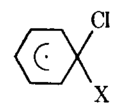
the liquid phase. The somewhat lower yields in ^{38}Cl-for-OCH_3 and ^{38}Cl-for-NH_2 substitution are most likely due to the presence of H atoms at the substituent X, thus giving rise to competing H abstraction.

Substitution of Cl, Br, I, and NO_2, on the other hand, shows a strongly overproportional decrease in yield with increasing *n*-pentane concentration, particularly in the case of the ^{38}Cl-for-Cl exchange. As was demonstrated previously by Berei and Stöcklin[7] by means of iodine scavenger experiments, these substitution processes exhibit considerable contribution from thermal processes, a fact which is particularly true for the ^{38}Cl-for-Cl exchange. This is apparently the case whenever the substitution process is thermoneutral or exothermic (X = Cl, Br, NO_2, I). The yield for these thermal and/or multistep processes decreases in the order of Cl > Br $\simeq$ NO_2 > I; i.e., with the exception of NO_2, the yields decrease with decreasing C–X bond energy of the leaving group X. Exothermic and thermoneutral homolytic replacement of substituents such as I, Br, Cl, NO_2 by Cl atoms is well known,[25,26] and it is assumed that these replacement reactions proceed via σ-complex formation:[27]

$\cdot Cl + C_6H_5X \longrightarrow$  $\longrightarrow C_6H_5Cl + X\cdot$ (7)

The fact that yields decrease with decreasing C–X bond energy does not, however, follow from reaction 7, in which case the reverse effect would be expected. An explanation can be provided by assuming a major competition from X abstraction

$$\cdot Cl + C_6H_5X \rightarrow C_6H_5 \cdots X \cdots Cl \rightarrow XCl + C_6H_5\cdot \quad (8)$$

which might overcompensate the replacement reaction when going from Cl to Br to I substituents. This is also reflected by the concomitant increase in corresponding inorganic yields (cf. Table III).

That the chlorine atom preferentially attacks the region of the maximum electron density distribution, viz. the halogen substituent, is also reflected by the relatively high yield of Cl-for-F substitution as compared to Cl-for-H substitution in fluorobenzene. It can be seen from Figure 4 that the one-step Cl-for-H substitution yield is almost identical with the Cl-for-F substitution yield, despite the fact that there are 5 times as many H atoms available per molecule as F-atoms. This result cannot possibly be explained by the small difference between the bond energies.

References and Notes

(1) G. Stöcklin, "Chemie Heisser Atome", Verlag Chemie, Weinheim, 1969.
(2) D. Urch, "International Review of Science. Series 1. Inorganic Chemistry", Vol. 8, p 149, Butterworths, London, 1972.
(3) H. J. Ache, W. Herr, and A. Thiemann in "Chemical Effects of Nuclear Transformations", Vol. II, IAEA, Vienna, 1961, p 111.
(4) J. E. Willard in "Chemical Effects of Nuclear Transformations", Vol. I, IAEA, Vienna, 1961, p 215.
(5) G. Stöcklin and W. Tornau, *Radiochim. Acta*, **6**, 86 (1966).
(6) G. Stöcklin and W. Tornau, *Radiochim. Acta*, **9**, 95 (1968).
(7) K. Berei and G. Stöcklin, *Radiochim. Acta*, **15**, 39 (1971).
(8) L. Vasáros, H.-J. Machulla, and W. Tornau, *J. Chromatogr.*, **62**, 458 (1971).
(9) E. J. Knust and M. Schüller, *J. Chromatogr.*, **114**, 207 (1975).
(10) A. E. Richardson and R. Wolfgang, *J. Am. Chem. Soc.*, **92**, 3480 (1970).
(11) H.-J. Machulla and G. Stöcklin, *J. Phys. Chem.*, **78**, 658 (1974).
(12) S. S. Kontis and D. S. Urch, *Radiochim. Acta*, **15**, 21 (1971).
(13) F. A. Trotman-Dickenson in "Advances in Free Radical Chemistry", Vol. 1, G. H. Williams, Ed., Logos, London, 1965.
(14) E. N. Avdonina, *Radiokhimiya*, **15**, 720 (1973).
(15) Cf., e.g., E. C. Kooyman in ref 13.
(16) H. L. Allred, *J. Inorg. Nucl. Chem.*, **17**, 215 (1961).
(17) P. Spagnolo, L. Testaferri, and M. Tiecco, *J. Chem. Soc. B*, 2006 (1971).
(18) D. H. McDaniel and H. C. Brown, *J. Org. Chem.*, **23**, 420 (1958).
(19) D. F. DeTar, 17th Organic Symposium, June 1961.
(20) R. E. Bühler and M. Ebert, *Nature (London)*, **214**, 1220 (1967).
(21) G. A. Russel, *J. Am. Chem. Soc.*, **80**, 4987, 4997, 5002 (1958).
(22) G. A. Russel, *Tetrahedron*, **8**, 101 (1960).
(23) R. M. White and F. S. Rowland, *J. Am. Chem. Soc.*, **82**, 4713 (1960).
(24) N. V. Pozdeev, A. N. Nesmeyanov, and B. G. Dzantiev, *Radiokhimiya*, **5**, 395 (1963).
(25) B. Miller and C. Walling, *J. Am. Chem. Soc.*, **79**, 4187 (1957).
(26) B. Milligan, R. L. Bradow, J. E. Rose, H. E. Hubbert, and A. Roe, *J. Am. Chem. Soc.*, **84**, 158 (1962).
(27) M. L. Poutsma in "Methods in Free-Radical Chemistry", Vol. 1, E. S. Huyser, Ed., Marcel Decker, New York, N.Y., 1969.

