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# *Physical and Inorganic Chemistry*

## Conformational Analysis of Macromolecules. VI. Helical Structures of *o*-, *m*-, and *p*-Chlorobenzyl Esters of Poly-L-aspartic Acid[1]

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**Abstract:** Energy calculations have been carried out for the single-stranded *o*-, *m*-, and *p*-chlorobenzyl esters of poly-L-aspartic acid to determine their most stable helical conformations. The *ortho* and *meta* isomers were found to form left-handed $\alpha$-helices; the *para* isomer formed a right-handed one. The predictions for the *ortho* and *meta* isomers have subsequently been verified by experiment, and the result for the *para* isomer is in agreement with previous theoretical and experimental observations. An analysis has been made of the energy contributions which influence the helix sense in these polymers. As an aid in providing a physical basis for the empirical energy functions used in these computations, molecular orbital calculations (with the EHT and CNDO/2 methods) have been carried out on the following model compounds: methyl acetate, toluene, chlorobenzene, *o*-, *m*-, and *p*-chlorotoluenes, benzyl acetate, and *o*-, *m*-, and *p*-chlorobenzyl acetates.

In previous papers[4-6] (referred to as papers III, IV, and V, respectively) of this series, the screw senses of the $\alpha$-helical forms of about 25 homopolyamino acids have been calculated, with agreement between the calculated and experimental screw senses in all cases but one (poly-$\beta$-ethyl-L-aspartate[6]). For those polymers with long side chains, the energies for rotation around the single bonds of the side chains were shown to make very important contributions to the total energy. In this paper the authors explore in more detail the symmetry and barriers of the energy functions for rotation about single bonds in the side chains of several aspartate polymers by carrying out semiempirical molecular orbital calculations (with the Extended Hückel Theory (EHT) of Hoffmann[7] and the Complete Neglect of Differential Overlap (CNDO/2) method of Pople, *et al.*[8,9]) for appropriate model compounds. These molecular orbital methods are used simply to improve and strengthen the physical basis (symmetry and magnitude of rotational energy functions, charge distributions, and method of accounting for near-neighbor electrostatic interactions) on which the empirical energy functions and their parameters, which are described elsewhere,[4-6] are obtained.

A second purpose of this paper is the computation of the screw sense of the $\alpha$-helical forms of the *o*- and *m*-chlorobenzyl esters of poly-L-aspartic acid. Similar calculations had already been carried out[6] for the *p*-chlorobenzyl esters of poly-L-aspartic acid and poly-L-glutamic acid; the calculations for the former were in agreement with experiment[10-13] and those for the

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[4] R. A. Scott and H. A. Scheraga, *J. Chem. Phys.*, **45**, 2091 (1966).

[5] T. Ooi, R. A. Scott, G. Vanderkooi, and H. A. Scheraga, *ibid.*, **46**, 4410 (1967).

[6] J. F. Yan, G. Vanderkooi, and H. A. Scheraga, *ibid.*, **49**, 2713 (1968).

[7] R. Hoffmann, *ibid.*, **39**, 1397 (1963); **40**, 2474, 2480, 2745 (1964).

[8] J. A. Pople and G. A. Segal, *ibid.*, **44**, 3289 (1966).

[9] J. A. Pople and M. Gordon, *J. Amer. Chem. Soc.*, **89**, 4253 (1967).

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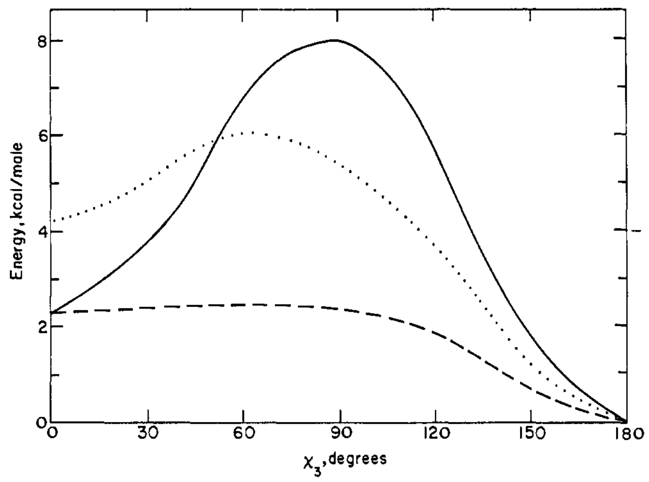

Figure 1. Energy for rotation about the (C=O)—O bond of esters, for $\chi_2 = \chi_4 = 180°$, the minimum-energy values for these angles: —, experimental curve for ethyl acetate (eq 1); ·····, calculated for methyl acetate by the CNDO/2 method; -----, calculated for methyl acetate by the EHT method.

latter were subsequently verified by experiment.[14] It was, therefore, of interest to extend these calculations to the *ortho* and *meta* isomers to determine whether the asymmetric location of the chlorine atom on the benzene ring in these polymers would influence the helix sense of poly-$\beta$-benzyl-L-aspartate (as it is affected by substitution in the *para* position). Because of the modifications made in the methods of computation, as a result of the insight gained from the molecular orbital calculations, the calculations for the *para* isomer[6] are repeated here. Since the *ortho* and *meta* isomers had not been reported previously, they were synthesized and examined[15] by ORD and CD to check the predictions from the calculations reported here.

