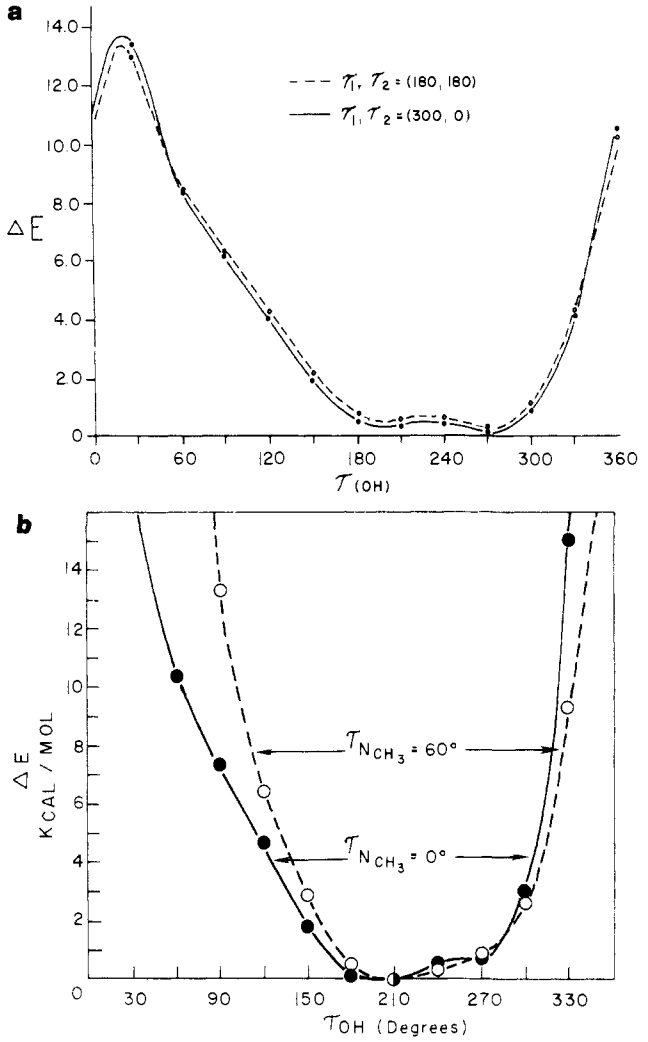

Figure 2. (a) Rotational behavior of the C_{14}-OH group in naloxone with the equatorial allyl group at (300°,0°) and (180°,180°). (b) Rotational behavior of the C_{14}-OH group in axial oxymorphone with NCH_3 protons alternately staggered and eclipsed.

the C_{14}-OH group and an H atom of the allyl group is 1.75 Å. In the region of low-energy conformations of the C_{14}-OH (ΔE < 3 kcal/mol), the curves differed by <0.2 kcal/mol.

The rotational behavior of the allyl group in equatorial [Table IV and Figure 1 (B and C)] and axial (Table XII) naloxone closely resembles that of equatorial and axial (Table XI) nalorphine in low-energy conformations. Significant energy differences (>1 kcal/mol) are found only for high-energy axial conformers with the allyl group oriented near the C_{14}-OH. An equatorial local minima [Figure 1 (B)] at (300°,240°) is consistent with naloxone crystal structures (309°,262°).[22]

The dimethylallyl derivative in its axial (Table XIII) and equatorial [Table V and Figure 1 (D and E)] orientations is conformationally similar to naloxone. However, the added methyl groups increase the rotational energy barriers and decrease the range of low-energy conformers, thus decreasing the flexibility of the substituent. Although this increased rigidity is overestimated due to lack of optimization of the methyl hydrogen conformations at each calculated point, the basic hindering effect of the methyl groups should be real.