Heat Capacities of Ureas and Water in Water and Dimethylformamide

O. D. Bonner,* Jana M. Bednarek, and Ruth K. Arisman

Contribution from the Department of Chemistry, University of South Carolina, Columbia, South Carolina 29208.

Received June 11, 1976

Abstract: Apparent molal heat capacities are reported for eight ureas and substituted ureas and for water in *N,N*-dimethylformamide as a solvent. Data for four substituted ureas are reported in water as a solvent. Excess apparent molal heat capacities are calculated and group contributions to the excess heat capacities are calculated in aqueous solutions. It is found that the relative hydrogen bonding strengths of protons to the peptide carbonyl oxygen are NH_2 > OH_2 > NH. Denaturation of proteins is postulated to be caused either by interaction of the urea NH_2 protons with the peptide carbonyl or by a hydrophobic interaction when alkyl-substituted ureas are most effective.

Although aqueous solutions of urea, substituted ureas, and guanidinum salts are found to be effective denaturants of proteins, the mechanism of the denaturation process is incompletely understood. On the one hand there is evidence for direct "binding" of denaturants to proteins.[1,2] Alternatively, it has been proposed that changing of the water structure by the denaturant plays a role in the denaturation process. Unfortunately, in spite of the many data that are too numerous to reference there is no unanimity of opinion as to the exact effect of urea, for example, on the solvent structure.[3] Perhaps a portion of the problem is in the semantics involved when one speaks of "structure making" or "structure breaking" solutes. Heat capacity measurements have proven useful in elucidating solute–solvent interactions,[4–8] but one limitation in the cal-

Table I. Specific Heat and Apparent Molal Heat Capacities in *N,N*-Dimethylformamide (cal deg^{-1} mol^{-1})