## Molecular Orbital Calculations for Model Compounds

The following model compounds, which are structurally related to the side chains of the polymers under consideration here, were treated by the molecular orbital methods: methyl acetate, toluene, chlorobenzene, *o*-, *m*-, and *p*-chlorotoluenes, benzyl acetate, and *o*-, *m*-, and *p*-chlorobenzyl acetates; in addition, previous molecular orbital calculations[16] for N-methylacetamide provide a check on the validity of the energy functions for rotation about the bonds of the backbone chain.

For given bond lengths and bond angles, the conformations of these model compounds are defined in terms of the dihedral angles for rotation about the bonds of the molecules. The (polymer) backbone dihedral angles, $\phi$, $\psi$, and $\omega$, for rotation about the N-C$^\alpha$, C$^\alpha$-C$'$, and C$'$-N bonds, respectively, are those defined by Edsall, *et al.*[17] The definitions of the dihedral angles of the aspartate ester side chain follow the general principles of Edsall, *et al.*,[17] and are given in ref 6. For clarity in the discussion, the dihedral angles of the model compounds will be designated so that they have the same nomenclature as the equivalent dihedral angles of the aspartate ester side chain. Thus, $\chi_1$ (the dihedral angle for rotation about the C$^\alpha$-C$^\beta$ bond of the side chain) is analogous to rotation about the C-H bond in the model compounds, and therefore has no meaning for the latter; $\chi_2$ is the dihedral angle for rotation about the $H_3C$—(C=O) bond, $\chi_3$ for rotation about the (C=O)—O bond, $\chi_4$ for rotation about the O-$CH_2$ bond, and $\chi_5$ for rotation about the $CH_2$-Ph bond (whether or not the ring is substituted). $\chi_5 = 0°$ for toluene (or chlorotoluene) when one of the ring H atoms (or Cl in the case of *o*- or *m*-chlorotoluene) is *cis* to one of the H atoms of the methyl group, and for *o*- and *m*-chlorobenzyl acetate when the Cl atom of the C-Cl bond is *cis* to the O atom of the O-$CH_2$ bond; with these definitions, the fully extended aspartate ester side chain has the dihedral angles $\chi_1 = \chi_2 = \chi_3 = \chi_4 = \chi_5 = 180°$.

The computer programs for the molecular orbital calculations are the same as those used previously[16] for model amides, except that, for those compounds containing chlorine atoms, the generalized version of the EHT program[18] (*i.e.*, EHTSPDF) was used. Our program for the CNDO/2 method could not be used for chlorine-containing compounds. The input data for the programs include the Cartesian coordinates of each atom, the number of valence electrons, the Slater exponents for s and p orbitals, and ionization potentials for 2s and 2p orbitals. These parameters for hydrogen, carbon, nitrogen, and oxygen atoms are the same as were used in similar calculations for amides,[16] while the parameters for chlorine were those of Hoffmann, *et al.*[18] For consistency with previous polymer results,[4-6] both the C-H and N-H bond lengths have been taken as 1.0 Å, even though 1.09 Å was used for the C-H bond length in the amide calculations.[16] The C-Cl bond length (1.70 Å), and all other bond lengths and bond angles, were the same as those used previously.[5,6] All bond lengths and bond angles were maintained constant in the calculations (since variations in these quantities cannot be justified within the limits of validity of the molecular orbital methods used). However, the dihedral angles were varied systematically, and the total energy of the molecule and the gross electron population, *i.e.*, the partial charges of each atom, were computed for each conformation; the variation of electron density with dihedral angles was found to be small.

### A. Methyl Acetate

By varying $\chi_3$ in 30° increments (for values of $\chi_2$ and $\chi_4$ equal to 60°, 180°, and 300°), the energy $U(\chi_3)$, for rotation about the (C=O)—O bond of methyl acetate, was computed by the CNDO/2 and EHT methods; some of the data are shown in Figure 1. As can be seen in Figure 1, these curves cannot be represented by

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(16) J. F. Yan, F. A. Momany, R. Hoffmann, and H. A. Scheraga, *J. Phys. Chem.*, **74**, 420 (1970).

(17) J. T. Edsall, P. J. Flory, J. C. Kendrew, A. M. Liquori, G. Nemethy, G. N. Ramachandran, and H. A. Scheraga, *Biopolymers*, **4**, 121, 1149 (1966); *J. Biol. Chem.*, 241, 1004, 4167 (1966); *J. Mol. Biol.*, **15**, 399 (1966); 20, 589 (1966).

(18) R. Hoffmann, G. D. Zeiss, and T. W. Van Dine, *J. Amer. Chem. Soc.*, 90, 1485 (1968).

