J|A|C|S
ARTICLES
Published on Web 04/05/2002

Cation−π Interactions: An Energy Decomposition Analysis and Its Implication in δ-Opioid Receptor−Ligand Binding

Yirong Mo,*,† Govindan Subramanian,*,‡ Jiali Gao,§ and David M. Ferguson||

Contribution from the Department of Chemistry, Department of Medicinal Chemistry, and Minnesota Supercomputer Institute, University of Minnesota, Minneapolis, Minnesota 55455, Xencor Inc., 111 West Lemon Avenue, Monrovia, California 91016, and Transtech Pharma, 4170, Mendenhall Oaks Parkway, Suite 110, High Point, North Carolina 27265

Received October 31, 2001

Abstract: The nature and strength of the cation−π interaction in protein−ligand binding are modeled by considering a series of nonbonded complexes involving N-substituted piperidines and substituted monocylic aromatics that mimic the δ-opioid receptor−ligand binding. High-level ab initio quantum mechanical calculations confirm the importance of such cation−π interactions, whose intermolecular interaction energy ranges from −6 to −12 kcal/mol. A better understanding of the electrostatics, polarization, and other intermolecular interactions is obtained by appropriately decomposing the total interaction energy into their individual components. The energy decomposition analysis is also useful for parametrizing existing molecular mechanics force fields that could then account for energetic contributions arising out of cation−π interactions in biomolecules. The present results further provide a framework for interpreting experimental results from point mutation reported for the δ-opioid receptor.

Introduction

The [μ-, κ-, and δ]-opioid receptors belong to the superfamily of G-protein coupled receptors, consisting of a seven α-helical transmembrane domain interlinked through the extra- and intracellular loops.[1] In the absence of crystal structures for either the receptor or the ligand-bound complex, the understanding of receptor−ligand interactions relies heavily on information derived from site-directed mutagenesis experiments.[2] To this end, we constructed a three-dimensional structural model of the transmembrane (TM) domain of the opioid receptors to help to gain insights into the molecular nature governing receptor−ligand interactions.[3] Through this study, a salt bridge between Asp128 in TM-III and the cationic amine group of the ligand (substituted piperidines) has been identified as a key anchoring point in the receptor−ligand complex.[4]

However, a recent pharmacological study revealed that single-point mutations of Asp128 in TM-III of the δ-opioid receptor to either aspargine or alanine do not affect the binding of δ-selective opioid agonists such as DPDPE, deltorphin II, and opiate antagonists such as naltrindole.[5] This observation is in contrast to the traditional hypothesis that cationic amines interact with biogenic amine receptor families of GPCR's through the anionic receptor site.[6] The absence of such an anchor point in the Asp128Asn and Asp128Ala mutants also prompts an interesting question on the origin of the δ-receptor−ligand interactions responsible for the ligand association. An examination of the neighboring receptor residues in the binding pocket indicates that Tyr129 may participate in cation−π interactions[7] with the ligand and, consequently, stabilize the complex. Indirect experimental support of this hypothesis stems from the fact that the binding affinities of nonselective antagonists such as bremazocine and naloxone at the δ-opioid receptor are impaired by over 90-fold when Tyr129 is mutated to alanine (Tyr129Ala).[8] In contrast, Tyr129Phe mutation results only in modest reduction in binding affinities of the above two ligands. Similarly, the transmembrane helical residues such as Phe222 (V), Trp274 (VI), and Tyr308 (VII) lining the binding pocket have also been shown to affect the binding of several δ-opioid ligands. These experimental evidences clearly point to the specific roles these

* To whom correspondence should be addressed. Phone: 336-841-0300. Fax: 336-841-0310. E-Mail: G.S., gsubramanian@ttpharma.com; Y.M., ymo@xencor.com.
† Xencor Inc.
‡ Transtech Pharma.
§ Department of Chemistry and Minnesota Supercomputer Institute.
|| Department of Medicinal Chemistry and Minnesota Supercomputer Institute.

(1) Dhawan, B. N.; Cesselin, F.; Raghubir, R.; Reisine, T.; Bradley, P. B.; Portoghese, P. S.; Hammon, M. *Pharmacol. Rev.* **1996**, *48*, 567.
(2) (a) Edvardsen, Ø.; Kristiansen, K. *7 Transmembrane Journal* **1997**, *6*, 1. (b) Horn, F.; Weare, J.; Beukers, M. W.; Hörsch, S.; Bairoch, A.; Chen, W.; Edvardsen, Ø.; Campagne, F.; Vriend, G. *Nucleic Acids Res.* **1998**, *26*, 275.
(3) (a) Metzger, T. G.; Paterlini, M. G.; Portoghese, P. S.; Ferguson, D. M. *Neurochem. Res.* **1996**, *21*, 1287. (b) Subramanian, G.; Paterlini, M. G.; Portoghese, P. S.; Ferguson, D. M. *J. Med. Chem.* **2000**, *43*, 381.
(4) (a) Heerding, J.; Raynor, K.; Kong, H.; Yu, L.; Reisine, T. *Regul. Pept.* **1994**, *54*, 119. (b) Kong, H.; Raynor, K.; Reisine, T. *Regul. Pept.* **1994**, *54*, 155. (c) Surratt, C. K.; Johnson, P. S.; Moriwaki, A.; Seidleck, B. K.; Blaschak, C. J.; Wang, J. B.; Yuhasz, S.; Uhl, G. R. *Regul. Pept.* **1994**, *54*, 289. (d) Surratt, C. K.; Johnson, P. S.; Moriwaki, A.; Seidleck, B. K.; Blaschak, C. J.; Wang, J. B.; Uhl, G. R. *J. Biol. Chem.* **1994**, *269*, 20548.
(5) (a) Befort, K.; Tabbara, L.; Bausch, S.; Chavkin, C.; Evans, C.; Kieffer, B. *Mol. Pharmacol.* **1996**, *49*, 216. (b) Befort, K.; Zilliox, C.; Filliol, D.; Yue, S.; Kieffer, B. L. *J. Biol. Chem.* **1999**, *274*, 18574.
(6) Bikker, J. A.; Trumpp-Kallmeyer, S.; Humblet, C. *J. Med. Chem.* **1998**, *41*, 2911 and references therein.
(7) Dougherty, D. A.; Stauffer, D. A. *Science* **1990**, *250*, 1558.
(8) Befort, K.; Tabbara, L.; Kling, D.; Maigret, B.; Kieffer, B. L. *J. Biol. Chem.* **1996**, *271*, 10161.