Naltrexone shows somewhat increased conformational rigidity over naloxone in axial (Table XIV) and equatorial [Table VI and Figure 1 (F and G)] orientations of the N-substituent. Equatorial conformations which are local minima resemble local minima of naloxone in that one edge of the cyclopropyl ring is superimposable on the allyl double bond in its low-energy conformations. A local minima at (300°,90°) is consistent with crystal structures of cyclazocine (300°,97°) in both protonated and neutral forms.[27]

The rotational behavior of equatorial nalbuphone (Table VII) shows a local minima (210°,180°) consistent with a recent crystal structure of nalbuphine (174°,180°).[28] Although less flexible than any other N-substituent in this series, it shares its lowest energy conformations [(300°,300°) and (210°,180°)] with naltrexone as shown in Figure 1 (H and I). In its axial conformation (Table XIV), it is constrained primarily to one conformation (150°,300°) with two others [(60°,180°) and (120°,180°)] 2.8 and 2.6 kcal/mol above the global minimum.

The results of the equatorial phenethyl rotations are shown in Tables VII–IX with three different conformers of the phenyl ring (τ_3). Minimum energy conformers are of two different types, shown in Figure 1 (J and K): a flexible extended chain conformation with τ_1 = 210° or 300° and τ_2 = 180° (in all three tables) and another with the phenyl group directly above the N–H bond [τ_1,τ_2,τ_3 = (210°,60°,60°) (330°,300°,120°)]. The second type of conformer and the small barrier between τ_1 = 210° and τ_1 = 300° in the extended chain conformers were not observed in our previous study of *N*-phenethylmorphine due to the coarser rotational grid used in that work.[11]

In the axial phenethyloxymorphone calculations, the N-substituent favors an approximately extended chain conformation (150°,180°,0°) with (60°,180°,0°) and (150°,300°,0°) local minima 1.4 and 2.4 kcal/mol higher in energy, a rotational behavior somewhat similar to nalbuphone.

Discussion

The results summarized in Table II do not support the hypothesis that axial N-substituents are required for agonist activity while equatorial substituents confer antagonist activity. No correlation exists between agonist/antagonist potency variation and calculated axial–equatorial energy differences in either the base or protonated form of the opiates studied. For example, the equatorial conformer is the lowest energy conformer for all compounds studied. Moreover, axial conformations are equally accessible in naloxone and oxymorphone, indicating an equally hindering effect of the C_{14}-OH group on agonists and antagonists. In general, there is no systematic increase in axial conformational energies as agonist potency decreases.

The inversion barriers calculated for the neutral molecules are insensitive to N-substitution and, hence, show no correlation to agonist/antagonist potencies. Comparison of the calculated inversion energies (Table II) with those calculated for ammonia and aniline indicates that the inversion process can occur in these fused ring opiates at about the same rate as in ammonia and that the axial conformer is accessible, particularly in the protonated form of the isolated molecule. There is no evidence, however, that this inversion or the presence of axial conformers is related to drug action at the receptor.

The rotational behavior of the equatorial *N*-allyl group in naloxone, with the C_{14}-OH in its minimum energy conformation, is identical within computational accuracy to that calculated for the allyl group in nalorphine. As shown in Figure 1 (B and C) the allyl group in naloxone retains the two types of low-energy conformers found in nalorphine (τ_1 = 180° and 300°, τ_2 = 120° and 240°) which we previously associated with its dual agonist and an-

tagonist behavior.[11] In low-energy regions of axial nalorphine and naloxone the same statements can be made. Thus, the total abolition of agonist activity in naloxone cannot be explained by differing accessible allyl conformations from those in nalorphine nor by direct interaction of the axial C_{14}-OH with low-energy *N*-allyl conformers.

In previous work we have suggested that agonist/antagonist behavior could be regulated by different low-energy equatorial N-substituent conformers[11] and that in the oxymorphone series such conformers interact indirectly with the C_{14}-OH group through the receptor.[14] Complex formation could occur by simultaneous interaction of the C_{14}-OH, cationic nitrogen, and certain N-substituents with a model anionic receptor site (sulfate or phosphate). The ternary complex formed is stable only for a small range of N-substituent conformations. These could correspond to effective antagonist–receptor interaction and account for the relatively strict structural requirements for pure antagonism. The role of the C_{14}-OH group in diminishing agonism then would be to cause preferential selection of only one type of low-energy conformer for receptor complex formation from among the several hypothesized "agonist" and "antagonist" N-substituent conformations.