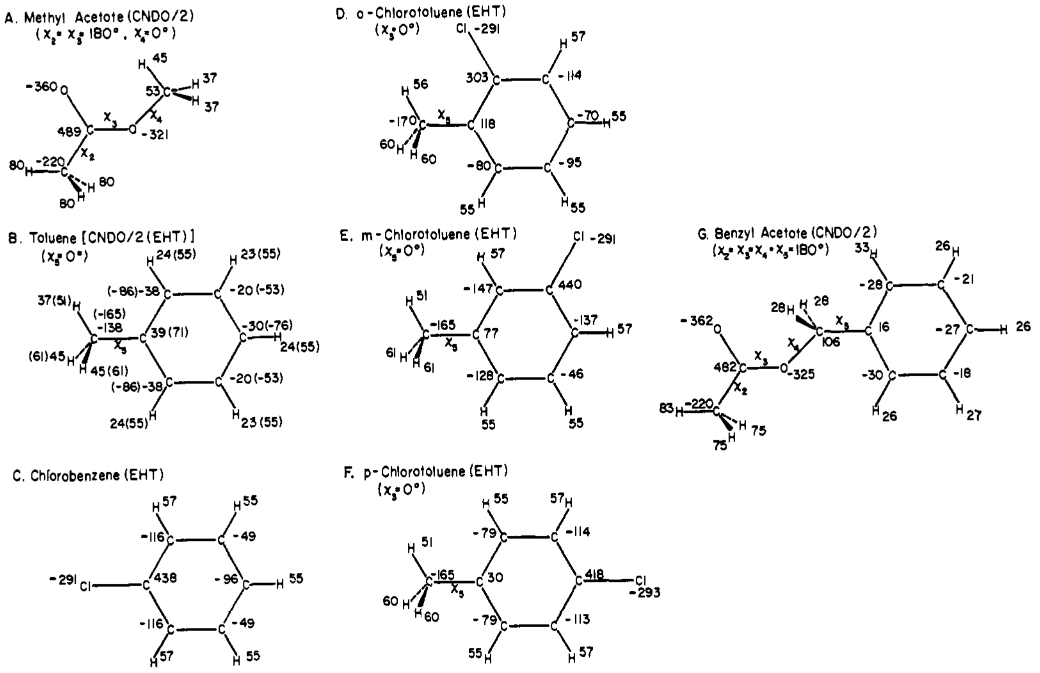

Figure 2. Computed partial charges for the molecules and conformations shown. Where not designated explicitly, the conformation is planar. The values shown should be divided by 1000 to obtain electronic charge units. The numbers in parentheses for toluene were obtained by the EHT method.

a simple cosine curve or by a sum of two cosine terms. The experimental curve for ethyl acetate was previously represented[6] by

$$U_{\text{exptl}}(\chi_3) = (2.3/2)(1 + \cos \chi_3) + (6.85/2)(1 - \cos 2\chi_3) \quad (1)$$

This curve, normalized to zero at $\chi_3 = 180°$, is also shown in Figure 1.

While the theoretical curves do not reproduce the experimental curve exactly, they have the same general shape, with the energy being lowest at $\chi_3 = 180°$ and highest roughly midway between $\chi = 0°$ and $\chi = 180°$. More important, because of the relatively high barrier, the region of experimental interest will be that in the neighborhood of $\chi_3 = 180°$. Therefore, since the three curves are in fair agreement in this region, the experimental one (eq 1) will be used for conformational energy calculations on polyamino acids having side-chain ester groups; in fact, in similar calculations on poly(lactic acid), Brant, *et al.*,[19] fixed $\chi_3$ at 180°.

The electron distribution on each atom of methyl acetate is shown in Figure 2A. These charges were obtained by the CNDO/2 method,[16] since the EHT method usually exaggerates the charge separation of the carbonyl group.[16] Since these charges are close to the empirical ones used in papers IV and V for the ester group, one acquires additional confidence in the empirical values (which reproduce the dipole moment[5]), and the authors will use the empirical values[5] in the conformational energy calculations on the polyamino acids reported here.

The dipole moment of methyl acetate, computed by the CNDO/2 method with the correction of Pople and Gordon,[9] is shown in Table I (experimental dipole moments from ref 20). It is seen to be in reasonably good agreement with the experimental value.

Table I. Dipole Moments of Model Compounds

| | | Dipole moment, D | | |
|---|---|---|---|---|
| | | Calcd | | |
| Compound | Formula | CNDO/2 | EHT | Exptl |
| Methyl acetate | $CH_3COOCH_3$ | 1.54 | | 1.7 |
| Toluene | $CH_3C_6H_5$ | 0.15 | 0.52 | 0.37 |
| Chlorobenzene | $C_6H_5Cl$ | | 2.17 | 1.75 |
| *o*-Chlorotoluene | $CH_3C_6H_4Cl$ | | 1.60 | 1.57 |
| *m*-Chlorotoluene | $CH_3C_6H_4Cl$ | | 2.13 | 1.77 |
| *p*-Chlorotoluene | $CH_3C_6H_4Cl$ | | 2.31 | 2.21 |
| Benzyl acetate | $CH_3COOCH_2C_6H_5$ | 1.57 | | 1.82 |

## B. Toluene, Chlorobenzene, and Chlorotoluenes

Since the parameters for chlorine are not yet available in the CNDO/2 program used here, CNDO/2 calculations were carried out only for toluene, whereas EHT calculations were carried out for all of the molecules in this group. The data were obtained for the range 0–30° (in 10° increments) for toluene and *p*-chlorotoluene, and for the range 0–180° (in 30° increments) for *o*- and *m*-chlorotoluene. The barriers for rotation of the methyl group are shown in Table II.