		m							
		0	0.358	0.5	1.0	2.0	3.0	4.0	15.1
Urea	Sp ht	0.4755		0.4830	0.4898	0.5015			
	ϕC_p	44.3		44.0	43.7	43.1			
Thiourea	Sp ht	0.4755		0.4786	0.4810	0.4845	0.4869	0.4885	
	ϕC_p	43.1		42.6	42.1	41.4	40.9	40.4	
Acetamide	Sp ht	0.4755		0.4792	0.4828	0.4893	0.4953	0.5002	
	ϕC_p	35.8		35.8	35.8	35.8	35.8	35.7	
1,1-Dimethylurea	Sp ht	0.4755	0.4770						
	ϕC_p		46.1						
1,3-Dimethylurea	Sp ht	0.4755		0.4760	0.4765	0.4775	0.4785	0.4796	
	ϕC_p	43.0		43.0	43.0	43.1	43.2	43.3	
1,3-Dimethylthiourea	Sp ht	0.4755		0.4740	0.4728	0.4711	0.4702	0.4696	
	ϕC_p	46.3		46.4	46.5	46.8	47.2	47.4	
Tetramethylurea	Sp ht	0.4755		0.4748	0.4742	0.4732	0.4724	0.4714	
	ϕC_p	53.8		53.8	53.8	53.8	53.8	53.7	
Tetramethylthiourea	Sp ht	0.4755		0.4725	0.4700	0.4657	0.4619		
	ϕC_p	56.4		56.4	56.6	56.6	56.5		
Water	Sp ht	0.4755		0.4818	0.4880	0.5003	0.5120	0.5233	0.6282
	ϕC_p	21.3		21.2	21.4	21.4	21.4	21.4	21.4

culation of "excess heat capacities" of solid solutes has been an estimation of their "intrinsic heat capacity" in solutions. The heat capacity data for solutes in *N,N*-dimethylformamide and water which we are reporting were undertaken to obtain a comprehensive set of data from which one might better estimate the extent of the interaction of denaturants with both proteins and water.

Experimental Section

Apparatus. The heat capacity measurements were made using a calorimeter which has been described in detail.[8] The apparatus is capable of detecting temperature changes of 5×10^{-5} K and the average values of specific heats are probably in error by less than 0.1%. The experiments were performed in the temperature range 297.6–298.6 K.

Solvent. *N,N*-Dimethylformamide was obtained as the Certified ACS reagent grade with the principle impurity being water. The solvent was dried over 5-Å molecular sieves for a minimum of 3 days prior to use. Infrared spectra indicated that this treatment effectively removed the water.

Solutes. Urea, thiourea, and acetamide were obtained either as the Fisher Certified ACS reagent grade or Baker analyzed reagent grade. The samples of 1,1-dimethylurea, 1,3-dimethylurea, and tetramethylthiourea were somewhat less pure and these compounds were recrystallized from chloroform. All solid solutes were vacuum-dried at 45 °C for 24 h before using. Tetramethylurea was obtained from Aldrich Chemical Co. and was vacuum-distilled and dried over molecular sieves before using.

Concentrations. Solutions were prepared by weight and concentrations are accurate to at least 0.1%. The data are interpolated to rounded concentrations in Tables I and III except in the case of slightly soluble solutes. Values at infinite dilutions were obtained by least-squares linear extrapolation of data below 1.0 *m*. Insufficient data were available for extrapolaties to infinite dilution of 1,1-dimethylurea in *N,N*-dimethylformamide and for tetramethylthiourea in water. The value of ϕC_p° for these compounds was assumed to be that obtained in the dilute solution in each instance.

Results and Discussion

Denaturant Interactions. The experiments reported in this paper were designed to separate, if possible, the effects of denaturants on protein-water systems. One possible role of the denaturant is direct interaction with the protein and this process is sometimes called "binding" of the denaturant.[1,2] It has been noted that one structural requirement[9] for denaturing effectiveness is the presence of an NH or NH_2 functional group. This suggests that the interaction might be hydrogen bonding between the proton of the denaturant and the carbonyl oxygen of the peptide group.

Table II. Excess Apparent Molal Heat Capacities at Infinite Dilution in *N,N*-Dimethylformamide (cal deg^{-1} mol^{-1})

	ϕC_p° (exptl)	C_p (intrinsic)[a]	ϕC_p° (excess)
Urea	44.3	26.7	17.6
Thiourea	43.1	26.7	16.4
Acetamide	35.8	28.8	7.0
1,1-Dimethylurea	46.1	40.0	6.1
1,3-Dimethylurea	43.0	40.0	3.0
1,3-Dimethylthiourea	46.3	40.0	6.3
Tetramethylurea	53.8	53.8	0.0
Tetramethylthiourea	56.4	53.8	2.6
Water	21.3	17.2	4.1

[a] Assumed values based upon data of Figure 1.