10.1021/ja0174433 CCC: $22.00

aromatic residues play in δ-opioid receptor toward ligand recognition.

Although it has not been specifically suggested for opioid receptor−ligand binding, cation−π interactions are known to play a key role in numerous biological recognition processes,[9] and cation−π interactions have been identified to be responsible for quaternary ammonium ion binding in enzymes.[10] Here, we examine the importance of cation−π interactions in model systems that could mimic the appropriate binding environments in the δ-opioid receptor−ligand complex.[11]

There have been several computational studies of cation−π interactions, ranging from the prototype benzene-ammonia[12] to more complex examples.[9c] Gao et al. reported the free energy of association between tetramethylammonium cation and benzene in water using a combined quantum mechanical and Monte Carlo simulation method.[13] Dougherty and co-workers emphasize the importance of electrostatic interactions between the cation and the quadrupole moments of an aromatic ring,[14] whereas additional terms, including induced dipole, polarization, dispersion, and charge transfer, must be included in quantitative models.[15] Kollman and co-workers showed that molecular mechanics models with explicit polarization terms can reproduce cation−π binding energies, whereas pairwise potentials generally perform poorly in comparison with high-level ab initio results.[16] Using a perturbation approach, Cubero et al. confirmed that the contribution from polarization effects in cation−aromatic interactions is 70% of the total electrostatic energy at the optimum Na^+-benzene contact.[17] These studies demonstrate that it is necessary to gain a fundamental understanding of cation−π interactions to develop empirical models for the prediction of receptor−ligand binding constants.

In the present study, ab initio quantum chemical calculations are performed on the prototype complexes that mimic the tyrosine-ligand environments in the opioid receptor−ligand complex (Figure 1). An energy decomposition analysis is performed subsequently to arrive at the relative contributions of various energy terms (electrostatics, polarization, etc.) toward the total intermolecular interaction energy.[18] The findings from this work provide additional insight on cation−π interactions in general, and the quantitative results will be useful for the development of more efficient force fields directed toward the accurate modeling of receptor−ligand interactions.

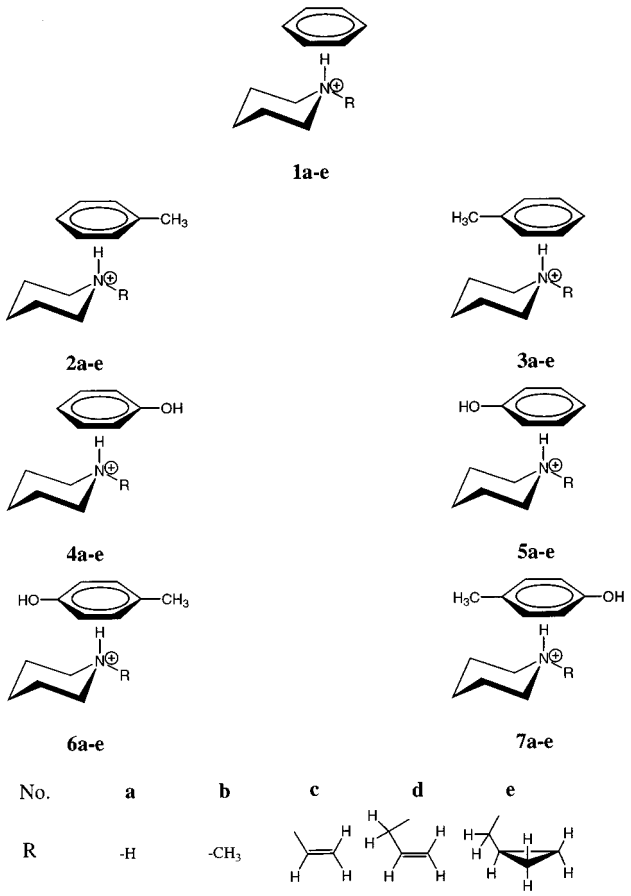

Figure 1. Schematic representation of the nonbonded complexes with various R groups.

Methods

The binding energy, ΔE_b, between two monomers **A** and **B** is defined as the difference of the energy of the complex and the sum of the energies of individual species.

$$\Delta E_b = E(\mathbf{AB}) - E(\mathbf{A}^o) - E(\mathbf{B}^o) + E_{BSSE} \quad (1)$$

where E_{BSSE} is the correction for the basis-set superposition error, $E(\mathbf{AB})$ is the energy of the complex **AB**, and $E(\mathbf{A}^o)$ and $E(\mathbf{B}^o)$ are energies of monomer $\mathbf{A}^o$ and $\mathbf{B}^o$, respectively. Because the geometry of the individual monomer is different in the complex than in isolation, geometrical deformation energy can be defined to emphasize the fact of the structural alteration due to intermolecular interactions. Thus, the process of complex formation may be divided into two steps: (1) a structural deformation from the equilibrium geometry of the isolated monomer ($\mathbf{A}^o$ and $\mathbf{B}^o$), which is specified by superscript "o", to that when the monomer (**A** and **B**) is in the complex, and (2) the interaction energy to bring these two "deformed" species into the complex configuration. Thus,

$$\Delta E_b = \Delta E_{def} + \Delta E_{int} \quad (2)$$

where $\Delta E_{def} = E(\mathbf{A}) + E(\mathbf{B}) - E(\mathbf{A}^o) - E(\mathbf{B}^o)$, and ΔE_{int} is defined in eq 3 below.