According to both methods, the rotational energy function for toluene has sixfold symmetry with a very small barrier. As seen in Table II, the introduction of a chlorine atom in the *para* position does not affect the symmetry or the magnitude of this small barrier. The contributions of nonbonded interactions to the barriers for $\chi_5$ in toluene and in *p*-chlorotoluene, computed with the empirical functions used previously,[6] were found to be negligible.

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(19) D. A. Brant, A. G. Tonelli, and P. J. Flory, *Macromolecules*, 2, 228 (1969).

(20) A. L. McClellan, "Tables of Experimental Dipole Moments," W. H. Freeman and Co., San Francisco, Calif., 1963, pp 175–134.

**Table II.** Rotational Barriers for Variation of $\chi_5$

| Molecule | Symmetry | Position of lowest energy, deg | Barrier, kcal/mole CNDO/2 | EHT | Nonbonded | Exptl |
|---|---|---|---|---|---|---|
| Toluene | 6-Fold | 30 | 0.07 | 0.03 | 0.002 | 0.5[a] |
| *p*-Chlorotoluene | 6-Fold | 30 | | 0.001 | 0.002 | |
| *m*-Chlorotoluene | 3-Fold | 60 | | 0.01 | 0.003 | |
| *o*-Chlorotoluene | 3-Fold | 60 | | 1.77 | 1.44 | |

[a] M. V. Volkenstein, "Configurational Statistics of Polymeric Chains," Interscience Publishers, New York, N. Y., 1963, p 72.

When a chlorine atom is introduced into the *ortho* or *meta* position, the rotational energy function is found to have threefold symmetry, with an appreciable barrier for the *ortho* compound but a negligibly small one for the *meta* compound. The contributions from the empirical nonbonded energy[6] to the barriers in these compounds were found to be of the same order of magnitude as those found by the EHT method (see Table II). It is seen that, in all of the compounds listed in Table II, the empirical nonbonded energy presumably accounts for most of the barrier computed by the molecular orbital procedure.

The electron densities, computed for toluene by both molecular orbital methods, and for the chloro derivatives by the EHT method, are shown in Figure 2. The dipole moments computed from these partial charges agree fairly well with experimental values (see Table I); the EHT method provides reasonable values of dipole moments for compounds not containing carbonyl groups, while the CNDO/2 method is applicable even when a carbonyl group is present.[16] The results of Table I provide additional confidence in the use of empirical charges,[6] based on dipole moment data, for conformational energy calculations; see below for the proper procedure to use these charges in such calculations.

## C. Benzyl Acetate

In order to determine the effect on the rotational barrier for variation of $\chi_5$ (the angle of rotation about the $CH_2$–Ph bond) when an ester group is substituted for a hydrogen atom in the methyl group of toluene, we consider here benzyl acetate (and, in the next section, the chlorobenzyl acetates). Using both the EHT and CNDO/2 methods, calculations were carried out for 30° increments in $\chi_5$ between 0° and 180°, with $\chi_2$, $\chi_3$, and $\chi_4$ fixed at 180° (the conformations with least steric hindrance) to reduce the number of computations. The rotational energy function was found to have twofold symmetry with barrier heights of 1.63 and 1.92 kcal/mole according to the EHT and CNDO/2 methods, respectively.

The partial charges obtained with the CNDO/2 method are shown in Figure 2G. Using these charges, the value of 1.57 D of Table I was obtained for the corrected CNDO/2 dipole moment, in fair agreement with the experimental value. The EHT charges were not computed because of the aforementioned problem with molecules containing the carbonyl group.

## D. Chlorobenzyl Acetates

*o*-, *m*-, and *p*-chlorobenzyl acetates serve as good models for the chlorine-substituted aspartyl side chains of interest here. In order to reduce the computing time, $\chi_2$, $\chi_3$, and $\chi_4$ were fixed at 180° and $\chi_5$ was varied in 30° increments between 0° and 180°; despite this restriction on the values of $\chi_2$, $\chi_3$, and $\chi_4$, the information gained about the rotational barrier for variation of $\chi_5$ should be valid. Because of the presence of chlorine, the CNDO/2 method could not be used; hence, partial charges and dipole moments were not computed. Only the EHTSPDF method was used, since it was shown to give[16] reliable indications of barrier heights and symmetries of energy functions.

For *p*-chlorobenzyl acetate, the rotational energy function for variation of $\chi_5$ was found to have twofold symmetry (like benzyl acetate, and in contrast to the sixfold symmetry in toluene and in *p*-chlorotoluene), with a barrier of 1.43 kcal/mole. As in the cases of toluene and *p*-chlorotoluene, the contribution of the nonbonded energy (using empirical functions[6]) to this barrier was found to be small (*i.e.*, 0.15 kcal/mole); thus, this barrier arises primarily from energy contributions other than those represented by empirical nonbonded energy functions.