Another possible role of the denaturant is that of altering the structure of the solvent. It is reasonably well established from various types of data that alkyl groups "enhance" the water structure about them. A few examples of different types of measurements which support this phenomenon are heat capacity,[5,10] viscosity,[11] colligative properties,[12,13] and proton magnetic resonance.[14] At least one estimate has been made of this contribution to the excess heat capacity of alkyl-substituted ureas in aqueous solution. Two other more direct interactions are possible, however, between the various ureas and water. These are those between the water protons and the urea carbonyl group and between the urea protons and the lone electron pairs on the water oxygen atom. An attempt to separate all of these interactions will be made in the remaining discussion.

Intrinsic Heat Capacities. It has been sufficiently emphasized[5,7,8] that heat capacity measurements are a powerful tool in elucidating solvent structural changes and solute-solvent interactions. The principal difficulty involved in making precise calculations is in obtaining an accurate estimate of the intrinsic heat capacity of solutes. It is recognized that the properties of a solute in solution most nearly resemble those of a supercooled liquid. It is also recognized that heat capacities of substances are different in the solid and liquid phases. A few examples[15] show this difference to be quite appreciable for many substances and to be greater than a factor of 2 for water. This observation casts some doubts on the accuracy of excess heat capacities, which have been reported for solid solutes when the

Table III. Specific Heat and Apparent Molal Heat Capacities in Aqueous Solutions (cal deg^{-1})

	Thiourea		1,3-DMTU[a]		TMTU[b]		Acetamide[c]	
m	Sp ht	ϕC_p	Sp ht	ϕC_p	Sp ht	ϕC_p	Sp ht	ϕC_p
0.0	0.9983	9.3	0.9983	54.5	0.9983		0.9983	38.0
0.2295					0.9880	85.8		
0.5	0.9676	12.2	0.9750	55.6			0.9881	38.0
1.0	0.9417	15.1	0.9555	56.7			0.9785	38.0
1.5	0.9205	18.2						
2.0			0.9216	57.6			0.9610	38.1
3.0			0.8934	58.1			0.9450	38.1
3.5			0.8815	58.4				
4.0							0.9310	38.2

[a] 1,3-Dimethylthiourea. [b] Tetramethylthiourea. [c] These data may be compared with those of ref 21, which was unpublished at the time this work was performed.

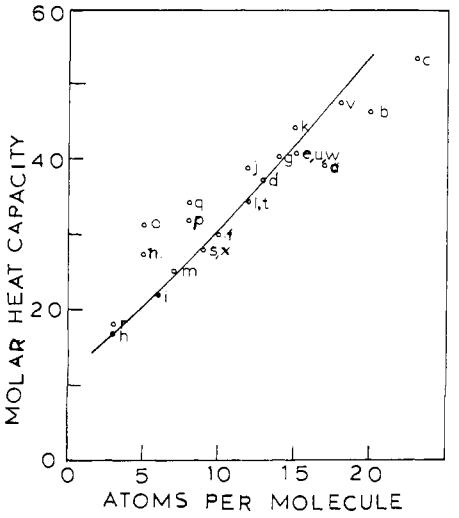


Figure 1. Molal heat capacities of some liquids (cal mol^{-1} K^{-1}): a, 2-methylbutane; b, *n*-hexane; c, *n*-heptane; d, methyl ethyl ketone (from this work); e, diethyl ether; f, acetone; g, ethyl acetate; h, HCN; i, hydrazine; j, unsym dimethylhydrazine; k, trimethylhydrazine; l, *N,N*-dimethylformamide (from this work); m, nitromethane; n, chloroform; o, carbon tetrachloride; p, 1,2 dibromoethane; q, trichloroethane; r, carbon disulfide; s, dimethyl sulfide; t, methyl diethyl sulfide; u, methyl propyl sulfide; v, methyl butyl sulfide; w, diethyl sulfide; x, ethyl mercaptan.

heat capacity of the pure *solid* is used as the intrinsic heat capacity.

