

mixture was allowed to cool to room temperature and directly applied to a Sephadex G-25 gel filtration column and eluted with water. The desired metalloprotein complex was then separated from the small amounts of unreacted peptide using ion exchange chromatography on CM-Sephadex C-25 with a 0-2 M NaCl salt gradient in 50 mM MES pH 6.5 buffer. Final purification as well as desalting was performed using RP HPLC to afford about 15 mg of the desired metalloprotein. Formation of the desired complex was established by FIB mass spectroscopy  $m/z = 5563$  (Figure 5) and the following characteristic ultra-violet and visible absorption bands:  $\lambda_{\max}$  (nm) 255, 300, and 470 br.

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ation are gratefully acknowledged for their support for of this work. We are also grateful to Dr. C. Singh and V. N. Balaji for the computer-generated model, M. Pique for the graphics presentation, Dr. G. Siuzdak for mass spectroscopic analysis, and colleagues B. Oskouian for help in the design and R. Lerner, K. C. Nicolaou, P. Wright, L. Walters, and J. Skolnick for valuable discussions.

**Registry No.** 2,2'-Bipyridyl-5-carbonyl-GELAQKLEQALQKLA-NH<sub>2</sub>, 138060-62-5; ethyl 6-chloronicotinate, 49608-01-7; 2-(trimethylstannyl)pyridine, 13737-05-8; 5-(ethoxycarbonyl)-2,2'-bipyridine, 56100-24-4; 5-carboxy-2,2'-bipyridine, 1970-80-5.

## Resonance Interactions in Acyclic Systems. 3. Formamide Internal Rotation Revisited. Charge and Energy Redistribution along the C-N Bond Rotational Pathway

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Contribution from the Department of Chemistry, Yale University, New Haven, Connecticut 06511. Received January 24, 1991

**Abstract:** The changes that occur during the rotation of the amino group of formamide have been studied in some detail. Geometry optimizations at the MP2/6-31G\* level confirmed the relatively large increase in C-N bond length but small decrease in the C-O length on going from the planar structure to the rotational transition state. A calculation of the force constants for formamide and for the transition state showed that the carbonyl force constant changed relatively little, but the C-N constant changed by about 30%. The path followed in the rotation was studied starting with the saddle point geometry and following it computationally down to the ground state. The geometrical changes are discussed. The electron populations were calculated for a number of structures along the reaction coordinate by numerical integration of the charge density within uniquely defined atomic volumes. The oxygen population was little affected by the rotation and the main charge shift was between carbon and nitrogen. The electrostatic potentials for the structures also were examined and converted to effective charges for spherically symmetrical atoms. All of the analyses indicated that essentially all of the interactions leading to the rotational barrier originate in the C-N bond and that the oxygen does not participate to a significant extent. The direction of the charge shift between C and N was in opposite directions for the electron populations derived by integration of the charge density, and by fitting the electrostatic potentials. However, this was due to the difference in the definition of the atoms, being anisotropic in the first case and spherically symmetrical in the second. All of the observations can be rationalized on the basis of the assumption that stabilization of the lone pair on nitrogen is the most important factor in determining both structures and energies.

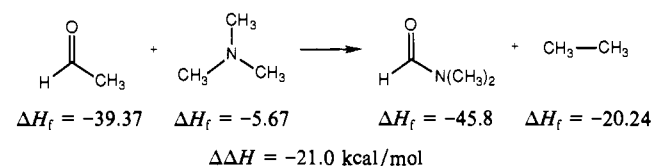
### 1. Introduction

The amide group is one of the most important functional groups in chemistry. Its planarity and relatively high barrier to rotation about the C-N bond are important factors in determining the conformations of peptides and related compounds. Most of the properties of amides are readily rationalized by postulating amide resonance of the type



Invocation of partial double bond character for the C-N bond allows one to account for the rotational barrier, since this presumably stabilizing interaction would be lost when the amino group is rotated by 90°. Similarly, planarity of the amino group would be required in order to achieve a maximum resonance interaction. Addition of a nucleophile to the carbonyl group would result in loss of the resonance interaction, and so one would correctly predict that these additions would be unfavorable. The resonance interaction model also provides a convenient explanation for the observed stabilization achieved by the combination of an amino

group and a carbonyl group. The magnitude of this interaction can be estimated from the following hypothetical reaction (kcal/mol)<sup>1</sup>



The energy change is close to that for the rotational barrier, indicating that the corresponding reaction forming the 90° rotated structure would be close to thermoneutral.

Despite the success of the above resonance picture in explaining a large body of data, there are some observations which cast doubt on this simple interpretation. In an earlier investigation of the rotational barrier in formamide, in which the observed barrier was successfully reproduced via ab initio molecular orbital calculations using the 6-31G\*\* basis set, the changes in electron population and structure on rotation were found not to be in accord with an electron donation from the amino group to the carbonyl

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(1) Cox, J. D.; Pilcher, G. *Thermochemistry of Organic and Organometallic Compounds*; Academic Press: New York, 1970.

**Table I.** Structural Parameters of Formamide Conformers<sup>a</sup>

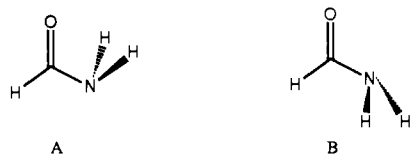
parameter	planar			90° structure (A)		270° structure (B)	
	HF	MP2	obs <sup>d</sup>	HF	MP2	HF	MP2
energy (6-31G*)	-168.93070	-169.40538		-168.90569	-169.37878	-168.90114	-169.37462
$\Delta E$ (kcal/mol)	0.0	0.0		15.69	16.69	18.55	19.30
energy (6-31G**)	-168.94048			-168.91510		-168.91072	
$\Delta E$ (kcal/mol)	0.0			15.98		18.67	
$r_{C=O}$	1.1927	1.2238	1.219	1.1832	1.2169	1.1789	1.2124
$r_{CN}$	1.3489	1.3606	1.360	1.4273	1.4421	1.4230	1.4368
$r_{CH}$	1.0910	1.1046	1.098	1.0876	1.1004	1.0943	1.1090
$r_{NH}$	0.9957 <sup>b</sup>	1.0107 <sup>b</sup>	1.002	1.0055	1.0225	1.0046	1.0220
	0.9929 <sup>c</sup>	1.0084 <sup>c</sup>	1.002				
$\angle N-C-O$	124.95	124.73	124.5	125.05	125.38	123.27	122.83
$\angle H-C-N$	112.66	112.37	112.7	113.48	113.06	116.39	116.71
$\angle C-N-H$	119.33 <sup>b</sup>	118.95 <sup>b</sup>	118.8	108.48	107.24	106.78	108.35
	121.79 <sup>c</sup>	121.83 <sup>c</sup>	121.4				
	0.0	0.0	0.0	57.06	55.74	121.71	123.27

<sup>a</sup>Total energies are in hartrees, bond lengths in Å, and bond angles in deg. Geometries were determined using the HF/6-31G\* method except where noted. <sup>b</sup>Hydrogen eclipsed with carbonyl. <sup>c</sup>Hydrogen eclipsed with aldehyde hydrogen. <sup>d</sup>O=C-N-H torsional angle.

oxygen.<sup>2</sup> First, it was found that whereas the C-N bond increased in length by 0.08 Å on a 90° rotation, the carbonyl bond length decreased by only 0.01 Å (Table I). If the resonance model were really descriptive, one might have expected a more equal change in bond lengths. Second, the integrated electron population<sup>3</sup> at the nitrogen was found to be greater in the lower energy planar form than in either of the rotated saddle point structures.<sup>2</sup> The electrons were taken from the carbon, and the electron population at the carbonyl oxygen was almost unchanged during the rotation. Here, the direction of the charge shift is opposite to that expected from the resonance formulation, and the oxygen does not appear to be significantly affected by the rotation.