The rotational energy functions for *o*- and *m*-chlorobenzyl acetates are more complicated, and are shown in Figures 3 and 4, respectively. Interactions between the chlorine atom and the ester group are primarily responsible for the distortion from a symmetrical shape. The EHT curve for the *ortho* isomer (in Figure 3), including the large repulsion at low values of $\chi_5$, can be well simulated (dotted curve in Figure 3) by a sum of empirical nonbonded energies[6] and a twofold cosine expression of the form

$$U(\chi_5) = (U_{\chi_5}/2)(1 - \cos 2\chi_5) \qquad (2)$$

By subtracting the nonbonded interactions from the solid curve of Figure 3, a value 2.40 kcal/mole was obtained for $U_{\chi_5}$. The values of $U_{\chi_5}$, computed in a similar manner for the *meta* and *para* isomers, are 1.78 and 1.43 kcal/mole, respectively. If the empirical electrostatic energy (computed from the empirical partial charges used previously[6]) is added to the dotted curve of Figure 3, the dashed one is obtained. It can be seen that the inclusion of the electrostatic energy does not lead to good agreement with the curve computed by the EHT method. We conclude from this (and from similar results described below for the *meta* isomer) that a different interpretation must be given to eq 2 than was previously[4-6] given to it. Previously,[4-6] eq 2 was regarded as an intrinsic torsional energy, and the empirical electrostatic energy was added in separately. From the results in Figures 3 and 4, one must regard eq 2 as containing the contributions of both the previously designated intrinsic torsional and electrostatic energies.

We digress here to discuss this point further. The result described above arises from the attempt to simulate dipole–dipole interactions by a monopole approxi-

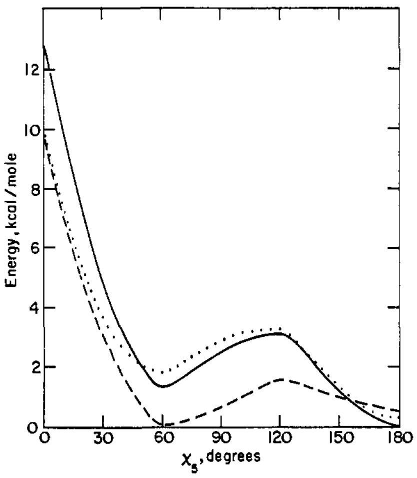

Figure 3. Energy for rotation about the $CH_2$—$(C_6H_4)Cl$ bond of *o*-chlorobenzyl acetate, for $\chi_2 = \chi_3 = \chi_4 = 180°$: —, calculated by the EHT method; ·····, calculated with empirical nonbonded energy function[6] plus $(2.40/2)(1 - \cos 2\chi_5)$; -----, calculated by adding the empirical electrostatic energy[6] to the above dotted curve.

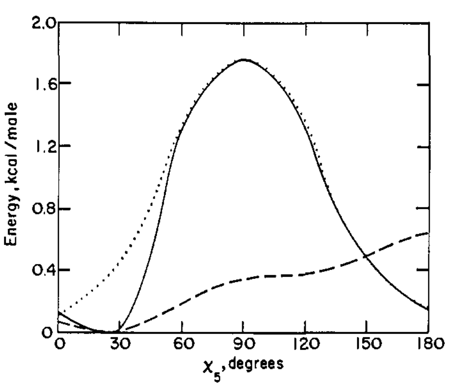

Figure 4. Energy for rotation about the $CH_2$—$(C_6H_4)Cl$ bond of *m*-chlorobenzyl acetate, for $\chi_2 = \chi_3 = \chi_4 = 180°$: —, calculated by the EHT method; ·····, calculated with empirical nonbonded energy function[6] plus $(1.78/2)(1 - \cos 2\chi_5)$; -----, calculated by adding the empirical electrostatic energy[6] to the above dotted curve.

mation. For example, in chlorobenzyl aspartate polymers, we assign partial charges to the amide (CONH), ester (COOC), and C–Cl groups, and (properly) compute the (CONH)–(COOC) and (CONH)–(C–Cl) electrostatic interactions; this procedure is valid because the charged groups, simulating the respective dipoles, are separated by two neutral carbon atoms ($C^\alpha$ and $C^\beta$). However, the (COOC)–(C–Cl) electrostatic interaction creates the problem encountered in the previous paragraph, because these groups are too close to each other for the monopole approximation to the dipole–dipole interaction to be valid. It is for this reason that this electrostatic interaction was omitted from the previous calculations,[6] and the EHT computations in this paper justify this procedure. First of all, it is an approximation to assign partial charges to some atoms and not to others, since calculations by the Poland–Scheraga[21] and the EHT[16] and CNDO/2[16] methods indicate that every atom in the molecule possesses a partial charge. In fact, the molecular orbital procedures indicate that the charge distribution, as represented by molecular orbitals, is spread throughout the molecule in a diffuse manner. One represents this diffuse charge distribution, for example, by using the Mulliken analysis to obtain the point (partial) charges[16] located on each atom. Probably, one should compute the electrostatic pair interactions, with charges on *all* atoms, in conformational energy calculations. The alternative procedure has been adopted previously[4-6] of localizing these charges exclusively on the polar groups, *e.g.*, CONH, COOC, C–Cl, etc. Secondly, this latter procedure is probably valid when the polar groups are far apart, as they are, for example, in the (CONH)–(COOC) interaction cited above; *i.e.*, under these conditions, the dipole–dipole interaction is fairly well accounted for. However, when the polar groups are very close together (specifically, when they are involved in 1,4-type interactions, *i.e.*, when the interacting atoms are separated by three bonds), then the authors believe that, to a first approximation, the cosine formula of eq 2 already includes the short-range electrostatic interaction, and the latter should not be added in as a separate contribution, as indicated in Figures 3 and 4 (of course, the 1,4-type nonbonded empirical energy does have to be included). In summary, the EHT calculations of Figures 3 and 4 provide the basis for *not* including 1,4-type electrostatic interactions in empirical conformational energy calculations on polypeptides. Specifically, in the polymers to be considered here, the electrostatic interactions between the (COOC) and (C–Cl) dipoles in the same side chain will be omitted, as they were previously.[6] Of course, such interactions between these groups in neighboring side chains do have to be taken into account.