Recently, we developed a block-localized wave function (BLW) method, which allows the localization of charge density in specific regions of a molecule in molecular orbital calculations.[18] An important feature of the BLW method is the definition of an intermediate wave

(9) (a) Burley, S. K.; Petsko, G. A. *FEBS Lett.* **1986**, *203*, 139. (b) Scrutton, N. S.; Raine, A. R. C. *Biochem. J.* **1996**, *319*, 1. (c) Ma, J. C.; Dougherty, D. A. *Chem. Rev.* **1997**, *97*, 1303. (d) Gallivan, J. P.; Dougherty, D. A. *Proc. Natl. Acad. Sci. U.S.A.* **1999**, *96*, 9459.
(10) (a) Sussman, J. L.; Harel, M.; Frolow, F.; Oefner, C.; Goldman, A.; Toker, L.; Silman, I. *Science* **1991**, *253*, 872. (b) Stauffer, D. A.; Karlin, A. *Biochemistry* **1994**, *33*, 6840. (c) Lin, Z.; Johnson, M. E. *FEBS Lett.* **1995**, *370*, 1.
(11) (a) Balboni, G.; Guerrini, R.; Salvadori, S.; Tomatis, R.; Bryant, S. D.; Bianchi, C.; Attila, M.; Lazarus, L. H. *Biol. Chem.* **1997**, *378*, 19. (b) Bryant, S. D.; Balboni, G.; Guerrini, R.; Salvadori, S.; Tomatis, R.; Lazarus, L. H. *Biol. Chem.* **1997**, *378*, 107.
(12) Rodham, D. A.; Suzuki, S.; Suenram, R. D.; Lovas, F. J.; Dasgupta, S.; Goddard, W. A., III; Blake, G. A. *Nature* **1993**, *362*, 735.
(13) Gao, J.; Chou, L. W.; Auerbach, A. *Biophys. J.* **1993**, *1*, 43.
(14) (a) Stauffer, D. A.; Barrans, J. R. E.; Dougherty, D. A. *Angew. Chem., Int. Ed. Engl.* **1990**, *29*, 915. (b) Dougherty, D. A. *Science* **1996**, *271*, 163.
(15) (a) Heginbotham, L.; MacKinnon, R. *Neuron* **1992**, *8*, 483. (b) Kumpf, R. A.; Dougherty, D. A. *Science* **1993**, *261*, 1708.
(16) (a) Caldwell, J. W.; Kollman, P. A. *J. Am. Chem. Soc.* **1995**, *117*, 4177. (b) Eriksson, M. A. L.; Morgantini, P. Y.; Kollman, P. A. *J. Phys. Chem.* **1999**, *103*, 4474.
(17) (a) Cubero, E.; Luque, F. J.; Orozco, M. *Proc. Natl. Acad. Sci. U.S.A.* **1998**, *95*, 5976. (b) Cubero, E.; Orozco, M.; Luque, F. J. *J. Phys. Chem. A* **1999**, *103*, 315.
(18) (a) Mo, Y.; Peyerimhoff, S. D. *J. Chem. Phys.* **1998**, *109*, 1687. (b) Mo, Y.; Zhang, Y.; Gao, J. *J. Am. Chem. Soc.* **1999**, *121*, 5737. (c) Mo, Y.; Gao, J.; Peyerimhoff, S. D. *J. Chem. Phys.* **2000**, *112*, 5530.

function, where charge transfer between interacting partners is "turned off".[18] This intermediate wave function is defined by expansion of the molecular orbitals over basis orbitals located only within each Lewis resonance structure block. Consequently, the intermolecular interaction can be evaluated in a series of successive steps, and the energy variation can be decomposed into various intuitive terms. For a dimer composed of two monomers **A** and **B**, the intermolecular interaction (ΔE_{int}) is defined as

$$\Delta E_{int} = E(\Psi_{AB}) - E(\Psi_A^o) - E(\Psi_B^o) + E_{BSSE} \quad (3)$$

with the explicit consideration of the basis-set superposition error (BSSE). Starting from the individual monomers, whose wave functions are Ψ_A^o and Ψ_B^o, respectively, we construct the initial block-localized wave function

$$\Psi_{AB}^o = \hat{A}(\Psi_A^o \Psi_B^o) \quad (4)$$

The energy variation compared to the total energy of isolated monomers is in fact the sum of electrostatic and exchange interactions, which can be further decomposed as reported earlier.[18c] However, the exchange interaction (Pauli repulsion) is a quantum mechanical effect and does not occur in force fields that are used to model biomolecules and condensed systems. Thus, the energy difference between the initial block-localized wave function and monomers is considered as the electrostatic energy term (ΔE_{es})

$$\Delta E_{es} = E(\Psi_{AB}^o) - E(\Psi_A^o) - E(\Psi_B^o) \quad (5)$$

In Ψ_{AB}^o, the electron densities of the monomers are the same as when they are separated. However, the approach of the two monomers will inevitably perturb each other's individual electron densities, and this effect is ascribed to polarization. This can be measured by the energy variation between the optimal BLW wave function