## 2. Structural Data

Before proceeding to an interpretation of the origin of the properties of formamide, it appeared necessary to be sure that the calculated structural changes on rotation were correct. It is possible (although unlikely) that they were an artifact resulting from the use of an RHF treatment. In order to see whether or not correction for electron correlation would affect the calculated structural changes, we have carried out geometry optimizations at the MP2/6-31G\* level which has been found to give bond lengths and angles that are in very good agreement with experimental data.<sup>4</sup> The structural parameters for formamide and the saddle point rotamers A and B are compared with the RHF results and the experimental data in Table I. It can be seen that although the bond lengths increase somewhat on correction for electron correlation, the difference in the calculated changes in C-N and C-O bond lengths persists. Again, it appears that the main changes are in the C-N bond and that the C-O bond is relatively unaffected by rotation.



The calculated barrier height at the MP2/6-31G\* level (16.7 kcal/mol) is in good agreement with the experimental barrier (18–19 kcal/mol).<sup>5</sup>

Information has been obtained on the geometrical changes that occur for formamide vibrationally excited for the CN bond ro-

**Table II.** Some Significant Force Constants (mdyn/Å) in Formamide Conformers

force constant	planar	90° structure (A)	270° structure (B)
C-N stretch	8.22	6.15	6.20
C=O stretch	15.80	16.95	17.30
C=O/C-N interaction	1.59	1.01	1.00

tation, and they involve significant stretching of the CN bond and a decrease in HNH angles,<sup>6</sup> as has been found in the above calculations. Structural data for amides that are prevented from achieving planarity because of geometrical constraints have recently been reported.<sup>7</sup> In both cases, the CN bond was found to have lengthened without much of a change in the CO bond length. Thus, these data are in very good accord with the theoretical predictions.

## 3. Vibrational Spectrum

It is well-established that the rotation of the amino group of an amide leads to a significant change in the carbonyl stretching frequency.<sup>8</sup> This increase in frequency could be caused by either a change in the carbonyl stretching force constant or a change in the interaction between the carbonyl group and other groups such as the C-N bond. Since the magnitude of a stretching force constant can, in a qualitative sense, be related to the strength of a bond, any change in bond order which occurs during rotation about the C-N bond should be reflected in the calculated force constants. In order to explore this problem, we have investigated the infrared spectrum of formamide. Since it is not possible to observe the spectrum of the saddle point conformers A and B, it was necessary to make use of theoretical calculations in order to obtain the desired information.

The HF/6-31G\* frequency calculations lead to force constants that are about 20% too large because the theoretical geometry has bond lengths that are somewhat too short and the observed vibrational frequencies are for anharmonic vibrations whereas the theoretical force constants are for harmonic vibrations which have a higher frequency. Nevertheless, it is generally found that the force constants for corresponding modes are overestimated by essentially the same amount,<sup>9</sup> and so the calculated values may usefully be compared. The force constants of interest are those

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**Table III.** Calculated and Experimental Vibrational Frequencies of Formamide ( $\text{cm}^{-1}$ )

exptl <sup>a</sup>	planar <sup>b</sup>	90° structure (A) <sup>b</sup>	270° structure (B) <sup>b</sup>
289	99	444 <sup>i</sup>	398 <sup>i</sup>
565	544	565	595
602	593	850	833
1030	1021	904	836
1059	1041	1068	1029
1255	1213	1216	1213
1378	1380	1363	1384
1572	1575	1573	1583
1734 <sup>c</sup>	1760 <sup>c</sup>	1788 <sup>c</sup>	1816 <sup>c</sup>
2852	2923	2962	2880
3451	3492	3362	3368
3545	3614	3433	3440

<sup>a</sup>Data from: Evans, J. C. *J. Chem. Phys.* **1954**, *22*, 1228. King, S. T. *J. Phys. Chem.* **1971**, *75*, 405. <sup>b</sup>Corrected by multiplying all values under 2000  $\text{cm}^{-1}$  by 0.88 and all those over 2000  $\text{cm}^{-1}$  by 0.91. <sup>c</sup>Carbonyl stretching mode.

involving the C, N, and O, and they are summarized in Table II. It can be seen that the carbonyl stretching force constant does not change much and the major changes are found with the C—N stretching force constant and the C=O/C—N interaction constant. The carbonyl force constant for rotamer A is close to that calculated for acetaldehyde using the same basis set (16.58 mdyne/Å).<sup>10</sup> The CO force constant decreases only by 7% on going to the planar form, whereas the CN constant increases by 32%.

In using theoretically calculated force constants, it has been found that a simple scaling scheme normally reproduces the experimental frequencies with a root-mean-square error of only about 25  $\text{cm}^{-1}$ .<sup>11</sup> The application of these scaling factors leads to the scaled frequencies that are given together with the observed values in Table III. Except for the lowest frequency band, there is quite good agreement. This low-energy band corresponds to an  $\text{NH}_2$  wagging mode which is known to have positive anharmonicity resulting from a stiff quartic potential which is not represented in a harmonic frequency analysis.<sup>12</sup>

Analysis of the vibrational data given in part in Table II suggests that the increase in carbonyl stretching frequency in the rotated conformers arises from changes in the C=O force constant and through the significant coupling interaction of the carbonyl stretch with the C—N bond stretching mode. The small change in the carbonyl stretching force constant upon rotation about C—N bond suggests that the nature of that bond is not largely affected, while the C—N bond becomes much softer in the transition structures. This is consistent with the large observed lengthening of the C—N bonds in structures A and B and the relatively insensitivity of the C=O bond length to the C—N bond changes.

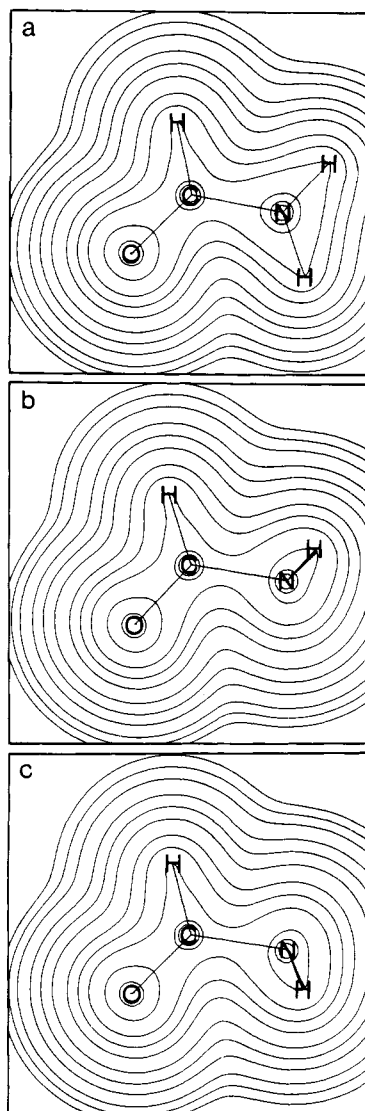
The carbonyl vibrational frequency was calculated to increase by 28  $\text{cm}^{-1}$  on going from the planar to the 90° rotated form (A) and by 56  $\text{cm}^{-1}$  in going to the 270° rotated form (B). These changes are consistent with the ca. 60  $\text{cm}^{-1}$  higher carbonyl stretching frequency seen in the twisted bridgehead lactam 1-azabicyclo[2.2.2]octan-2-one compared to open chain amides.<sup>7</sup>

Even though the change in the carbonyl force constant is small, it actually predicts a somewhat greater change in the calculated difference in the harmonic carbonyl vibrational frequencies between the planar and rotated conformers than is found. Consequently, the large C=O/C—N cross-term must also be taken into

(10) Wiberg, K. B.; Walters, V.; Colson, S. D. *J. Phys. Chem.* **1984**, *88*, 4723.