Returning to the *meta* isomer, it is seen again that the EHT curve (in Figure 4) can be well simulated by a sum of empirical nonbonded energies[6] and a cosine expression (eq 2) with $U_{\chi_5} = 1.78$ kcal/mole. Again, it is seen that the erroneous inclusion of the empirical electrostatic energy between the (COOC) and (C–Cl) groups would lead to poor agreement with the curve computed by the EHT method.

EHT and empirical nonbonded energies[6] were also computed for the chlorobenzyl acetates by keeping $\chi_2$, $\chi_3$, and $\chi_5$ fixed at 180° to obtain information about the energy for variation of $\chi_4$. These results are shown in Figure 5. It can be seen that both the EHT and nonbonded energy functions have minima at $\chi_4 = 90°$, 180°, and 270°, instead of at the values suggested previously.[6]

Further theoretical molecular orbital calculations are being carried out to investigate the rotational energies for all amino acid side chains, and for the polypeptide backbone; these results will be reported in a future paper.

## Empirical Energies for Polymers

The molecular orbital results reported here and elsewhere[16] were used primarily to improve the physical basis for, and give us increased confidence in, the empirical energy functions[4-6] which have previously[6] yielded good agreement between computed and experimental helix senses in homopolymer derivatives of poly-β-benzyl-L-aspartate[6] and other polyamino acids.[4-6] Thus, the nonbonded functions, and partial charges for the amide (CONH), ester (COOC), and C–Cl groups, used before,[6] are employed here (with a

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[21] D. Poland and H. A. Scheraga, *Biochemistry*, 6, 3791 (1967).

**Table III.** *o*-, *m*-, and *p*-Chlorobenzyl Esters of Poly-L-aspartic Acid; Dihedral Angles and Contributions to the Total Energy for Right- and Left-Handed α-Helical Conformations of Low Energy[a]

| Polymer[b] and type of conformn[c] | Dihedral angles, deg |  |  |  |  |  |  | Side-chain energy, $U_s$ |  |  |  |  |  |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
|  | $\phi$ | $\psi$ | $\chi_1$ | $\chi_2$ | $\chi_3$ | $\chi_4$ | $\chi_5$ | Torsional | Nonbonded | Electrostatic | Backbone energy | Total energy | $\Delta U$ (R − L)[d] |
| *o*-ClPBLA |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Rt(−) | 129.5 | 123.9 | 300.5 | 341.2 | 166.7 | 213.3 | 174.0 | 0.58 | −14.37 | 1.69 | −8.40 | −20.50 |  |
| Rt(−) | 127.4 | 126.8 | 292.3 | 12.2 | 181.4 | 198.0 | 319.9 | 1.16 | −14.52 | 1.11 | −8.14 | −20.39 |  |
| Lt(−) | 227.0 | 239.3 | 178.3 | 7.9 | 187.8 | 146.8 | 33.7 | 0.99 | −13.60 | −0.68 | −8.41 | −21.70 |  |
| Ll(+) | 228.1 | 239.0 | 179.5 | 222.8 | 180.0 | 189.3 | 170.9 | 0.20 | −11.65 | −0.22 | −8.44 | −20.11 | +1.20 |
| *m*-ClPBLA |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Rt(−) | 131.5 | 122.3 | 307.0 | 334.9 | 166.6 | 215.9 | 348.5 | 0.68 | −14.77 | 1.56 | −8.42 | −20.95 |  |
| Rt(−) | 130.0 | 125.0 | 295.7 | 305.0 | 185.2 | 275.9 | 178.6 | 0.10 | −12.50 | 1.40 | −8.22 | −19.22 |  |
| Lt(−) | 227.3 | 239.5 | 168.0 | 25.1 | 202.4 | 150.8 | 16.1 | 1.62 | −15.02 | 0.29 | −8.42 | −21.53 |  |
| Ll(+) | 228.9 | 237.9 | 189.6 | 262.8 | 187.9 | 267.7 | 357.3 | 0.43 | −12.18 | 0.49 | −8.48 | −19.74 | +0.58 |
| *p*-ClPBLA |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Rt(−) | 131.7 | 123.0 | 298.0 | 303.5 | 182.8 | 274.5 | 171.6 | 0.05 | −13.15 | 1.33 | −8.30 | −20.07 |  |
| Rt(−) | 129.4 | 124.3 | 302.5 | 340.5 | 167.6 | 217.4 | 161.8 | 0.64 | −14.09 | 1.78 | −8.38 | −20.05 |  |
| Lt(−) | 226.1 | 240.9 | 166.0 | 27.3 | 203.9 | 152.8 | 14.2 | 1.80 | −13.76 | 0.83 | −8.30 | −19.43 |  |
| Lt(+) | 229.9 | 237.0 | 300.3 | 148.6 | 198.4 | 84.9 | 3.3 | 0.89 | −12.10 | 0.62 | −8.47 | −19.06 | −0.64 |