One method of estimating the intrinsic heat capacity of a solute (as it exists in solution) is to note the values which have been reported for liquid substances at 25 °C. A rather extensive list of these is readily available[16] and C_p values have been plotted as a function of the number of atoms per molecule in Figure 1. None of these liquids are hydrogen bonded and the heat capacities will represent primarily London and dipole forces. It will be noted that with the exception of the hydrocarbons which fall below the line and the halogenated hydrocarbons which lie above the line, the heat capacities of the remainder of the compounds of C, H, O, S, and N vary almost linearly with the atomicity. The average deviation from a smooth curve is less than ±1 cal mol^{-1} deg^{-1}. An additional check on the validity of using these approximations is the agreement between the heat capacity values obtained for these solutes in solutions where the solute-solvent and solvent-solvent interactions are similar and the values for the pure liquids. This method of estimation, although consistent, is admittedly not exact. Nevertheless, large deviations of the heat capacities of the solutes of this paper from such calculated "intrinsic" values should represent interaction with or alteration of the solvent.

Denaturant–Peptide Interactions. The experimentally determined specific heat data and calculated apparent heat capacities, ϕC_p, in *N,N*-dimethylformamide as solvent, appear in Table I. This solvent is convenient as a model containing the protein peptide linkage since (1) it is a liquid and (2) there is no proton available to interact with the denaturant carbonyl group. Excess apparent molal heat capacities, $\phi C_p^\circ - C_p$ (intrinsic), are calculated in Table II for the solutes in the amide solvent. It would be expected that the tetramethyl compounds would exhibit an apparent heat capacity in solution equal to their intrinsic heat capacity, so it is probable that the calculation of an excess heat capacity for tetramethylthiourea means that too low a value was assumed for its intrinsic heat capacity. The fact that C_p (excess) is larger for 1,3-dimethythiourea than for 1,3-dimethylurea indicates that possibly a slightly larger intrinsic value should be assumed for all of the thio compounds. This adjustment to the values obtained from using as standards the compounds in Figure 1 may be rationalized by observing that CS_2, which contains the C=S group, has a heat capacity which places it above the smooth curve, while the other thio compounds fall almost directly on it. It is felt, however, that a more consistent view is obtained by using the "average" intrinsic heat capacity values which appear in Tables II and IV without making any arbitrary adjustments. The largest excess heat capacities are for these compounds containing an NH_2 group and the order is urea > thiourea > acetamide > 1,1-dimethylurea. This is the expected order when one considers the electron donating character of the methyl group. The values of C_p (excess) are smaller for those compounds containing NH groups and the value for water lies between that of the NH_2 compounds and the NH compounds.

Denaturant–Water Interactions. The denaturant-water systems are much more complicated because of the large number of possible interactions. The NH and NH_2 groups of the denaturants may hydrogen bond to the water; the water protons may hydrogen bond to the carbonyl group of the denaturant; there is ample evidence[5,7,10-14] that alkyl groups "enhance" the water structure; and finally in the case of the thioureas there is the possibility of the C=S group interacting with the solvent in some fashion. We have attempted to sort out the various effects by comparing the apparent heat capacities of seven ureas and thioureas and acetamide in aqueous solutions. This, in effect, permits one to evaluate the individual contributions of the five functional groups. An exact agreement between experimental heat capacities and those calculated from group contributions would not be expected, since the different neighboring groups in each compound will alter to some extent the interaction of a given group with the solvent. Hopefully, however, a qualitative answer should emerge. It may be noted that all of the oxygen-containing compounds have the -C(=O)N< arrangement, which is unique to the

Table IV. Excess Molal Heat Capacities at Infinite Dilution in Aqueous Solutions (cal deg^{-1} mol^{-1})

Solute	ϕC_p°	C_p (intrinsic)[c]	ϕC_p° (excess) Exptl	Calcd[a]
Urea	20.9[b]	26.7	−5.8	−5.8
Thiourea	9.3	26.7	−17.4	−17.2
Acetamide	38.0	28.8	9.2	9.2
1,1-Dimethylurea	59.2[b]	40.0	19.2	19.3
1,3-Dimethylurea	65.6[b]	40.0	25.6	25.6
1,3-Dimethylthiourea	54.5	40.0	14.5	14.2
Tetramethylurea	103.9[b]	53.8	50.1	44.2
Tetramethylthiourea	85.5	53.8	32.0	32.8
Water	18.0	17.2	0.8	

[a] $[\phi C_p - \phi C_p$ (intrinsic)]. [b] Data from ref 7. [c] Assumed values based upon data of Figure 1.