(11) Vibrational frequencies calculated using the HF/6-31G\* level of theory compare very well with experimental values if all frequencies below 2000  $\text{cm}^{-1}$  are scaled by 0.88 and all higher frequencies scaled by 0.91. Cf.: Wiberg, K. B.; Walters, V.; Colson, S. D. *J. Phys. Chem.* **1984**, *88*, 4723. Wiberg, K. B.; Dempsey, R. C.; Wendoloski, J. J. *J. Phys. Chem.* **1984**, *88*, 5596. Wiberg, K. B.; Walters, V. A.; Wong, K. N.; Colson, S. D. *J. Phys. Chem.* **1984**, *88*, 6067.

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**Figure 1.** Projection density plots for all the electrons of (a) formamide, (b) rotamer B, and (c) rotamer A. The amino group is to the right and the oxygen is to the left.

account when comparing these data. In this case, the cross-term serves to reduce the difference between the calculated carbonyl stretching frequencies of the planar and transition-state forms. It might also be noted that the  $\sim 60\text{-cm}^{-1}$  change in vibrational frequency is small in comparison to the difference between C=O and C—O stretching frequencies ( $\sim 600\text{ cm}^{-1}$ ).

Although the force constants are interesting and provide some indication of the changes in bonding on rotation, it would be more useful to have a direct measure of the bond orders. The force constants are affected by bond orders, bond lengths, and other factors and are therefore at best an indirect indicator of bond order. Some time ago, we proposed a bond index that was the covalent bond order in zero differential overlap semiempirical MO calculations.<sup>13</sup> This has been extended to ab initio calculations by others,<sup>14</sup> but we have not been satisfied with these procedures because they have the inherent problems associated with the use of atomic orbitals in calculating populations and other properties.<sup>15</sup> Recently, Cioslowski<sup>16</sup> has proposed a related covalent bond order based on the atomic overlap matrix as defined in the theory of

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(15) Cf.: Glaser, R. *J. Comput. Chem.* **1989**, *10*, 118.

(16) Cioslowski, J.; Mixon, S. T. *J. Am. Chem. Soc.* **1991**, *113*, 4142.

**Table IV.** Calculated Covalent Bond Orders for Formamide Conformers<sup>a</sup>

bond	planar			90° (A)			270° (B)		
	$\pi$	$\sigma$	total	$\pi$	$\sigma$	total	$\pi$	$\sigma$	total
C=O	0.458	0.668	1.127	0.571	0.677	1.248	0.584	0.666	1.249
C-N	0.229	0.655	0.884	0.046	0.844	0.891	0.048	0.839	0.887
C-H	0.003	0.895	0.898	0.004	0.917	0.921	0.004	0.920	0.924
NH <sub>a</sub>	0.029	0.745	0.775			0.830			0.841
NH <sub>b</sub>	0.031	0.760	0.792						
N...O	0.134	0.154	0.288	0.057	0.157	0.215	0.061	0.153	0.214

<sup>a</sup>Separation into  $\sigma$  and  $\pi$  is not easily defined for out-of-plane hydrogens in the rotated forms.

atoms in molecules.<sup>3</sup> We have applied this procedure to the three rotamers of formamide giving the results shown in Table IV.

The carbon-carbon double bond in ethylene is calculated to have a covalent bond order of 1.96, close to the nominal 2.0.<sup>15</sup> One might then note the relatively small covalent bond order for the carbonyl group, suggesting that it has considerable ionic character. This is in good accord with the large C=O bond dipole,<sup>17</sup> and with calculations of the electron populations at C and O of carbonyl groups.<sup>18</sup> The much greater electronegativity of oxygen as compared with carbon leads to a charge shift from C to O. This may be seen in projection density<sup>19</sup> plots in which all of the charge density above and below the molecular plane is collapsed onto that plane. Plots for the total charge density are shown in Figure 1, and those for just the  $\pi$  electrons are shown in Figure 2. It might be noted that both the  $\sigma$  and  $\pi$  components of the bond are polarized, and so the total covalent bond order is divided approximately equally between  $\sigma$  and  $\pi$ .

In Figure 1 it can be seen that the oxygen contours occupy most of the volume associated with the C=O bond leading to a large electron population for the oxygen and a small population for the carbon. In this way, a carbonyl group is best written as C<sup>+</sup>-O<sup>-</sup>, and the relatively short bond length may be attributed to a coulombic attraction between C and O. The contours in the vicinity of the carbonyl group are essentially unchanged on rotation about the CN bond, again indicating a lack of participation of this bond in the C-N bond rotation process.

The resonance picture is concerned with the  $\pi$  electrons, and their contours in Figure 2 again show a large shift in charge density from carbon to oxygen. However, there is essentially no change for the C=O bond on rotation about the CN bond. It is clear that the carbonyl group is a spectator and not an active participant in the C-N bond rotation. The small change in C-O bond order on rotation may be attributed to the changes in charge at the atoms and a change in the ionic character of the bond.<sup>20</sup> There is no significant change in the C-N covalent bond order on rotation. However, in the planar form there is a significant  $\pi$ -covalent bond order (0.229) which almost disappears in the 90° rotated form. In the latter, this is compensated for by an increase in the  $\sigma$  bond order. This is in agreement with the finding that there is less C-N charge separation in the rotated form than in the planar form (see below).

All of these data are consistent with the proposal that any resonance interactions present in formamide do not affect the carbonyl C=O bond to the large extent required to support the standard resonance model. Recent VB calculations on formamide also support this view.<sup>21</sup>

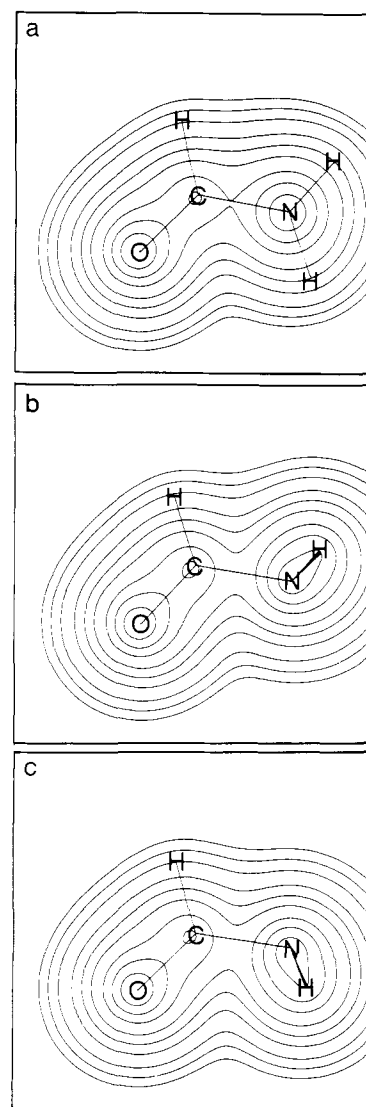
(17) Aldehydes and ketones have dipole moments of 2.6–2.7 D, most of which may be attributed to the carbonyl group.

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(20) The electron populations for planar formamide were as follows: C, 4.023; H, 1.035; O, 9.392; N, 8.476; H, 0.529 and 0.544. For 90° rotated formamide they were as follows: C, 4.249; H, 1.005; O, 9.341; N, 8.222; H, 0.592. We have examined the populations and bond orders for a number of types of compounds containing groups and have found a linear relationship between the population at oxygen and the C-O bond order (Wiberg, K. B.; Hadad, C. M.; Rablen, P. R.; Cioslowski, J. To be published).

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**Figure 2.** Projection density plots for the  $\pi$  electrons of (a) formamide, (b) rotamer B, and (c) rotamer A.