[a] The energy is expressed in kilocalories per mole per residue. [b] *o*-ClPBLA, *m*-ClPBLA, and *p*-ClPBLA are the *o*-, *m*-, and *p*-chlorobenzyl esters, respectively, of poly-L-aspartic acid. [c] R and L denote right- and left-handed α-helices, respectively; l(+) and l(−) mean that the side-chain conformations are of the longitudinal type, and that they point toward the C-terminal end (+), or toward the N-terminal end (−); t(+) and t(−) denote transverse side-chain conformations which wrap around the backbone in a clockwise (+) or counterclockwise (−) direction, when the helix is viewed from the N-terminal end. [d] $\Delta U$(R − L) is the total energy difference between the right- and left-handed conformations of minimum energy.

dielectric constant of unity, as before[6]). No 1,4-type electrostatic interactions were included (*i.e.*, the interaction between the COOC and C–Cl groups of the same side chain was omitted since it was included in the energy for variation of $\chi_5$); however, 1,4-type nonbonded interactions were included. Intrinsic torsional energies were represented, as before,[4-6] by the usual cosine forms, which do not include the nonbonded energy contributions. The barriers for variation of $\phi$, $\psi$, $\chi_1$, $\chi_2$, $\chi_3$, and $\chi_4$ were identical with those used previously, but the minima of the rotational function for variation of $\chi_4$ were taken at 90°, 180°, and 270° (see Figure 5). For variation of $\chi_5$, eq 2 was used, with the barriers given in section D. The amide group was held fixed at $\omega = 0°$, the planar *trans* conformation. The hydrogen-bond energy function, and all cutoff criteria were the same as before.[6] Thus, the only differences between this and the previous paper[6] occur in the rotational energy functions for varying $\chi_4$ and $\chi_5$.

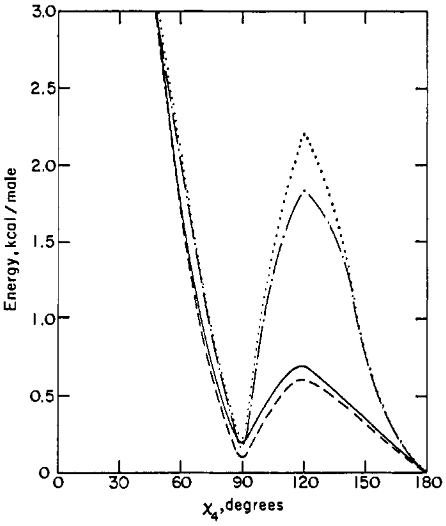

Figure 5. Energy for rotation about the O–CH$_2$ bond of *o*- and *m*-chlorobenzyl acetates, for $\chi_2 = \chi_3 = \chi_5 = 180°$. The shapes of the curves for *p*-chlorobenzyl acetate are similar to both of those for the *meta* isomer, and are not shown: —, calculated for the *ortho* isomer by the EHT method; -----, calculated for the *meta* isomer by the EHT method; ·····, empirical nonbonded energy function[6] for the *ortho* isomer; —·—·—·, empirical nonbonded energy function[6] for the *ortho* isomer.

## Procedure for Energy Minimization for Polymers

The same computer program discussed previously[6] was used to generate ten peptide units with nine side chains, treating all the dihedral angles of a given residue as independent variables and using the regularity condition. Energy minimization was carried out at conformations near the right- and left-handed α-helical regions, *i.e.*, $\phi = 132°$, $\psi = 123°$ for the right-handed helix, and $\phi = 228°$, $\psi = 237°$ for the left-handed one. The starting conformations were those corresponding to minima in the torsional energy functions for variations of the side-chain dihedral angles. Thus, initial values of $\chi_1$ were 180° and 300° (since the 60° position was shown to be one of high energy[5]); $\chi_2$ was initially set at 60°, 180°, and 300°, $\chi_3$ at 180°, and $\chi_4$ at 90°, 180°, and 270°. The initial values for $\chi_5$ differed for each compound, being 60°, 180°, and 300° for the *ortho* isomer (see Figure 3), 0° and 180° for the *meta* isomer (see Figure 4), and only 0° for the *para*-isomer (since the 0° and 180° positions are identical from symmetry considerations). Thus, there were 54, 36, and 18 starting conformations for the *ortho*, *meta*, and *para* isomers, respectively, for each helical sense of the backbone chain.