$$\Psi_{AB}^{BLW} = \hat{A}(\Psi_A \Psi_B) \quad (6)$$

and the initial BLW Ψ_{AB}^o

$$\Delta E_{pol} = E(\Psi_{AB}^{BLW}) - E(\Psi_{AB}^o) \quad (7)$$

In the above equation, the optimization of Ψ_{AB}^{BLW}, where the orbitals in Ψ_A or Ψ_B are orthogonal, and the orbitals between Ψ_A and Ψ_B are nonorthogonal, comprises the major task of the BLW method. In fact, Ψ_{AB}^{BLW} corresponds to the adiabatic state where electron transfer between **A** and **B** is deactivated. Because routine Hartree−Fock (HF) wave function is constructed from the molecular orbitals that are delocalized over the whole system, it corresponds to an adiabatic state. The energy difference (ΔE_{ct}) between BLW and HF wave function (+BSSE, which is estimated by the counterpoise correction method)[19] is attributed to the charge-transfer effect

$$\Delta E_{ct} = E(\Psi_{AB}^{HF}) - E(\Psi_{AB}^{BLW}) + E_{BSSE} \quad (8)$$

Consequently, the overall intermolecular interaction at the HF level is decomposed as a sum of all the above energy terms

$$\Delta E_{int} = \Delta E_{es} + \Delta E_{pol} + \Delta E_{ct} \quad (9)$$

Numerous energy decomposition schemes have been proposed in the literature for probing and explaining the role of noncovalent intermolecular forces in receptor−ligand interactions, enzyme−substrate binding, antigen−antibody recognition, and notably the hydrogen-bonding mechanism in nonbonded bimolecular complexes.[18,20−23] Compared with these existing schemes, the BLW energy decomposition (BLW-ED) method reported in this paper can uniquely define and optimize the wave function for the localized structure (Lewis structure) where charge transfer between monomers is turned off. This provides a rational ground to examine the polarization and charge-transfer contributions. Our previous comparison[18] of the BLW-ED method with Morokuma's scheme[20] also demonstrated the stability of our method with respect to the basis sets.

To investigate the model opioid receptor−ligand complex, noncovalently bound bimolecular complexes involving various substituted piperidine (to mimic the environments of opioid ligands such as naloxone, naltrexone, morphine, etc.) and aromatic rings are considered (Figure 1). In particular, the former includes protonated amino group with hydrogen, methyl, vinyl, allyl, and cyclopropylmethyl as N-substitutions to the piperidine ring, while the aromatic ring is comprised of benzene, toluene, phenol, and cresol resulting in 20 unique interacting pairs. However, for some pairs there are other possible interacting sites, which result in multiple low energy states for a pair. Thus, a total of 35 stable states for these 20 unique pairs were located using the HF/6-31G(d) basis sets implemented in the Gaussian 98[24] program. The energy decomposition calculation using the BLW approach was performed for each of these identified nonbonded complexes. The BLW-ED code was developed at the University of Minnesota and ported into GAMESS[25] to take advantage of its direct self-consistent field capability. Although the self-consistent field approach slightly underestimates the interaction energy as compared to the experimental values,[26] the present limitation of the BLW approach in handling correlated wave functions restricts the choice to the use of HF methodology only.

Results and Discussion

The binding and interaction energies along with the individual energy components obtained through the BLW-ED listed in Table 1 provide guidance for the unique trends observed for the 35 investigated complexes. Clearly, the cation−π interactions are significant for most of the systems considered, with predicted binding energies ranging from −6 to −12 kcal/mol. This is close to the −12.5 kcal/mol interaction energy obtained for the methylammonium···benzene complex at the HF/6-31+G* level by Dougherty et al.[27] In fact, these authors have further shown that the cation−π interaction is more stabilizing than the salt-bridge interaction energy in aqueous solution.[27]

(19) Boys, S. F.; Bernardi, F. *Mol. Phys.* **1970**, *19*, 553.

(20) (a) Kitaura, K.; Morokuma, K. *Int. J. Quantum Chem.* **1976**, *10*, 325. (b) Morokuma, K.; Kitaura, K. In *Chemical Applications of Atomic and Molecular Electrostatic Potentials*; Politer, P., Truhlar, D. G., Eds.; Plenum: New York, 1981.

(21) (a) Stevens, W. J.; Fink, W. H. *Chem. Phys. Lett.* **1987**, *139*, 15. (b) Stone, A. J. *The Theory of Intermolecular Forces*; Oxford University Press: Oxford, 1996.

(22) (a) Reed, A. E.; Curtiss, L. A.; Weinhold, F. *Chem. Rev.* **1988**, *88*, 99. (b) Glendening, E. D.; Streitwieser, A. *J. Chem. Phys.* **1994**, *100*, 290. Glendening, E. D. *J. Am. Chem. Soc.* **1996**, *118*, 2473. (c) Schenter, G. K.; Glendening, E. D. *J. Phys. Chem.* **1996**, *100*, 17152.

(23) van der Vaart, A.; Merz, K. M., Jr. *J. Phys. Chem.* **1999**, *A103*, 3321.