Table V. Group Contributions to Excess Molal Heat Capacities at Infinite Dilution in Aqueous Solutions

Group	ϕC_p° (excess), cal deg^{-1} mol^{-1}
C=O	4.2
NH_2	−5.0
C=S	−7.2
NH	1.5
CH_3	10.0

ureas and peptides, and it is felt that the carbonyl groups of esters, ketones, etc. and the $-NH_2$ or >NH groups of amines cannot be meaningfully compared with those of these compounds because of the absence of resonance possibilities such as $-C(-O^-)=N^+<$.

The experimentally measured specific heat data and the calculated apparent heat capacities are given in Table III. Excess molal heat capacities are tabulated in Table IV. One may observe from an inspection of the data that the apparent molal heat capacities of urea and thiourea are less than the intrinsic values. This reminds one of the behavior of the simple electrolytes and in the case of thiourea the value of C_p (excess) of −17.4 cal deg^{-1} mol^{-1} is approaching[8] that of LiCl or NaCl. This observation appears to substantiate the "structure breaking" effect of urea and thiourea in that the solutions of these substances contain fewer or weaker hydrogen bonds than does pure water. Group contributions to excess heat capacities which appear in Table V were calculated in the following manner. Urea, 1,1-dimethylurea, and acetamide were used to evaluate the $-CH_3$, C=O, and NH group contributions. An average value for C=O minus C=S contributions was obtained from the differences in excess heat capacities of urea and thiourea and of the 1,3-dimethyl-substituted compounds. The average value of the NH contribution was obtained by the subtractions of the known group contributions from the total excess heat capacities of 1,3-dimethylurea and 1,3-dimethylthiourea. These data indicate that both the NH_2 and C=S groups are structure breaking. The peptide carbonyl group, which is similar to the urea group, is a strong hydrogen bond acceptor[17] and it is not surprising that it would make a positive contribution to the heat capacity of the system. The positive contribution of the methyl group is, of course, expected.[5,10-14]

The negative contribution of the NH_2 group to the heat capacity of the system is perhaps the most surprising result which was obtained. The data in dimethylformamide as a solvent indicated that this group bonds more strongly than the water protons to the amide carbonyl group and one would expect the same order of bonding strengths to the water oxygen electron pairs and thus a positive contribution. One must consider though the short-range order in pure water, in which many water molecules are hydrogen bonded in a tetrahedral arrangement. It may be possible that the strength of the $NH_2\cdots OH_2$ bonds causes a reorientation of the water molecules in the vicinity of the urea molecule, which results in *fewer* $OH_2\cdots OH_2$ hydrogen bonds; thus a net negative contribution to the heat capacity of the system.

Summary

The question which one might ask at this point concerns the manner in which the interactions which have been elucidated are related to the denaturation process. These data substantiate the previous observations that urea (and also thiourea) are solvent structure breakers. The magnitude of this effect is less, however, than nondenaturing salts such as lithium or sodium chloride. The alkyl-substituted ureas are, in fact, structure makers. The effect of the ureas on the solvent appears not to be the most important aspect of denaturation.

These preliminary data involving the ureas and dimethylformamide indicate that the NH_2 groups of ureas interact more strongly with the peptide carbonyl group than do the protons of water. The interaction of the NH groups is, on the other hand, weaker than that of the NH_2 protons and certainly no stronger than that of the OH_2 protons. This observation is in agreement with the many studies of denaturant "binding" by proteins. It also points out the erroneous conclusion which has been drawn from the often quoted work of Klotz and Franzen,[18] who showed that water–carbonyl bonding in an aqueous solution of *N*-methylacetamide was considerably stronger than the interpeptide bonds. All data are, on the other hand, consistent with the relative hydrogen bonding strengths to a peptide carbonyl group in the order urea > water > peptide NH. The importance of this point is such that it should be verified by other types of direct measurements.