If this hypothesis is correct, any substitution that detracts from the strict geometric requirements found for (C_{14}-OH)–(anion)–(*N*-allyl) antagonist complexing should enhance the agonist/antagonist potency ratio. Two such alterations, the C_{14}-OCH_3 derivative of naloxone and the C_{15}-OH analogue, should restore agonism. Removal of the C_{14}-OH group altogether to make *N*-allylhydromorphone would also produce a mixed agonist/antagonist. The ability of the axial C_{14}-OH group alone to reduce intrinsic agonism has been recently demonstrated by the decreased agonism of naltrexone relative to its hydromorphone analogues in the guinea pig ileum.

As shown in Figure 1 (D and E) the equatorial *N*-dimethylallyl substitution in nalmexone has low-energy conformers similar to naloxone. However, these minima are steeper and energy barriers between them higher than for the N-substituent of naloxone. This loss of flexibility could detract from its ability to bind to the receptor simultaneously with C_{14}-OH in a low-energy "antagonist" conformation. Consistently, the data in Table I show that nalmexone has a much higher agonist/antagonist potency ratio and lower binding affinity to the guinea pig ileum than naloxone. In contrast, the axial–equatorial energy differences between naloxone and nalmexone are comparable and provide no particular clue as to why they are pharmacologically so different.

Naltrexone shows very similar N-substituent conformational behavior to naloxone for both axial and equatorial conformations [Figure 1 (F and G)]. The axial conformers of naltrexone are equally accessible as those of oxymorphone, again ruling out their role as regulators of agonist activity. The results firmly suggest that the equatorial methylcyclopropyl group confers pure or nearly pure antagonism on the molecule by conformationally occupying the same space as naloxone's allyl group while retaining some of its π-bond character. Thus, one would predict that substitution on cyclopropyl ring carbons would alter the locations of conformational local minimas and reduce the antagonism of naltrexone by reducing overlap with the local minima of the allyl group.

Comparing nalbuphone [Figure 1 (H and I)] to naltrexone's conformational behavior, nalbuphone is more rigid, decreasing its ability to form complexes with the receptor simultaneously with the C_{14}-OH group while in its antagonist conformation. In addition to this steric restriction, a diminshed π character would further decrease its ability to form charge-transfer complexes with a receptor anion. Thus, the agonist conformer would exist to a greater extent than in naloxone and naltrexone and overall binding would diminish as is observed.

In equatorial phenethyloxymorphone, it is suggested that the conformers with the phenyl ring directly over the quaternary amine group are ruled out by steric constraints at the receptor site. Such conformers would directly interfere with cationic nitrogen–anionic receptor interaction presumed to be an important part of agonist activity. Our data suggest that the *N*-phenethyl substituent may then bind to the receptor in an approximately extended chain conformation [Figure 1 (J and K)] allowing the phenyl group adequate flexibility to interact with an additional binding site somewhat removed from the anionic site of the receptor. Binding to this site could enhance the agonist potency as seen in animal studies[5] while preserving the "oxymorphone-like" drug–receptor interaction.

Conclusion

Axial–equatorial energy differences of N-substituents in the oxymorphone series show no correlation with opiate agonist/antagonist potency ratios. No direct interaction of the C_{14}-OH group with low-energy axial or equatorial conformations of the N-substituents occurs and, hence, cannot explain the effect of C_{14} substitution on pharmacological activity. A rather speculative but consistent explanation of the potency and binding data suggests itself if it is assumed that the cationic nitrogen of agonists and antagonists interacts with a large anion such as sulfate or phosphate to form a drug–receptor complex. The nature of this complex could then be affected by both C_{14} and N substitution. The binding of the axial C_{14}-OH group to the receptor could preferentially select the antagonist form of the N-substituent to form the most stable ternary receptor complex. With this suggested hypothesis, allyl- and methylcyclopropyl N-substituents would be prototypes for optimum formation of such ternary complexes since they have a comparatively high degree of flexibility and π character. Nalmexone and nalbuphone are less flexible and have less π character and thus would form weaker ternary complexes reducing the effect of the C_{14}-OH group (i.e., creating a less pure antagonist). The lack of antagonist activity of methyl and phenethyl N-substituents could then be explained by their lack of interaction with the receptor anionic site. The *N*-phenethyl substituent could, however, interact with a more distant binding site, enhancing its binding and agonist potency. Further characterization of the interaction of different N-substituents with a model anionic site is now in progress.