#### 4. Atomic and Structural Changes during Rotation

It was of interest to us to see how the structural parameters and atomic properties change during rotation about the C-N bond. To accomplish this, we started with the lower energy saddle point conformer (A) and carried out a search for the path connecting this conformer to the planar form using a highly damped mode-walking algorithm with the force constants evaluated analytically (HF/6-31G\*) at each point.<sup>22</sup> The calculated rotational pathway was found to begin with the amino hydrogens gauche to the carbonyl group, followed by rotation of the pyramidal NH<sub>2</sub> with little change in hybridization until the hydrogen

(22) The calculation was performed using a harmonic mode-walking algorithm implemented in the developmental version of GAUSSIAN90.<sup>29</sup>

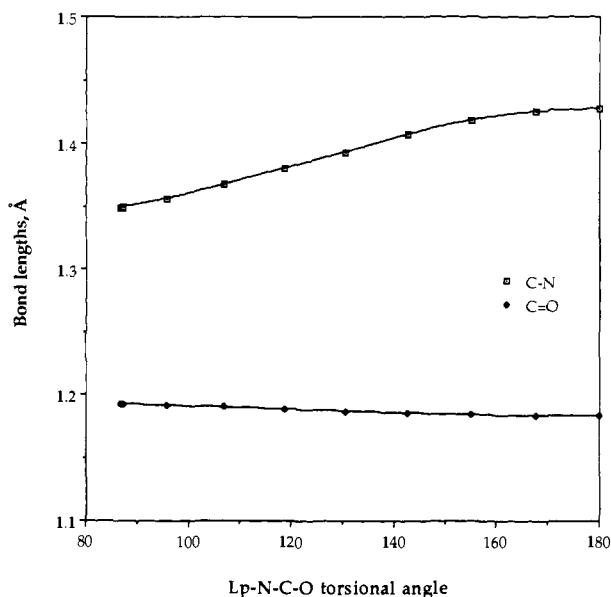


Figure 3. Bond length changes for formamide on rotation about the C-N bond.

rotating toward the carbonyl group overshoots by about  $8^\circ$ . At this point, the hydrogen becoming anti to the carbonyl is still about  $25^\circ$  out of plane, but the calculated energy of this structure is within 0.5 kcal/mol of the planar form. This demonstrates that considerable distortion of the amino group in the planar structure is possible with little energy cost.

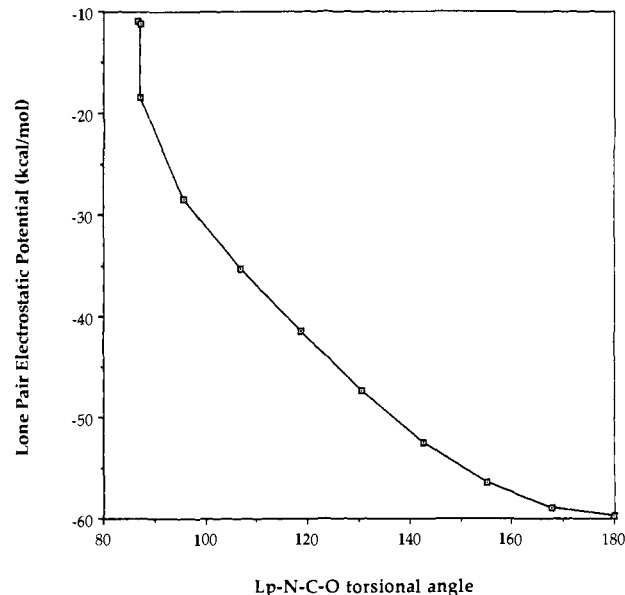
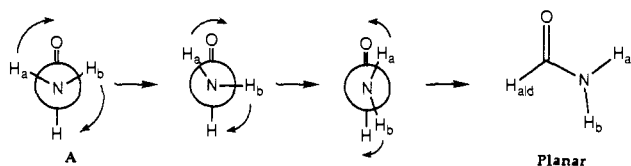
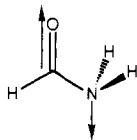


Figure 4. Electrostatic potential minima in the vicinity of the amide nitrogen.

The mode-walk technique resulted in our finding 33 intermediate structures, from which 11 were chosen for further study using the 6-31G\*\* basis set. The reaction coordinate was taken as the torsional angle between the carbonyl and the nitrogen "lone pair". The latter was defined by a line from the nitrogen to the point at which the electrostatic potential was most negative. The C=O and C-N bond length changes that occur during rotation are shown in Figure 3 as a function of this reaction coordinate. Whereas the C-N bond length increases significantly on rotation from the planar to rotamer A, the changes in the C-O bond length are negligible.



Nitrogen lone-pair dihedral angle with C=O bond

Figure 4 shows the dependence of the "lone pair" electrostatic potential value on the progress of the transition from the planar form to rotational transition state A. It can be seen from the figure that when the amide is close to planar the electrostatic potential well is quite shallow, with a potential minimum of only  $-10.9$  kcal/mol. The electrostatic potential well deepens considerably even with small, energetically-negligible deformations from the planar structure, and finally arrives at a value of  $-59.7$  kcal/mol at the rotational transition structure. This potential well is more shallow than the value expected for a typical primary amine such as methylamine, which has an electrostatic potential well value of  $-82.5$  kcal/mol, presumably as a result of the proximity of the orthogonal electronegative carbonyl group.

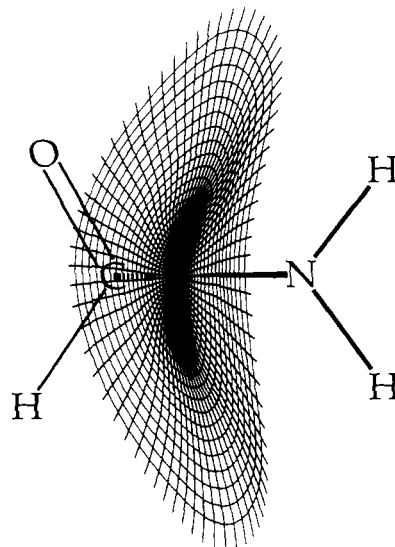
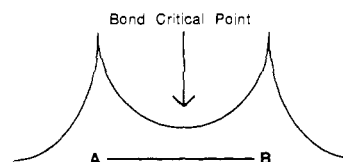


Figure 5. An example of an interatomic surface.

## 5. Electron Populations

We may now turn to one of our special concerns, the flow of electron population and electronic kinetic energy between the atoms in the structures connecting the planar and saddle point conformers. The 6-31G\*\* atomic electron populations were evaluated at each of the 11 selected intermediate structures using Bader's theory of atoms in molecules.<sup>3</sup>

Direct analysis of the electron density is capable of providing all pertinent information concerning molecular behavior and energetics. To understand this technique, one must envision the molecule as a collection of atomic "attractors" (nuclei) surrounded by a sea of charge density. Between each pair of bonded atoms, there exists a pathway of greatest charge density called the *bond path*. Somewhere along this path there is a point of minimum density known as the *bond critical point*. A bond path connecting atoms A and B containing just such a critical point is illustrated below:



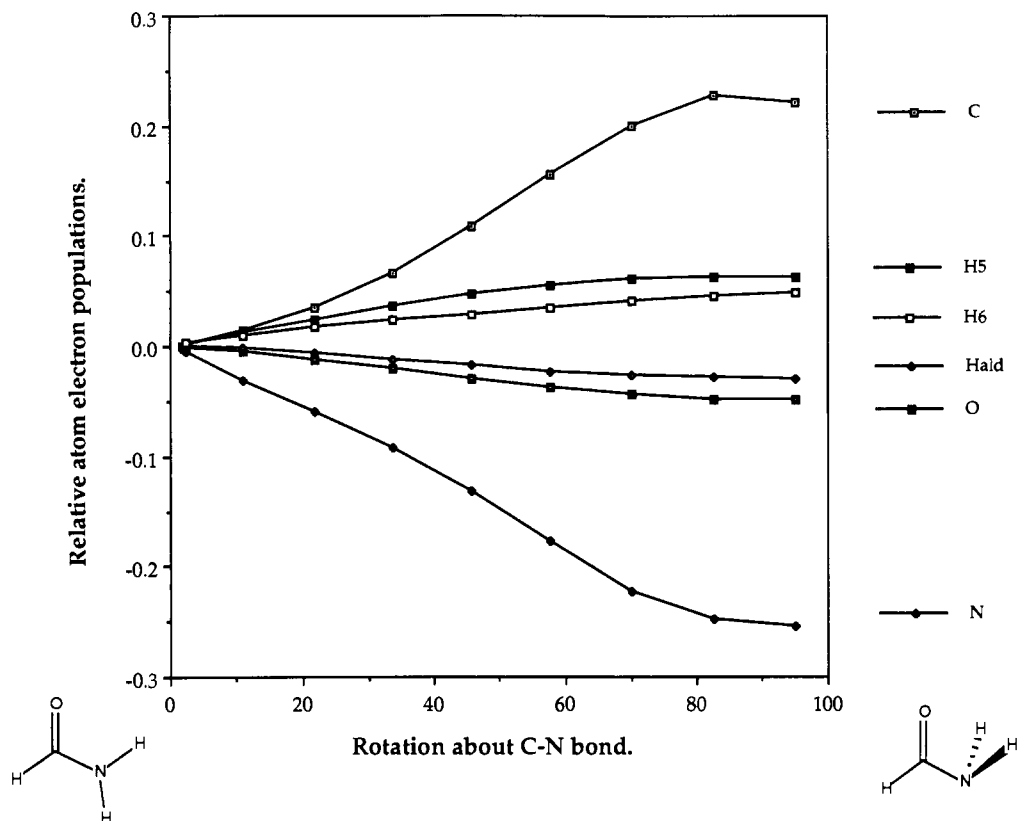


Figure 6. Changes in electron population on rotation about the C-N bond.

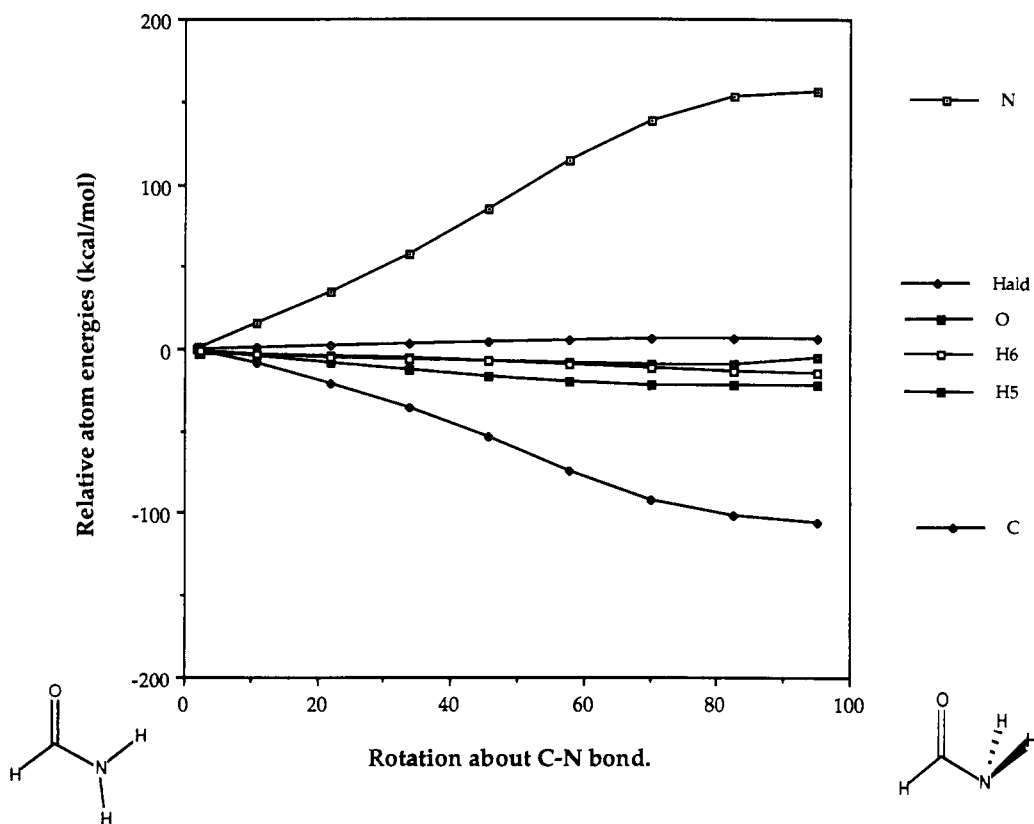


Figure 7. Changes in energy on rotation about the C-N bond.

Within the density analysis method, the bond critical point represents a special point where the electron populations of atoms A and B join. If one then starts at the bond critical point and constructs a set of rays normal to the bond path for which the charge density decreases most rapidly, an interatomic surface can

be defined as shown in Figure 5. A set of surfaces may be constructed, one for each bond, which serve to separate the molecule into a set of atomic domains.

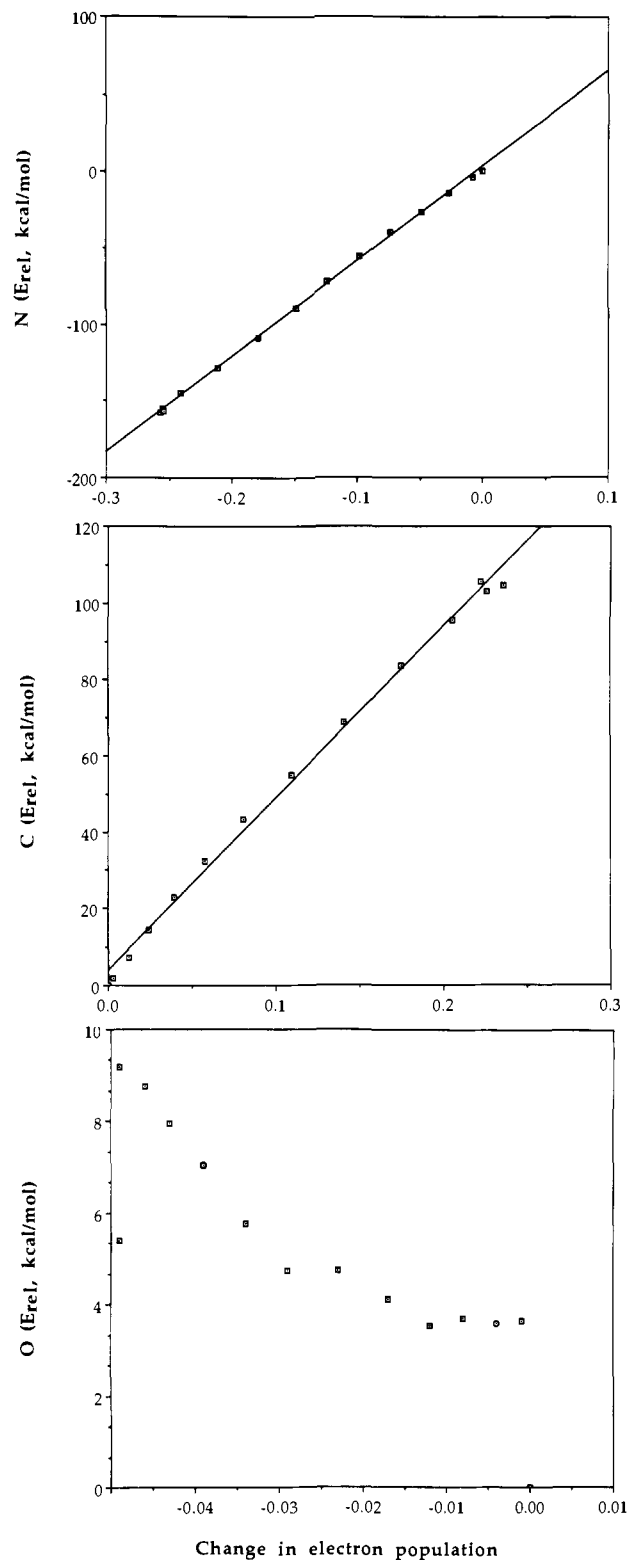
By integrating the appropriate function of the charge density over each atomic domain, the electronic properties of each atom

can be calculated. The most obvious of these properties is the atomic electron population, but the method has the added capability of calculating other properties such as the atomic electronic kinetic energies for each atom in a molecule. It is this capability that allows us to describe the rotational barrier in formamide from an *intramolecular* perspective.