It appears from an inspection of all of the pertinent data that the denaturation process may be of two types. For those proteins for which urea is a more effective denaturant than the alkyl-substituted ureas,[9] the most important effect is the "binding" of the urea[1,2] by an interaction between the protein carbonyl group and the urea NH_2 group. In other instances where the alkyl-substituted ureas are the more effective denaturants[19,20] the most important interaction is between the protein and the hydrophobic alkyl groups. Further evidence for the importance of the hydrophobic effect is the denaturation of these same proteins by alcohols and glycols of appreciable hydrocarbon content.[19]

Acknowledgment. This work was supported by PHS Grant No. 1 R01 19980 from the National Institute of General Medicine.

References and Notes

(1) H. B. Bull and K. Breese, *Arch. Biochem. Biophys.*, **139**, 93 (1970).

(2) E. P. K. Hade and C. Tanford, *J. Am. Chem. Soc.*, **89**, 5034 (1967).
(3) These references offer a critical evaluation of many of the pertinent data: (a) F. Franks, "Hydrogen Bonded Solvent Systems", A. K. Covington and P. Jones, Ed., Taylor and Francis, London, 1968, p 42; (b) A. Holtzer and M. F. Emerson, *J. Phys. Chem.*, **73**, 26 (1969).
(4) H. Ruterjans, F. Schreiner, V. Sage, and Th. Ackermann, *J. Phys. Chem.*, **73**, 986 (1969).
(5) T. S. Sarma and J. C. Ahluwalia, *Trans. Faraday Soc.*, **67**, 2528 (1971).
(6) P. Picker, P. A. Leduc, P. R. Philip, and J. E. Desnoyers, *J. Chem. Thermodyn.*, **3**, 631 (1971).
(7) P. R. Philip, G. Perron, and J. E. Desnoyers, *Can. J. Chem.*, **52**, 1709 (1974).
(8) O. D. Bonner and P. J. Cerutti, *J. Chem. Thermodyn.*, **8**, 105 (1976).
(9) J. A. Gordon and W. P. Jencks, *Biochemistry*, **2**, 47 (1963).
(10) P. R. Philip and J. E. Desnoyers, *J. Solution Chem.*, **1**, 353 (1972).
(11) R. L. Kay, T. Vituccio, C. Zawoyski, and D. F. Evans, *J. Phys. Chem.*, **70**, 2336 (1966).
(12) G. E. Boyd, A. Schwarz, and S. Lindenbaum, *J. Phys. Chem.*, **70**, 821 (1966).
(13) W. Y. Wen, S. Saito, and C. Lee, *J. Phys. Chem.*, **70**, 1244 (1966).
(14) M. Marciacq-Rousselot, A. de Trobriand, and M. Lucas, *J. Phys. Chem.*, **76**, 1455 (1972).
(15) G. N. Lewis and M. Randall, "Thermodynamics", 2nd ed, revised by K. S. Pitzer and L. Brewer, McGraw-Hill, New York, N.Y., 1961, pp 63, 72.
(16) R. Shaw, *J. Chem. Eng. Data*, **14**, 461 (1969).
(17) O. D. Bonner and Y. S. Choi, *Spectrochim. Acta Part A*, **31**, 1975 (1975).
(18) I. M. Klotz and J. S. Franzen, *J. Am. Chem. Soc.*, **84**, 3161 (1962).
(19) T. T. Herskovits, B. Gadegbakeu, and H. Jaillet, *J. Biol. Chem.*, **245**, 2588 (1970).
(20) T. T. Herskovits, H. Jaillet, and B. Gadegbakeu, *J. Biol. Chem.*, **245**, 4544 (1970).
(21) O. Kujohara, G. Perron, and J. E. Desnoyers, *Can. J. Chem.*, **53**, 3263 (1975).

