Neighbouring group participation by amide N–H and its possible biological significance

Sosale Chandrasekhar* and S. Srinivasa Gopalan

Department of Organic Chemistry, Indian Institute of Science, Bangalore 560 012, India

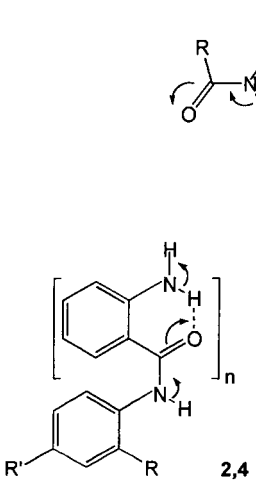
J. Chem. Research (S), 2000, 504–505
J. Chem. Research (M), 2000, 1237–1256

The titled phenomenon is possibly indicated in the hydrolysis of 4-(1-aziridin-1-ylmethanoyl)-5*H*-phenanthridin-6-one (**7b**); the relay version is believed to occur in organised polypeptides.

The possibility that a neighbouring amide group can stabilise an anionic centre *via* hydrogen bonding with its NH group has not been seriously tested: surprisingly, because of the preponderance of the amide NH group in proteins, and the possibility that the above effect can be amplified by relay in polypeptide units along their hydrogen bond chains (*e.g.* the α helix and the β sheet). Such charge dispersal may well be the key to the Pauling theory of the stabilisation of transition states at the active sites of enzymes.[3] Recent theoretical work strongly supports this possibility,[2] and prompts us to report our own experimental studies – initiated,[1] in fact, in the early 1990s.

The present studies focused on demonstrating the effect of a single amide NH moiety on nucleophilic addition to a carbonyl group, *via* hydrogen bonding to the carbonyl oxygen atom (**1**). In particular, the hydrolysis of carboxylic amides and esters in substrates with pendant carboxylic amide NH groups was marked for kinetic study. A vexing problem in such substrates is the possibility of nucleophilic catalysis[4a,5,6] by the pendant amide (likely at high pHs), and direct acid catalysis (likely at low pHs). Thus, substrates which were reactive at neutral pH, or those in which nucleophilic catalysis was not geometrically feasible, were sought. Three different systems – the anthranilate, the salicylate and the phenanthridone – were designed and studied.

The anthranilates: Initial studies were directed at the anthranilic acid (**2**) system,[7a] because of its internally chelated form. When methyl anthranilate (**2a**) was reacted with isatoic anhydride (**3**) in the melt (120 °C for 8 h), the expected anthranilate **2b** was accompanied by its higher analog **2c** in the ratio **2b:2c** = 1.8:1 (59 % total yield). The substantial formation of **2c** despite the presence of the ester **2a** in considerable excess indicated that **2b** was more nucleophilic than **2a** (supported by various control experiments). The inductive effect of the neighbouring ester group in **2a** is the simplest explanation, although the possibility of a hydrogen bond relay in **2b** also needs to be considered.