As shown in Figures 6 and 7 the flow of charge and energy between atoms proved to be smooth during the rotation.<sup>19</sup> It is significant that most of the changes involve only the carbonyl carbon and the amide nitrogen. The data in Figure 6 show that as the structure changes from the planar amide form to the rotated structure the carbonyl carbon gains electronic population at the expense of the amide nitrogen. This result is consistent with the earlier work of Wiberg and Laidig<sup>2</sup> and suggests that the barrier to rotation is due largely to the amino group being destabilized near the transition structure. This destabilization is clearly shown in the kinetic energy flow in Figure 7, where there is a smooth destabilization of the amide nitrogen and a smaller stabilization of the carbonyl carbon during the conformational change. This can be interpreted as being indicative of the relative electronegativities of the nitrogen and the carbonyl carbon, since the amount of stability that a particular type of atom can derive from a given amount of electron density provides a fundamental description of electronegativity. Figure 8 shows the relationship between changes in electron population and atomic stability for the amide nitrogen, the carbonyl carbon, and the carbonyl oxygen, respectively. The abscissa of each of these graphs indicates the relative stability of each atom with increasing electron population. The origins are with respect to the planar formamide values. It is significant that the slope of the nitrogen line is 1.38 times greater than that of carbon, while the changes at oxygen are small. These data are also consistent with the view that a carbonyl oxygen is very nearly saturated with electron density, and increasing the electronic population of this kind of atom does not give it extra stability. Recent work on enolate electronic structures supports this view.<sup>23</sup>

We have noted that the nitrogen in the planar form appears more electronegative than that in either saddle point conformer and has a larger electron population. The change in population at nitrogen can be accommodated by a simple hybridization argument. In the planar amino group, the hybridization at nitrogen would be close to  $sp^2$ . However, on rotation, the amino group undergoes pyramidalization, leading to H-N-H bond angles of  $106^\circ$  and bond path angles ( $102^\circ$ ) that are even smaller. Here, the hybridization would correspond to less than 25% s-character, and the nitrogen would be much less electronegative than in the planar form. This would lead to the calculated change in structure and electron population, since in the planar form the greater electronegativity of the  $sp^2$  hybridized nitrogen shortens the C-N bond and allows the nitrogen to withdraw more electron density from the carbonyl carbon. As a check on this hypothesis, we have carried out a geometry optimization for  $90^\circ$  rotated formamide in which the amino group was forced to be planar. This raised the energy by 6.6 kcal/mol above the transition structure (A) and led to electron populations that are between those of the planar form and the pyramidal saddle point form. It would appear that one reason why the planar transition form is not favored is because the nitrogen prefers to place its lone pair electrons in an orbital with high s-character which requires a pyramidal structure.

It is possible to separate the electron populations of the planar form into  $\sigma$  and  $\pi$  components.<sup>2</sup> The  $\pi$  populations are 1.866 ( $NH_2$ ), 0.408 (C), and 1.710 (O). In the absence of any interactions, the populations would have been 2.0, 1.0, and 1.0, respectively. Therefore, the  $\pi$  system charges are  $NH_2$ , +0.134 ( $NH_2$ ), +0.592 (C), and -0.710 (O). This may be compared with rotamer A for which the  $\pi$  populations are 1.990 ( $NH_2$ ), 0.406 (C), and 1.604 (O). There is essentially no change at carbon, but the oxygen gains 0.106  $\pi$  electron in the planar form. At the same time, it loses 0.054  $\sigma$  electron on going from the rotated to planar forms, leaving a net change of only 0.05 electron. All of



**Figure 8.** Relationship between the electron population and the kinetic energy for the C, N, and O atoms of formamide. Note that the energy scale for O is much different than that for C or N.

the relatively large shift in electron population at carbon is found in the  $\sigma$  system as would be expected for an inductive effect.

It is also of interest to look at the changes which occur in atomic sizes during rotation about the C-N bond. Figure 9 shows the distances from each atom of the C-N and C=O bonds to each bond critical point. Examination of this figure reveals that the largest change is in the radius of the carbonyl carbon atom bonding to the amide nitrogen. The nitrogen atom is somewhat affected, but not to as large an extent.

(23) Breneman, C. M. Unpublished work.

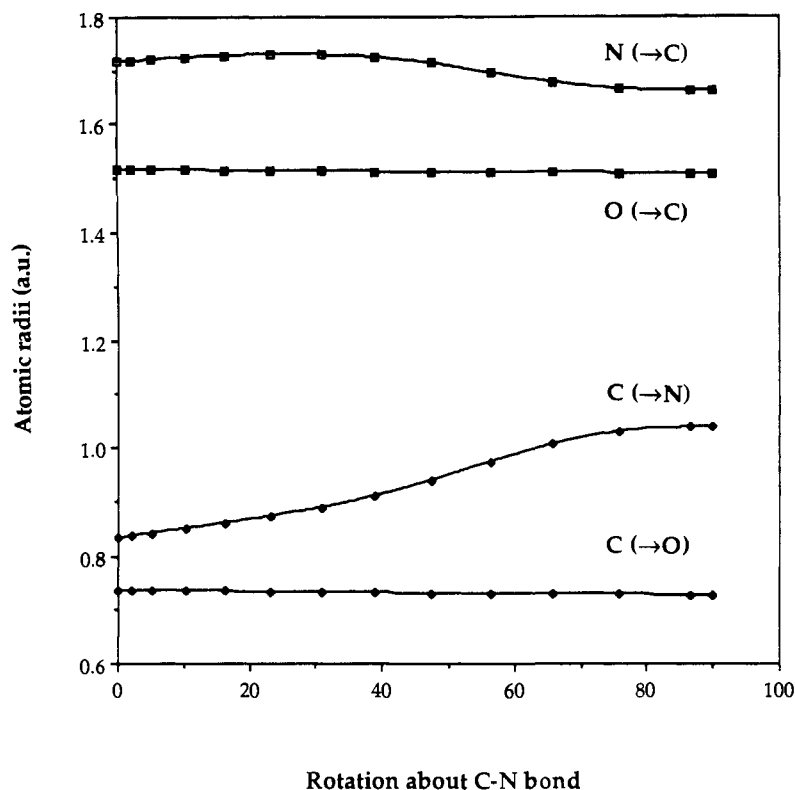


Figure 9. Changes in atomic sizes (distances from the atoms to the bond critical points) on rotation about the C-N bond.

