

## Effect of Lithium Ion on the Torsional Barrier in Amides

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*Summary*  $^1\text{H}$  N.m.r. studies show that the activation energy for internal rotation in  $[\text{}^2\text{H}]$ -*NN*-dimethylformamide is increased by  $3.4 \text{ kcal mol}^{-1}$  on complexation with  $\text{Li}^+$ .

THE nature of the binding of lithium cation to the peptide bond is of paramount significance in understanding the conformational changes in polypeptides and proteins brought about by lithium salts<sup>1,2</sup> and also possibly the therapeutic properties of these salts.<sup>3</sup> Specific binding of

$\text{Li}^+$  to amides has been examined in some detail by vibrational and electronic spectroscopy.<sup>4,5</sup> These optical spectroscopic studies, as well as molecular orbital calculations,<sup>2,6,7</sup> indicate that  $\text{Li}^+$  binds preferentially to the carbonyl oxygen of the amide, causing a decrease in the C=O bond order and increase in the C-N bond order. An important consequence of such bond order changes should be an increase in the barrier height to rotation about the C-N bond of amides. However, no satisfactory experimental evidence for a change in barrier height,  $E_a$ , due to inter-

action with  $\text{Li}^+$  is available in the literature. Thus, some workers<sup>8</sup> have reported only coalescence temperatures,  $T_c$ , in n.m.r. spectra or free energy changes at  $T_c$ . The recent n.m.r. study<sup>7</sup> of the barrier height in the *NN*-dimethylformamide (DMF) + LiCl system unfortunately suffers from the fact that the  $E_a$  value found by the authors for the parent amide itself is far from the correct value. Furthermore, these measurements were made on DMF rather than  $[\text{2H}]\text{DMF}$ ; presence of appreciable spin-coupling between the formyl proton and the Me protons<sup>9</sup> in DMF may vitiate the determination of  $E_a$ . In view of the importance of having reliable barrier heights to understand the nature of binding of  $\text{Li}^+$  to the peptide bond, we have carried out a careful determination of barrier heights in  $[\text{2H}]\text{DMF}$  and in  $[\text{2H}]\text{DMF} + \text{LiCl}$ , based on total line shape analysis of the n.m.r. spectra.

$[\text{2H}]\text{DMF}$  was synthesized by the procedure of Neumann and Jonas.<sup>9</sup> The effect of  $\text{Li}^+$  was examined by adding sufficient LiCl to neat  $[\text{2H}]\text{DMF}$  to make a 4M solution and warming to facilitate solvation. At this composition, the 1:4  $\text{Li}^+$ -amide complex<sup>4</sup> would be preponderant in solution. N.m.r. spectra were recorded using a Varian A-60 spectrometer fitted with a V-6040 variable temperature unit. Homogeneity was maximized at each temperature with respect to an internal dioxan reference. A sweep rate of 0.25 Hz  $\text{s}^{-1}$  and a sweep width of 50 Hz were used. Temperatures, determined with an ethylene glycol standard and the calibration equation of Van Geet,<sup>10</sup> are estimated to be accurate to  $< \pm 1^\circ\text{C}$ . Line shape analysis was carried out using the complete Gutowsky-Holm equations,<sup>11</sup> modified to account for unequal  $T_2$ 's.<sup>12</sup> The analysis consisted of digitizing the experimental spectra (ca. 40 points/Hz), curve-fitting employing the iterative program of Knott<sup>13</sup> (MLAB) on a PDP-10 computer, and finally making a visual comparison of experimental and theoretical line shapes on a visual display unit.† Arrhenius plots were also prepared by the MLAB program.

† In the curve-fitting process the values of  $T_2$  in the absence of exchange were taken to be 0.40 and 0.42 s, as found for the two methyl lines at 39 °C. The values for  $\Delta\nu$ , the chemical shift difference, were taken from the relation  $\Delta\nu = 10.26 - 0.0124 t$ , which represents a linear extrapolation of the values found at 39–90 °C, where exchange is negligible. We also carried out fits for  $[\text{2H}]\text{DMF}$  itself with  $\Delta\nu$  determined by the fitting programs at each temperature. [The reliability of these approaches has been examined for other amide systems (ref. 17)]. Although the values found for  $\tau$  with the latter method differed somewhat from those given here, the value of  $E_a$  was virtually unchanged.

‡ 1 kcal = 4.18 kJ.

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TABLE. Activation parameters for C-N bond rotation.<sup>a</sup>

		$[\text{2H}]\text{DMF}$	$[\text{2H}]\text{DMF} + \text{LiCl}$
$E_a/\text{kcal mol}^{-1}$	.. ..	$21.6 \pm 0.2$	$25.0 \pm 0.5$
$T_c/^\circ\text{C}$	.. ..	125.0	137.6
$\Delta G^\ddagger(T_c)/\text{kcal mol}^{-1}$	.. ..	$21.2 \pm 0.1$	$21.8 \pm 0.1$
$\Delta S^\ddagger(T_c)/\text{cal K}^{-1} \text{mol}^{-1b}$	.. ..	$-1.0 \pm 0.5$	$+5.8 \pm 1.3$
$\Delta\nu_{AB}/\text{Hz}$ (at 39 °C)	.. ..	9.85	12.15
$k(T_c)/\text{s}^{-1}$	.. ..	17.4	21.9

<sup>a</sup> The errors quoted represent only the probable errors in fitting data and do not reflect possibly larger systematic errors known to be inherent in n.m.r. studies of this type (ref. 17).

<sup>b</sup>  $\Delta S^\ddagger = (E_a - RT - \Delta G^\ddagger)/T$ .

We find the  $E_a$  value in pure  $[\text{2H}]\text{DMF}$  to be 21.6 kcal  $\text{mol}^{-1}$ .<sup>‡</sup> This value is close to that reported by Inglefield *et al.*<sup>14</sup> (20.8–21.6), Rabinowitz and Pines<sup>15</sup> (20.5), and Drakenberg *et al.*<sup>16</sup> (21.3). The value of Neumann and Jonas<sup>9</sup> (24.3) seems to be too high; however, the values of  $T_c$  and  $\Delta G^\ddagger$  at  $T_c$  found in our study are close to those reported by Neumann and Jonas. With added LiCl,  $E_a$  increases from 21.6 to 25.0 kcal  $\text{mol}^{-1}$  (Table), an appreciable change that can almost certainly be ascribed to an enhancement of the barrier to rotation of the C-N bond on complexation of  $\text{Li}^+$ . Such an increase in barrier has been predicted from optical spectra and M.O. calculations.

In a molecule such as DMF,  $\Delta S^\ddagger$  is expected to be near zero; the value found here ( $-1$  cal  $\text{K}^{-1} \text{mol}^{-1}$ ) is zero within experimental error. On addition of LiCl,  $\Delta S^\ddagger$  increases to nearly 6 cal  $\text{K}^{-1} \text{mol}^{-1}$ . Since  $\Delta S^\ddagger$  is known to be very sensitive to experimental errors, one must be cautious in interpreting changes of this magnitude,<sup>17</sup> but it seems likely that the increase is real and probably attributable to a reordering of ligands or solvent molecules as internal rotation occurs in the  $\text{Li}^+$  complex.

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