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Supplementary Material Available: Tables III–XVI (15 pages). Ordering information is given on any current masthead page.

References and Notes

(1) H. W. Kosterlitz and A. A. Waterfield, *Annu. Rev. Pharmacol.*, **15**, 29 (1975).
(2) H. Blumberg and H. B. Dayton, *Adv. Biochem. Psychopharmacol.*, **8**, 33 (1974).
(3) C. B. Pert and S. H. Snyder, *Mol. Pharmacol.*, **10**, 868 (1974).

(4) H. W. Kosterlitz, J. A. H. Lord, and A. J. Watt, "Agonist and Antagonist Actions of Narcotic Analgesic Drugs", Macmillan, London, 1972.
(5) G. A. Deneau, J. E. Villarreal, and M. H. Seevers in "Problems of Drug Dependence", National Academy of Sciences, Washington, D.C., 1973.
(6) K. Verebey, J. Volavka, S. J. Mule, and R. B. Resnick, *Clin. Pharmacol. Ther.*, **20**, 315 (1975).
(7) M. Gordon, A. W. Pircio, F. S. Caruso, and I. J. Pachter, paper presented to the Committee on Problems of Drug Dependence, Mexico City, N.M., 1974.
(8) E. Kutter, A. Herz, H. J. Teschemacher, and R. Hess, *J. Med. Chem.*, **13**, 801 (1970).
(9) J. Knoll, *Neuropharmacology*, **14**, 921 (1975).
(10) J. R. Smythies, private communication.
(11) G. H. Loew and D. S. Berkowitz, *J. Med. Chem.*, **18**, 656 (1975).
(12) J. J. Kaufman, H. Popkie, and W. Koski, *J. Am. Chem. Soc.*, **98**, 1342 (1976).
(13) J. J. Kaufman and E. Kerman, *Int. J. Quantum Chem.*, 8, 289 (1974).
(14) D. S. Berkowitz and G. H. Loew in "Opiates and Endogenous Opioids", Elsevier/North Holland, Amsterdam, 1976.
(15) A. P. Feinberg, I. Creese, and S. H. Snyder, *Proc. Natl. Acad. Sci. U.S.A.*, **73**, 4215 (1976).
(16) R. A. Y. Jones, A. R. Katritsky, A. C. Richards, and R. J. Wyatt, *J. Chem. Soc. B*, 122 (1970).
(17) D. C. Palmer and M. J. Strauss, *Chem. Rev.*, **77**, 1 (1977).
(18) A. F. Casy in "Modern Pharmacology", Vol. 1, R. M. Featherstone, Ed., Marcel Dekker, New York, N.Y., 1973, p 1.
(19) S. Diner, J. P. Malrieu, F. Jordan, and M. Gilliert, *Theor. Chim. Acta*, **15**, 100 (1969).
(20) G. H. Loew and J. R. Jester, *J. Med. Chem.*, 18, 1051 (1975).
(21) G. H. Loew, D. S. Berkowitz, and R. Newth, *J. Med. Chem.*, **19**, 863 (1976).
(22) I. L. Karle, *Acta Crystallogr., Sect. B*, **30**, 1682 (1974).
(23) R. J. Sime, M. Dobler, and R. L. Sime, private communication.
(24) R. L. Sime, R. Forehand, and R. J. Sime, *Acta Crystallogr., Sect. B*, **31**, 2236 (1975).
(25) J. Pople and D. Beveridge, "Approximate Molecular Orbital Theory", McGraw-Hill, New York, N.Y., 1970.
(26) L. Gylbert, *Acta Crystallogr., Sect. B*, **29**, 1630 (1973).
(27) I. L. Karle, R. D. Gilardi, A. V. Fratini, and J. Karle, *Acta Crystallogr., Sect. B*, **25**, 1469 (1969).
(28) R. J. Sime, M. Dobler, and R. L. Sime, *Acta Crystallogr., Sect. B*, **32**, 809 (1976).
(29) G. H. Loew, H. Weinstein, and D. Berkowitz in "Environmental Effects on Molecular Structure and Properties", B. Pullman, Ed., D. Reidel Publishing Co., Dordrecht, Holland, 1976.