## 6. A View from the Outside

The analysis given above is concerned with the molecule as seen by its nuclei. The population calculations are in good accord with simple hybridization arguments, as is usually found to be the case.<sup>24</sup> However, for intermolecular interactions, this approach may sometimes be inconvenient since the atomic charge, atomic dipoles, and higher terms must be considered. Here, it is helpful to view the molecule from the outside in order to see what an approaching reagent would experience. Two pertinent quantities are readily obtained from the wave functions: the Laplacian of the charge density ( $\nabla^2\rho$ ) and the electrostatic potential. The Laplacian of  $\rho$  has been used to predict regions of higher reactivity,<sup>25</sup> and the electrostatic potential has frequently been used to predict the regiochemistry of intermolecular interactions.<sup>26</sup>

To simplify the analysis, the electrostatic potential field can be used to define effective point charges for spherically symmetrical atoms. We have used a version of the CHELP program<sup>27</sup> (CHELPG), modified to be nearly rotationally invariant, to calculate nuclear-centered effective point charges that reproduce the electrostatic potential field outside the molecular van der Waals radius to a high degree of accuracy.<sup>28</sup> These charges, while not strictly representative of the interior charge distribution of the molecule, do convey a qualitative picture of what the molecule looks like to its surroundings. As indicated in Figure 10, the calculated CHELPG charges show that the amino group externally appears more positive in the planar structure than in the pyramidal transition state, as predicted by the resonance model, while the carbonyl carbon appears to become considerably more electrophilic in the rotated structure, also in accord with experiment. The data

show that the carbonyl oxygen remains relatively unchanged throughout the rotation. In each case, the Mulliken population analysis data are also included on the figures for comparison, even though Mulliken charges are known to be considerably basis-set dependent.<sup>29</sup>

In an effort to visualize the external view of the electrostatic potential and the Laplacian of the charge density ( $\nabla^2\rho$ ), we have calculated these properties at about 30 000 points on the molecular van der Waals surface (defined as the 0.002 e/au<sup>3</sup> surface in  $\rho$ ). The calculated values are represented by a color scale superimposed on the closely-spaced 3-D contours which represent the molecular surface in  $\rho$  (Figure 11). The electrostatic potential values at the VDW surface are in accord with intuition; the lowest values of the electrostatic potential are found in the molecular plane near the carbonyl oxygen in all three conformers, while in the rotated structures A and B a second region of low potential is seen near the nitrogen, where a "lone pair" might be expected to exist. There is a somewhat larger region of negative potential around the carbonyl oxygen in the planar structure than in either of the rotational transition-state structures, as predicted by the resonance model, but this is not reflected in the change in atomic charge at oxygen.

The Laplacian of  $\rho$  is normally examined in terms of its critical points, which lie closer to the nuclei than the VDW surface.<sup>24</sup> Nevertheless, we thought it might be of interest to see if there is any relationship between  $\nabla^2\rho$  at the critical points and at the VDW surface. It can be seen from Figure 11 that the nature of the Laplacian field around the carbonyl oxygen at the van der Waals radius is in conflict with the electrostatic potential; in the region where "lone pairs" might be expected, there are weak areas of greater positive  $\nabla^2\rho$ , implying regions of relative charge depletion, although at much shorter distances from the oxygen nucleus (<0.5 au) there are two negative peaks in the Laplacian field. These observations suggest that the critical points in  $\nabla^2\rho$  may predict the nature of the electrostatic potential at the VDW surface. This question will receive further study.

(24) Cf.: Wiberg, K. B.; Breneman, C. M. *J. Am. Chem. Soc.* **1990**, *112*, 8765.

(25) Carroll, M. T.; Chang, C.; Bader, R. F. W. *Mol. Phys.* **1988**, *63*, 387 and ref 3b.

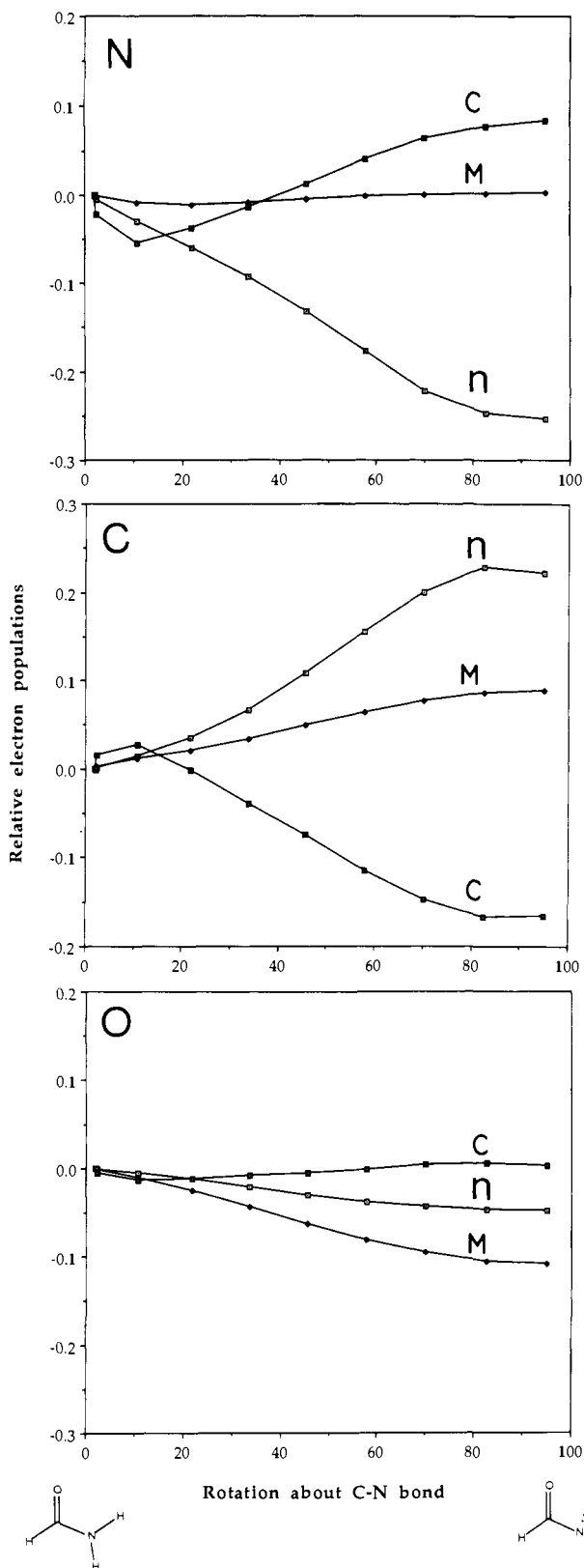
(26) Scrocco, E.; Tomasi, J. *Adv. Quantum Chem.* **1978**, *11*, 115.

(27) Chirlian, L. E.; Francl, M. M. *J. Comput. Chem.* **1987**, *6*, 894. QCPE program No. 524. Our modification consists of selecting points for fitting from a 3-D grid of 5000 points placed around the molecule with a point spacing of 0.3 Å. Points within a 2.0-Å radius from any atom are excluded.

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**Figure 10.** Changes in electron populations caused by rotation about the C-N bond. The lines indicate (n) the changes in electron population as determined by the theory of atoms in molecules, (C) the changes in effective charges determined by fitting the electrostatic potentials, and (M) the changes in the Mulliken populations.

It is important to note the anisotropy of the electrostatic potential near the nitrogen (Figure 11). *The only way in which this anisotropy can be reproduced by charges for spherically symmetrical atoms is to have part of it associated with other atoms such as the hydrogens attached to the nitrogen.* Thus, the charges

derived from the electrostatic potential are effective charges which will reproduce the data, and should not be thought of as correctly representing the charge at a given atom.

## 7. Conclusions

All of the results, including the geometry changes, the changes in force constants, and the changes in electron populations or effective charges, indicate that the oxygen in formamide does not play an active role in determining the rotational barrier and that the principal interactions involve only the carbon and nitrogen. A reasonable hypothesis is that the main determining factor is the need for the nitrogen to stabilize its lone pair electrons as well as possible. In the rotated form, A, the nitrogen places its lone pair electrons in an orbital with as much s-character as possible since the s-electrons are more strongly bound than are p-electrons, and as a result the other bonds are formed with orbitals having higher p-character. This may be seen in the less than tetrahedral observed H-N-H angles which correspond to even smaller bond path angles.<sup>30</sup> The nitrogen will then have relatively low electronegativity since this quantity decreases with increasing p-character. In the planar form, the nitrogen has an opportunity to use its lone pair to interact with the adjacent electron deficient carbon, and this will lead to stabilization. The interaction is seen in the CN  $\pi$  bond order of 0.229 in the planar form. In order to allow this interaction, the lone pair must occupy a p-orbital, which results in approximately 120° bond angles at nitrogen and  $sp^2$  hybridization. The increase in electronegativity of the nitrogen results in a charge shift from the carbon to the nitrogen in the  $\sigma$  system. The shift in charge density is in part due to a shift in the bond critical point toward the carbon leading to a larger volume element for the nitrogen. The role of the oxygen is mainly to polarize the carbonyl group and make the carbon able to interact with the nitrogen.