## Results and Discussion for Polymers

The four lowest energy conformations for each polymer, obtained after energy minimization from each of the starting conformations, are shown in Table III. These are very similar to those obtained previously[6] for poly-β-benzyl-L-aspartate and for the *p*-chloro derivative; hence, the drawings of the conformations in paper V[6] are also applicable here. The notations R, L, t, l, (+), and (−) are defined in paper V[6] (see also footnote *b* of Table III). The difference in total energy, $\Delta U(\mathrm{R} - \mathrm{L})$, between the right- and left-handed conformations of minimum energy, indicates that the *ortho* and *meta* isomers should be left-handed and the *para* isomer right-handed. The *para* isomer is known to be right-handed,[10-13] and the *ortho* and *meta* isomers were synthesized and the predicted helix senses verified.[15]

It is of interest to consider the low-energy conformations in some detail, and also the energy contributions which are responsible for determining whether the right- or left-handed form is more stable. First of all, the unsubstituted[6] and the chloro-substituted polymers have essentially the same values of $\chi_1$ and $\chi_2$. Since the amide–ester dipole–dipole interaction is determined by these angles, this energy is roughly the same in all of these polymers. For poly-$\beta$-benzyl-L-aspartate (PBLA),[6] the electrostatic energy is repulsive for both right- and left-handed $\alpha$-helices, but sufficiently less repulsive for the left-handed form to make this the most stable one. The contribution of the amide–ester dipole–dipole energy would be the same in the chloro-substituted aspartate polymers. On the other hand, for poly-$\gamma$-benzyl-L-glutamate (PBLG),[6] the electrostatic energy is attractive for the right-handed and repulsive for the left-handed $\alpha$-helix, because of the extra methylene group in the glutamate side chain; thus, the right-handed form of PBLG is the most stable one. This influence of the amide–ester dipole–dipole interaction in leading to different helix senses was already pointed out in paper IV[5] in the case of the methyl-L-aspartate and methyl-L-glutamate polymers.

When the influence of the C–Cl dipole is considered, specifically the electrostatic energy for the interaction between the C–Cl dipole and the nearest backbone amide group, the data of Table IV are obtained. The data of Table IV were computed for the lowest energy conformations, which are determined not only by the electrostatic energy but also by the other components of the energy function (see Table III). The side-chain conformations are Rt(−) and Lt(−) for the right- and left-handed helices, respectively. From Table IV, it is seen that the (C–Cl)–amide electrostatic energy favors the left-handed form in all cases, in the order *ortho* < *meta* < *para*. In the left-handed forms, the C–Cl dipole is nearly antiparallel to the nearest backbone amide dipole in the *ortho* and *meta* isomers (see Figure 6), while it is more perpendicular in the *para* isomer; the interdipole distance increases in going from the *ortho* to the *meta* to the *para* isomer. In the right-handed forms, the C–Cl and amide dipoles are less well aligned with respect to each other, being somewhat antiparallel for the *ortho* and *para* isomers, but now parallel for the *meta* isomer (see Figure 6). These orientations are reflected in the signs of the data of Table IV. In the case of the *ortho* and *meta* isomers, the (C–Cl)–amide and ester–amide electrostatic energies dominate the influence of the nonbonded and torsional energies, thereby favoring left-handedness; however, the *para* isomer is right-handed, even though the electrostatic energy favors left-handedness, because of the contribution from the torsional energy. In the case of the *meta* isomer (shown in Figure 6), the reversal of the orientation of the C–Cl dipole in the right-handed form is due to the nonbonded interaction, which overrides the electrostatic energy in this case.

**Table IV.** Electrostatic Energy for the Interaction of the C–Cl Group with the Nearest Backbone Amide Group

| | Electrostatic energy, kcal/mol | | |
|---|---|---|---|
| Polymer | Right-handed | Left-handed | $\Delta U_{\mathrm{elec}}(\mathrm{R} - \mathrm{L})$ |
| *o*-ClPBLA | −0.12 | −0.46 | 0.34 |
| *m*-ClPBLA | +0.03 | −0.59 | 0.62 |
| *p*-ClPBLA | −0.08 | −0.89 | 0.81 |

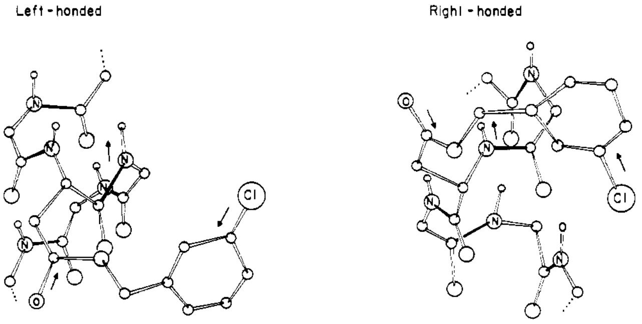

Figure 6. Orientation of the side chains of the left- and right-handed $\alpha$-helices [Lt(−) and Rt(−)] of *m*-ClPBLA. The solid arrows represent the directions of the C–Cl, ester, and amide dipoles, respectively.

## Conclusion

The energy minimization technique has been used to compute the preferred helix sense of the *o*-, *m*-, and *p*-chlorobenzyl esters of poly-L-aspartic acid. The predictions for the *ortho* and *meta* isomers have subsequently been verified by experiment,[15] and the result for the *para* isomer agrees with previous theoretical[6] and experimental[10-13] conclusions. It was of interest to find two more left-handed $\alpha$-helices (those of the *ortho* and *meta* isomers), since there are very few left-handed $\alpha$-helices known, *viz.*, only those of poly-$\beta$-methyl-L-aspartate[22,23] and poly-$\beta$-benzyl-L-aspartate,[24-26] both of whose helix senses have also been obtained correctly in the theoretical investigations.[5,6]

**Acknowledgment.** We are indebted to Professor R. Hoffmann for making the EHT and CNDO/2 computer programs available to us.

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