(24) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Zakrzewski, V. G.; Montgomery, J. A., Jr.; Stratmann, R. E.; Burant, J. C.; Dapprich, S.; Millam, J. M.; Daniels, A. D.; Kudin, K. N.; Strain, M. C.; Farkas, O.; Tomasi, J.; Barone, V.; Cossi, M.; Cammi, R.; Mennucci, B.; Pomelli, C.; Adamo, C.; Clifford, S.; Ochterski, J.; Petersson, G. A.; Ayala, P. Y.; Cui, Q.; Morokuma, K.; Malick, D. K.; Rabuck, A. D.; Raghavachari, K.; Foresman, J. B.; Cioslowski, J.; Ortiz, J. V.; Stefanov, B. B.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Gomperts, R.; Martin, R. L.; Fox, D. J.; Keith, T.; Al-Laham, M. A.; Peng, C. Y.; Nanayakkara, A.; Gonzalez, C.; Challacombe, M.; Gill, P. M. W.; Johnson, B. G.; Chen, W.; Wong, M. W.; Andres, J. L.; Head-Gordon, M.; Replogle, E. S.; Pople, J. A. *Gaussian 98*; Gaussian, Inc.: Pittsburgh, PA, 1998.

(25) Schmidt, M. W.; Baldridge, K. K.; Boatz, J. A.; Elbert, S. T.; Gordon, M. S.; Jensen, J. J.; Koseki, S.; Matsunaga, N.; Nguyen, K. A.; Su, S.; Windus, T. L.; Dupuis, M.; Montgomery, J. A. *J. Comput. Chem.* **1993**, *13*, 1347.

(26) Deakyne, C. A.; Meot-Ner (Mautner), M. *J. Am. Chem. Soc.* **1985**, *107*, 474.

Table 1. Energy Decomposition Results (kcal/mol) for Intermolecular Interactions[a]

complex	E_{BSSE}	ΔE_{el}	ΔE_{pol}	ΔE_{ct}	ΔE_{int}	ΔE_{def}	ΔE_b	type	complex	E_{BSSE}	ΔE_{el}	ΔE_{pol}	ΔE_{ct}	ΔE_{int}	ΔE_{def}	ΔE_b	type
1a	1.21	−4.86	−3.12	−2.59	−10.57	0.33	−9.24	II	**4d**	1.68	−6.26	−3.92	−1.70	−11.88	1.79	−10.09	III
1b	1.68	−4.25	−2.41	−1.41	−8.07	0.26	−7.81	I	**4e**	1.42	−7.05	−3.98	−1.63	−12.66	1.92	−10.74	III
1c	1.30	−3.17	−2.03	−1.60	−6.80	0.32	−6.48	I	**5a**	1.14	−5.54	−3.32	−2.35	−11.21	0.67	−10.54	II
1d	1.76	−2.52	−1.94	−1.56	−6.02	0.46	−5.56	I	**5b**	1.73	−5.15	−2.74	−1.67	−9.56	0.82	−8.74	I
1e	1.64	−3.86	−2.05	−1.28	−7.19	0.25	−6.94	I	**5c**	1.50	−7.07	−4.20	−1.56	−12.83	1.64	−11.19	III
2a	1.26	−5.02	−3.60	−2.64	−11.26	0.39	−10.87	II	**5d**	1.70	−2.64	−2.07	−1.63	−6.34	0.70	−5.64	I
2b	1.66	−4.35	−2.83	−1.58	−8.76	0.31	−8.45	I	**5e**	1.70	−4.83	−2.40	−1.47	−8.70	0.80	−7.90	I
2c	1.32	−3.29	−2.44	−1.76	−7.49	0.37	−7.12	I	**6a**	1.19	−5.83	−3.74	−2.26	−11.83	0.82	−11.01	II
2d	1.78	−2.68	−2.22	−1.61	−6.51	0.51	−6.00	I	**6b**	1.75	−5.60	−3.12	−1.65	−10.37	1.07	−9.30	I
2e	1.61	−3.92	−2.42	−1.46	−7.80	0.29	−7.51	I	**6c**	1.54	−7.78	−4.51	−1.64	−13.93	1.93	−12.00	III
3a	1.22	−4.99	−3.58	−2.68	−11.25	0.39	−10.86	II	**6d**	1.50	−7.07	−3.96	−1.49	−12.52	1.66	−10.86	III
3b	1.65	−4.47	−2.72	−1.36	−8.55	0.33	−8.22	I	**6e**	1.40	−8.38	−4.30	−1.59	−14.27	2.76	−11.51	III
3c	1.53	−3.15	−2.31	−1.73	−7.19	0.39	−6.80	I	**7a**	1.25	−6.15	−3.69	−2.47	−12.31	0.94	−11.37	II
3d	1.79	−2.71	−2.26	−1.67	−6.64	0.50	−6.14	I	**7b**	1.41	−8.51	−4.70	−1.85	−15.06	2.49	−12.57	III
3e	1.62	−4.02	−2.33	−1.27	−7.62	0.30	−7.32	I	**7c**	1.79	−7.82	−4.53	−1.60	−13.95	1.70	−12.25	III
4a	1.25	−5.84	−3.30	−2.44	−11.58	0.81	−10.77	II	**7d**	1.70	−6.95	−4.24	−1.79	−12.98	2.11	−10.87	III
4b	1.38	−7.64	−4.37	−1.79	−13.80	2.11	−11.69	III	**7e**	1.42	−8.30	−4.30	−1.64	−14.24	2.65	−11.59	III
4c	1.75	−7.14	−4.24	−1.53	−12.91	1.43	−11.48	III									

[a] E_{BSSE} = Energy due to basis-set superposition error. ΔE_{el} = Electrostatic energy. ΔE_{pol} = Polarization energy. ΔE_{ct} = Charge-transfer energy. ΔE_{int} = Intermolecular interaction energy. ΔE_{def} = Deformation energy. ΔE_b = Binding energy.