Atom/Bond Analysis of Conformational Properties of Molecules (PCILO–CNDO)

Olivia de la Luz Rojas[1]

Contribution from the Institut de Biologie Physico-Chimique, Laboratoire de Biochimie Théorique, associé au C.N.R.S., 75005 Paris, France; the Laboratoire de Physique Quantique, Université Paul Sabatier, 31077 Toulouse Cedex, France; and the Department of Biochemistry, Centro de Investigación y de Estudios Avanzados del I.P.N., Apartado Postal 14-740, Mexico 14, D.F. *Received July 28, 1975*

Abstract: A methodology to analyze the conformational properties of molecular systems in terms of local energy contributions is presented. The energy partitioning proposed here is based on a theoretical additive expansion in terms of one-bond, two-bond, three-bond, etc., contributions. The additive structure of the energy is built using fully localized bond MO's and a perturbation expansion. This methodology has been applied to the study of internal rotation barriers of the following molecular series: CH_3XH_n, $CH_3XH_{n-1}CH_3$ (X = C,N,O), and $X_3B{\cdot}NY_3$ (X,Y = H,F). The energy analysis is made in terms of one-, two-, and three-body contributions at the zeroth-, second-, and third-order perturbative corrections within the PCILO–CNDO scheme. The role of vicinal, geminal, and long-range interactions (third-neighbor bonds) is considered, the transferability of similar contributions from one series to the other, and the effects of substitutions. Our method confirms directly the important role of the delocalization tails, but also the nonnegligible role of the zeroth-order repulsive effects and intramolecular van der Waals dispersion energies.

The conformational properties and especially the internal rotation barriers represent a very good test of the accuracy of quantum chemical methods, since they concern changes of a few kilocalories per mole in a total energy of 10^5 to 10^7 kcal/mol. A theoretical approach such as the Hartree–Fock approximation provides the accurate values for the internal rotation barriers in numerous molecules;[2] the barrier is calculated by subtracting the total energies evaluated for each appropriate conformation of the molecule, but this procedure itself provides no information about the origin of the barrier.

The explanation of the sources of such barriers is a very interesting problem. In the past many different theoretical approaches have been carried out. Among them, Lowe[3] qualitatively explained the barriers about single bonds, comparing the results obtained with the following type of approaches: i.e., (1) using a decomposition of the total energy change with rotation into nonlocal physical components,[4] (2) using delocalized or canonical molecular orbitals, and (3) using localized MO's.

Approach 1 presents the following serious defects: (a) the qualitative description of a barrier (as repulsive or attractive dominant) can change depending on whether the calculation is carried out in the rigid-rotor or the geometry-optimized approximation,[5,6] and (b) for two calculations based on identical geometries, the use of different basis sets can change a calculated barrier from attractive to repulsive dominant.[4-7] Moreover, this approach does not allow a direct relation with the local geometry changes of the nuclear skeleton. With respect to approach 2, the delocalized nature of the canonical Hartree–Fock orbitals does not allow us to analyze the phenomena explicitly in terms of bonds or atomic interactions. Hence, we think that a local analysis in terms of local contributions is more promising; it may refer to atoms or to bonds.

The expression of the Hartree–Fock energy in the LCAO approximation allows a partition in one-, two-, three-, and four-atom terms; it is reduced to one- and two-atom contributions in the CNDO approximation.[8] Section IA briefly recalls this partition and illustrates its limitations in the analysis of a few energy conformational changes.[9-11] The important contributions to the energy changes sometimes concern monatomic or diatomic contributions between atoms whose relative positions are unchanged during the conformational change. Such contributions may come from some changes in the density matrix which are not explained.

In section IB, one considers the analysis of SCF results, referring to bonds (use of bond-like SCF localized MO's[12]). This methodology gives very interesting information about the local origin of the barriers, showing the dominant role of the tails of the SCF–MO's on vicinal groups, as well as the important role of the monoelectronic part of the CNDO (or INDO)