Notes

A Novel Synthesis and Biological Activity of Several 5-Halo-5′-amino Analogues of Deoxyribopyrimidine Nucleosides

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A novel synthetic procedure has been developed for the large-scale synthesis of 5-chloro-, 5-bromo-, and 5-iodo-5′-amino-2′,5′-dideoxyuridine (**4c**–e) as well as of two new analogues, 5-iodo-5′-amino-2′,5′-dideoxycytidine and 5-fluoro-5′-amino-2′,5′-dideoxyuridine (**4a** and **4b**), in good yield. The starting materials, 5-halo-2′-deoxyuridine and 5-halo-2′-deoxycytidine, are readily available and the method is straightforward. This report describes the synthesis and the biologial activities of these compounds.

Several 5-halo analogues of 2′-deoxyuridine and 2′-deoxycytidine have excellent biological activity as either antineoplastic or antiviral agents. These include 5-fluoro-2′-deoxyuridine, 5-iodo-2′-deoxyuridine, 5-iodo-2′-deoxycytidine, etc.[1-3] In an attempt to modify their toxicity with retention of antiviral activity, the 5′-hydroxyl moiety of several nucleoside analogues has been replaced with an amino group.[4,5] Of these, 5-iodo-5′-amino-2′,5′-dideoxyuridine (AIU, AIdUrd) has retained the antiviral activity of the parent compound, 5-iodo-2′-deoxyuridine (IdUrd), albeit with a lesser potency,[6,7] but remarkably has none of the toxicities associated with the 5′-hydroxyl analogues.[6-8] Although the original method of preparation of AIdUrd[4] provided a sufficient amount of compound for our in vitro studies[6,9,10] and topical use in initial animal experiments,[7,10] there was a need for an improved method of synthesis to provide the large amounts required for systemic administration in our proposed animal test systems. Starting material (IdUrd), 300 g, yields 10–15% product of AIdUrd. A novel synthetic procedure was developed which is applicable for the large-scale synthesis of 5-chloro-, 5-bromo-, and 5-iodo-5′-amino-2′,5′-dideoxyuridine (**4c**–e) as well as of two new analogues, 5-iodo-5′-amino-2′,5′-dideoxycytidine and 5-fluoro-5′-amino-2′,5′-dideoxyuridine (**4a** and **4b**). The present report describes the synthesis and the biological activities of these compounds.

Chemistry. The synthesis of a variety of 5-halo-5′-substituted deoxyribopyrimidine nucleoside analogues is outlined in Scheme I. Tosylation of 5-halo-2′-deoxyribonucleosides **1a**–**e** with *p*-toluenesulfonyl chloride in dry pyridine at 3 °C gave the corresponding 5-halo-5′-*o*-*p*-tolylsulfonyl derivatives **2a**–**e**. Compounds **2a**–**e** reacted with lithium azide in *N*,*N*-dimethylformamide at 70–75 °C for 2 h to afford the 5-halo-5′-azido analogues **3a**–**e**.[4,5,11] Treatment of **3a**–**e** with triphenylphosphine[12] in pyridine at room temperature, followed by hydrolysis with concentrated ammonium hydroxide, yielded a series of novel 5-halo-5′-amino nucleoside analogues **4a**–**e**. The physical properties, yields of the last step conversion, and elemental analyses are listed in Table I. Compounds **4d** and **4e** have been synthesized previously by treatment of the 5-mercuriacetate of 5′-amino-2′,5′-dideoxyuridine with bromine and iodine, respectively.[4,13] Although this method

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