On the basis of the adapted geometries (see Supporting Information) and the magnitude of the overall interaction energies, the complexes studied herein are categorized into three distinct groups: Type I exemplifies the cation−π complexes between N-substituted protonated piperidine and an aromatic ring, where the N−H$^+$ bond of the piperidine points toward the π electron density of the benzene (**1b**−**e**, **2b**−**e**, **3b**−**e**, **5d**, **5e**, and **6b**). The interaction energies range from −6.0 to −10.4 kcal/mol, while the binding energies vary between −5.6 and −9.3 kcal/mol after a small set-off by the deformation energy. Type II represents the prototype piperidine and the (un)-substituted monocyclic aromatic, where the alkyl group (R) in Type I is replaced by a hydrogen to form NH_2^+−π interactions (**1a**−**7a**). The stabilization of these complexes (ΔE_{BE}) approximates from −9.2 to −11.4 kcal/mol, with two polar N−H bonds pointing toward the π cloud of the aromatic ring. This leads to a stronger interaction between the monomeric units than that in Type I. Similar to Type I, the deformation energy in Type II is mostly less than 1 kcal/mol. Type III reflects on the interaction between the protonated amino group and the hydroxyl group of the aromatic ring (**4b**−**e**, **5c**, **6c**−**e**, and **7b**−**e**). This series of complexes possesses the strongest interaction among all three types and is more directed. In fact, Type III complexes are typical hydrogen-bonding interactions rather than cation−π interaction, whose strength ranges from −11.9 to −15.1 kcal/mol. However, the deformation cost in this type is also the highest among all, resulting in net binding energies ranging from −10.1 to −12.6 kcal/mol (Table 1).

When the total interaction energy is decomposed into the electrostatic, polarization, and charge-transfer energy components, our results confirmed the traditional view that electrostatic interactions are the most important in stabilizing cation−π complexes. Although this might be anticipated because of the in vacuo energetics, it is also clear that the electrostatic interaction does not solely dominate the total interaction energy since it explains only 40−58% of the total interaction energy in the complexes. Furthermore, its influence is more with an increase in the strength of the total interactions as can be seen from the electrostatic contributions for Type III complexes

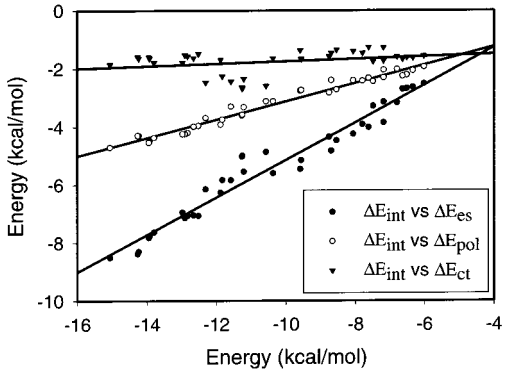

Figure 2. Correlation between the total interaction energy and individual energy terms.

(Table 1). As noted by others and confirmed further in the present study, polarization effects (ΔE_{pol}) are significant for cation−π interactions[16,17] and contribute about 30% towards the total interaction energy. Consistent with Cubero et al.'s findings,[17] the present data also show that the polarization energy is approximately 60% in magnitude when compared to the electrostatic term alone. The energy decomposition analysis also reinforces the importance of charge-transfer effects, which have been omitted in many molecular mechanical models in several previous investigations. In fact, the stabilization due to charge transfer from the π system to the cation fragment contributes 16−25% of the interaction energy. This observation also raises an interesting question. If polarization and charge-transfer effects are so important in cation−π interactions, how can simple electrostatic models employed in molecular mechanics force fields yield meaningful results for cation−π interactions in biological systems?

It is interesting to notice that both the polarization and the electrostatic stabilization energy have good correlations with the total interaction energy as evidenced from the correlation coefficient (r) of 0.92 and 0.87 with a slope of 0.39 and 0.78, respectively (Figure 2). On the other hand, the charge-transfer energy term is essentially independent of the variation of the total interaction energy (ΔE_{ct} vs ΔE_{int}) with a slope of 0.07.

For both Type I and III complexes, ΔE_{ct} is about 1.7 kcal/mol, irrespective of the magnitude of the total interaction. In

(27) Gallivan, J. P.; Dougherty, D. A. *J. Am. Chem. Soc.* **2000**, *122*, 870.

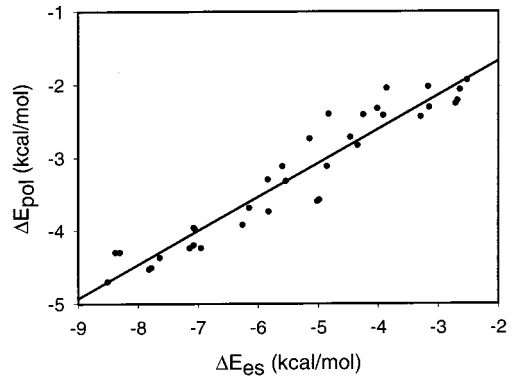

Figure 3. Correlation between electrostatic interaction energy and polarization energy.