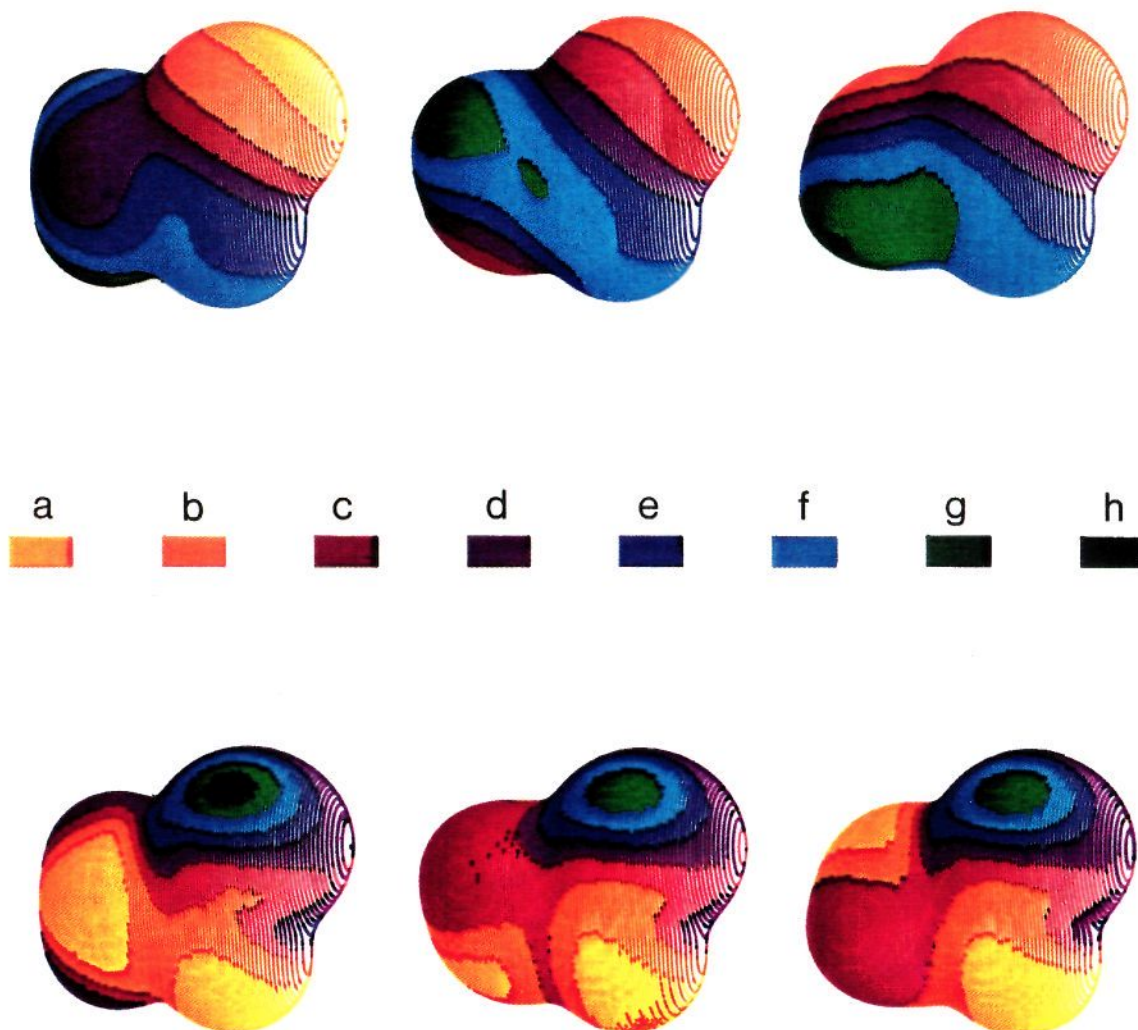
The atoms defined in this way are far from spherical, and the center of charge does not coincide with the nuclear position. As a result, there are atomic dipoles and higher terms, which may readily be calculated along with the populations derived from integration of the charge density. The dipole moment may be reproduced by summing the charge-distance products and adding the atomic dipoles.<sup>31</sup> Similarly, if one also includes the quadrupole terms, it is possible to reproduce the electrostatic potential outward from the van der Waals surface.

It is also possible to reproduce the electrostatic potential using a set of nuclear centered charges, and the values of these effective charges may be derived from the electrostatic potential. The charges defined in this way are for nonphysical spherically symmetrical atoms. They also are in agreement with the conclusion that the oxygen is not significantly involved in determining the barrier to rotation. The direction of charge flow between C and N derived from the effective charges is opposite from that obtained from the electron populations found by integration of the charge density. This is a result of the difference in the way the atoms are defined, and formamide is probably an extreme example of this difference. The electrostatic energy plots (Figure 11) illustrate the difficulty in representing the nitrogen by an effective charge. The electrostatic potential in the vicinity of the nitrogen is markedly anisotropic. *As a result, the effective charges must be balanced among the several atoms in such a way as to reproduce the anisotropy.*

It is important to note that the electrostatic potential outward from the van der Waals surface may be well reproduced either by the terms obtained using the theory of atoms in molecules or by the effective charges derived by fitting the electrostatic potential. The molecular dipole moment may also be reproduced in the same fashion. Thus, there is no fundamental conflict between the two sets of charges, despite the differences seen in Figure 10. The charges designated as "n" are for anisotropic (nonspherical) quantum mechanically defined atoms, and are the

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**Figure 11.** Electrostatic potential maps (upper) at the van der Waals surface ( $0.002 e/B^3$ ) for planar formamide (left), conformer A (center), and conformer B (right). The color code is (kcal/mol) as follows: (a)  $-52$  to  $-38$ ; (b)  $-38$  to  $-24$ ; (c)  $-24$  to  $-10$ ; (d)  $-10$  to  $+4$ ; (e)  $4$  to  $18$ ; (f)  $18$  to  $32$ ; (g)  $32$  to  $46$ ; and (h)  $46$  to  $60$ . Values of the Laplacian of  $\rho$  at the van der Waals surface are given in the lower plot. The color code ( $e/B^3$ ) is as follows: (a)  $0.0065$  to  $0.0077$ ; (b)  $0.0077$  to  $0.0089$ ; (c)  $0.0089$  to  $0.0101$ ; (d)  $0.0101$  to  $0.0113$ ; (e)  $0.0113$  to  $0.0125$ ; (f)  $0.0125$  to  $0.0137$ ; (g)  $0.0137$  to  $0.0149$ ; (h)  $0.0149$  to  $0.0160$ . The molecule is oriented with the oxygen at the upper right, the amine group at the left, and the aldehydic proton to the lower right.

leading terms of a series expansion that includes atomic dipoles, quadrupoles, etc. They correctly represent the charge distribution as seen by the atomic nuclei and reflect changes in hybridization. The effective charges marked "c" are for spherical *nonphysical* atoms and give a simple representation of the electrostatic potential at the VDW surface. As such, they provide a convenient way in which to think of the molecule in terms of intermolecular interactions.

## 8. Calculations

The calculations were carried out using GAUSSIAN-90<sup>32</sup> and standard

basis sets.<sup>33</sup> The electron populations and kinetic energies were calculated using PROAIM.<sup>34</sup> The bond orders were calculated using BONDER.<sup>35</sup>

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**Registry No.** HC(O)NH<sub>2</sub>, 75-12-7.

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