Table 2. Basis-Set Dependence on Energy Decomposition Results (kcal/mol) for Methyl-piperidine−Benzene (**1b**)

basis set	E_{BSSE}	ΔE_{el}	ΔE_{pol}	ΔE_{ct}	ΔE_{int}	ΔE_{def}	ΔE_{b}
6-31G(d)	1.68	−4.25	−2.41	−1.41	−8.07	0.26	−7.81
6-31G(d,p)	1.65	−4.13	−2.43	−1.46	−8.02	0.26	−7.76
6-311G(d)	0.91	−4.77	−2.84	−0.80	−8.41	0.25	−8.16
6-311G(d,p)	0.90	−4.46	−2.79	−0.88	−8.14	0.24	−7.90
6-31+G(d)	0.43	−4.10	−3.22	−0.67	−7.99	0.24	−7.75
6-311+G(d,p)	0.57	−4.10	−3.39	−0.41	−7.90	0.24	−7.66

contrast, the charge-transfer energy term is more significant ($\Delta E_{ct} \approx 2.5$ kcal/mol) in complexes **1a**−**7a**, where the positive charge is distributed equally between the two hydrogen atoms. This is reflected by the electrostatic component (ΔE_{el}) for these systems (Table 1). A plot of ΔE_{es} versus ΔE_{pol} (Figure 3) further confirms that these two components are linearly dependent ($r = 0.91$, slope 0.46) and that the sum of these two components dominates the total interaction energy. This relationship allows the modeling of the polarization effects quite effectively using enhanced electrostatic terms. This is one of the primary reasons that effective pairwise potential functions used in molecular mechanics force fields work exceptionally well in biomolecular modeling.[16]

Furthermore, given the fact that there is only a small and nearly constant contribution of the charge-transfer term (ΔE_{ct}) to the total interaction energy, it can be adequately modeled by the use of an offset value in the force field, rather than explicit representations (Figure 2). Figure 2 also implies that with decreasing cation−π interactions, all three individual energy components tend to converge. This suggests that the electrostatic, polarization, and charge-transfer energy terms make similar contributions to the total interaction energy for weakly bound cation−π complexes.

The computations and subsequent discussions in the present work (Table 1) are mainly based on the HF level with a very moderate 6-31G(d) basis set. However, the basis-set dependence of the results was investigated by choosing a representative cation−π model system (**1b**, Figure 1) that is pertinent in the current study and performed BLW-ED analysis with the augmentation of basis sets up to 6-311+G(d,p). Table 2 lists the energy terms at various calculation levels. As expected, the basis-set superposition error reduces with enlarged basis set, and in particular with the inclusion of diffuse functions. As for the energy components, the electrostatic energy term is relatively stable with respect to the basis sets. However, the charge-transfer energy varies from −1.41 to −0.41 kcal/mol when the basis set improves from 6-31G(d) to 6-311+G(d,p). The *lost* energy is regained in the polarization energy term, which increases from −2.41 to −3.39 kcal/mol. This illustrates the basis-set artifacts, as the gradual enlargement of basis sets tends to diffuse the definition of individual atoms. It is essential to find a balance where the basis set is large enough to derive the correct molecular geometries and energetics but small enough to maintain the individuality of atoms (or monomers) in a molecule. More discussions on the basis-set dependency of BLW-ED have been addressed in ref 18c. Because the main goal in the present work is to find the systematic correlations between various energy terms in cation−π systems, we will keep using the results at the HF/6-31G(d) level, although the polarization effect at this level is slightly underestimated.

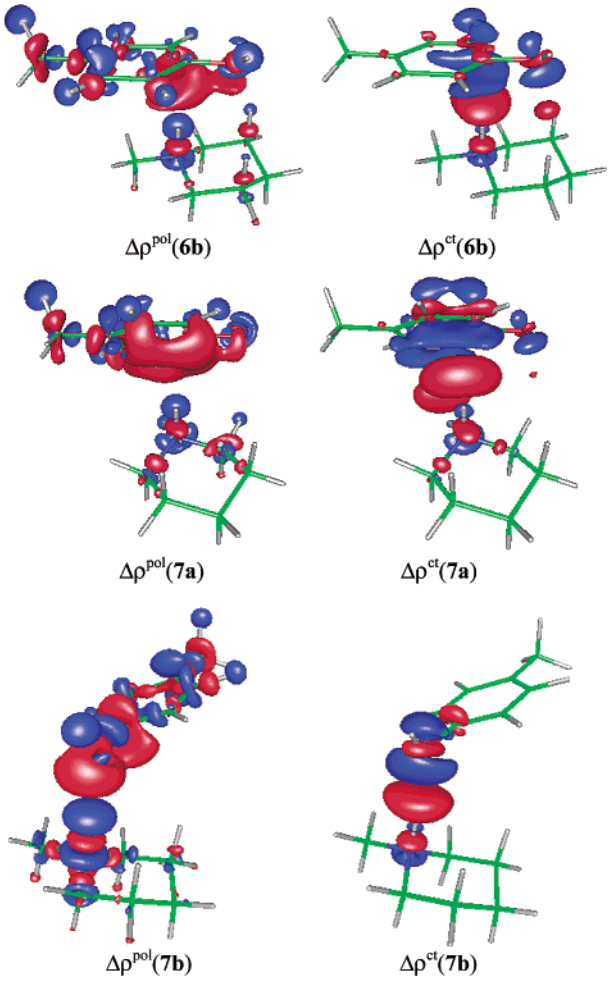

Figure 4. Electron density difference (EDD) maps showing the polarization effect ($\Delta\rho^{pol}$) and charge-transfer effect ($\Delta\rho^{ct}$). The isodensity values for $\Delta\rho^{pol}$ and $\Delta\rho^{ct}$ are 0.012 and 0.004 e/au^3, respectively.

In the BLW-ED method, the wave function for specific intermediate states can be derived, one being the charge-transfer effect "switched off". By comparing the wave functions Ψ^{o}_{AB} and Ψ^{BLW}_{AB}, the polarization effect can be visualized using the electron density difference (EDD) maps. Similarly, a comparison between Ψ^{BLW}_{AB} and Ψ^{HF}_{AB} illustrates the effect due to charge transfer. Figure 4 shows the EDD maps for three complexes **6b**, **7a**, and **7b** representing the three types of interacting pairs. Among the three pairs, both **6b** and **7b** comprise the protonated methylpiperidine and *p*-OH toluene, while **7a** is the complex of protonated piperidine and *p*-OH toluene. In Figure 4, $\Delta\rho^{pol}$

illustrates the charge polarization of the aromatic π ring systems due to the presence of a cation (contours in red indicate a gain in electron density, while blue curves represent a loss of charge density). Obviously the π system is disturbed markedly due to the neighboring cation, and the electron density is pulled to shift toward the direction of the cation. This charge migration not only enhances the electrostatic stabilization, but also moves the charge to the cation as shown in the $\Delta\rho^{ct}$ plots. Because most of the charge-density variation occurs in the substituted aromatics (**3** and **5**), the polarization effect mainly comes from the aromatic substitutions. It is also important to notice that the EDD maps demonstrate the polarization effect to be spread over the whole system (delocalized) unlike the localized nature of the charge-transfer effect (Figure 4).

Although population analysis is arbitrary in obtaining partial charges, it is still tempting to estimate the magnitude of charge transfer because partial atomic charges are one of the key features involved in force field parametrizations. Consequently, the population analyses on the wave functions, Ψ_{AB}^{BLW} and Ψ_{AB}^{HF}, are obtained to evaluate the amount of charge transferred (Δq) from the aromatic unit to the cation. This charge variation is defined as follows:

$$\Delta q = q_{+}(\Psi_{AB}^{HF}) - q_{+}(\Psi_{AB}^{BLW}) \qquad (10)$$

where q_{+} is the overall population in the cation fragment. In this study, the Mulliken population analysis (MA), natural orbital population analysis (NPA), and Merz−Besler−Kollman electrostatic fitting scheme (MBK) have been used. The NPA is based on the overall electron density rather than individual orbitals, and numerous applications have demonstrated its advantage over the Mulliken population analysis. Although all of the three schemes reveal consistent trends (see Supporting Information) with respect to the charge-transfer stabilization energies listed in Table 1, both MA and MBK estimate an average of 0.045e to be transferred, while NPA gives a much lower value (0.012e).

The goal of the present study was to finally translate these results from ab initio to force field domain to gain a better understanding on the nature of the δ-opioid receptor−ligand interactions. However, the limitation of the existing force field methods in treating cation−π interactions explicitly, or for that matter the effects of polarization, restricts our current efforts. Preliminary insight into the proposed hypothesis was obtained by performing automated receptor docking of opioid ligands such as naloxone, naltrexone, and naltrindole (see Supporting Information) onto the transmembrane domain of the homology modeled δ-opioid receptor. As anticipated, the docking solutions align the piperidine ring closer to Asp128 located in TM-III such that the corresponding N-substitutions direct toward TM-VII and the tyramine moiety toward TM-VI. Because the homology models align Tyr129 partially toward the binding pocket,[3] it is also possible that the cation−π interactions between the protonated piperidene ring and the aromatic (Tyr129) significantly stabilize the δ-opiod receptor−ligand complex. This hypothesis can be reinforced by the experimental point mutation results reported by Befort et al.[8] and further supported by the significant stabilization offered by such interactions through the model systems (Type I; Table 1). An alternate hypothesis whereby the tyramine moiety of the ligands π-stack with Tyr129 or possibly with the Tyr129Phe mutant could be completely ruled out, as docking solutions recognizing such an orientation could not be identified.

Conclusions

Ab initio quantum mechanical studies on a series of model systems confirmed the importance of noncovalent cation−π interactions that could have far-reaching implications in modeling opioid receptor−ligand complexes. The large magnitudes of stabilization observed for these model systems suggest a similar trend for opioid ligands as well and the possibility of cation−π stabilization associating the δ-opioid receptor−ligand complexes. This is further supported by the comparable strengths of the alternate salt bridges[27] that could associate the receptor−ligand complexes belonging to the aminergic neurotransmitter families. The BLW-ED approach used to decompose the total interaction energy helps delineate the contribution of the electrostatic, polarization, and charge-transfer energy terms toward the cation−π stabilization. The present results reveal that although the electrostatic interaction makes the largest contribution to cation−π binding, polarization and charge-transfer effects are not negligible. Although previous molecular mechanics studies have suggested improved results when explicit polarizable force fields are employed, simple electrostatic models still enjoy remarkable success in the modeling of biomolecular systems. The success of simple electrostatic models seems to be inconsistent with the argument that both polarization and charge-transfer terms play significant roles in cation−π interactions. However, our energy decomposition reveals good linear relationships between the individual energy terms and the total interaction energy. Therefore, a simple force field without explicit polarization and charge-transfer terms can adequately model biomolecules with remarkable success.[14] This finding also implies that although force fields can be used to investigate chemical and biochemical problems, including enzymatic reactions or drug design, it may be desirable to investigate specific contribution from charge polarization and charge transfer for systems where such effects are deemed to be important.

Acknowledgment. G.S. thanks Dr. Germana Paterlini (UMN) for encouragement on this work. The work is partially supported by the National Science Foundation (J.G.) and the National Institutes of Health (D.M.F., J.G.). The authors also thank the referees for their comments and suggestions toward improving the manuscript. The authors dedicate this work to the late Prof. Peter A. Kollman, UCSF, for his contributions in this field.

Supporting Information Available: Population analysis results, total and zero-point energies for the monomers and complexes, along with the 2-dimensional depiction of the 3-dimensional structures of the complexes detailing the orientation and key geometric parameters (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

JA0174433