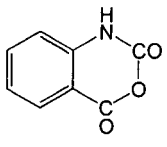


1

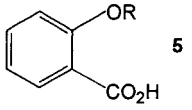
2,4

	R	R'
2	CO_2Me	H
4	H	CO_2Me

	n
a	0
b	1
c	2



3

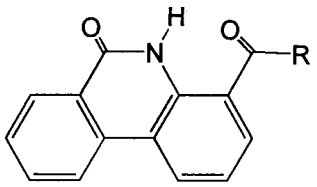


5

5	R
(a)	CO-Me
(b)	$CO-(CH_2)_2-NH-CO-Ph$
(c)	$[CO-(CH_2)_2-NH]_2-CO-Ph$
(d)	$CO-(CH_2)_2-NMe-CO-Ph$

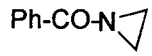
$HO-[CO-(CH_2)_2-NH]_n-CO-Ph$

6	n
(a)	1
(b)	2



7

7	R
(a)	OH
(b)	



8

Scheme 1

The salicylates: This system was chosen for study essentially because acetyl salicylic acid (aspirin, **5a**) is known to hydrolyse at neutral pH at a convenient rate (by a mechanism involving general base catalysis):[9] analogs with pendant secondary amide groups were designed. Thus, O-(*N*-benzoyl-β-alanlyl)salicylic acid (**5b**) and O-[*N*-(*N*-benzoyl-β-alanyl)-β-alanyl]salicylic acid (**5c**) were prepared (42% and 56% yield respectively), *via* DCC/DMAP promoted esterification of the respective β-alanines (**6a** and **6b**) with benzyl salicylate (not shown), followed by hydrogenolysis of the resulting benzyl esters (not shown). Also studied was the *N*-methyl derivative **5d.**

The kinetics of the ester hydrolyses of the analogs **5b, 5c** and **5d** were studied spectrophotometrically at pH 6.5 (as for

* To receive any correspondence. E-mail: sosale@orgchem.iisc.ernet.in

Table 1 Kinetic data[a] for the hydrolysis of the alanylsalicylic acids at 39 °C.

Compd no	$10^5\ k_{obsd}/s^{-1}$	$\Delta H^{\ddagger}$/kcal mol^{-1}	$\Delta S^{\ddagger}$/e.u.	$\Delta G^{\ddagger}$/kcal mol^{-1}
5a[b]	1.11	18.39 ± 0.01	–(22.5 ± 0.02)	25.38 ± 0.02
5b	2.15	18.0 ± 0.1	–(22.3 ± 0.1)	25.0 ± 0.1
5c	1.48	19.6 ± 0.6	–(18.0 ± 2.1)	25.2 ± 1.3
5d	2.50	19.2 ± 0.3	–(18.3 ± 1.1)	24.9 ± 0.7

[a]Correlation coefficients > 0.95; $\Delta G^{\ddagger}$ from at least three runs at 34–49 °C; [b]from ref. 12.

Table 2 Kinetic data[a] for the hydrolysis of the acylaziridines at 25 °C.

Compd no	$10^3\ k_{obsd}/s^{-1}$	$\Delta H^{\ddagger}$/kcal mol^{-1}	$\Delta S^{\ddagger}$/e.u.	$\Delta G^{\ddagger}$/kcal mol^{-1}
7b	3.10 ± 0.1	14.6 ± 1.1	–(21.6 ± 3.6)	21.0 ± 2.2
8	0.58 ± 0.1	8.6 ± 1.4	–(44.5 ± 4.7	21.9 ± 2.8

[a]$\Delta G^{\ddagger}$ based on three runs at 20–35 °C.

aspirin),[9] the data being collected in Table 1. The rate ratio **5a:5b:5c:5d** = 1:2:1.4:2.3 at 39 °C, and indicates either a marginal effect of amide NH, or a near cancellation of large effects. The nullification of large effects may arise from competing ground state and transition state stabilisation *via* chelation; an explanation for this is offered based on the 'hard and soft acids and bases principle'. Although an earlier report by Japanese workers had evidenced chelation in several ethyl β-alaninates by IR spectroscopy,[11a] **5b, 5c** and their benzyl esters did not offer firm evidence of chelation. Also, the anomalously high reactivity of the *N*-methyl derivative **5d** (in which no chelation is possible), and the use of aqueous media (which would disfavour chelation) were clearly sources of ambiguity.

The phenanthridone: A problem with the salicylates is ester resonance [$-^{+}O{=}C(R){-}O^{-}$], which would facilitate ground state chelation (assuming that this plays a role). This was sought to be avoided in the aziridine amide (**7b**) of phenanthridin-6-one-4-carboxylic acid (**7a**), essentially because amide resonance is largely thwarted in the *N*-acylaziridines.[13b]

The *N*-acylaziridine **7b** was prepared from the known[14] carboxylic acid **7a** *via* its acid chloride (53% yield; IR data did not exclude chelation in **7b,** but indicated considerable loss of amide resonance). The kinetics of the hydrolysis of **7b** were determined at pH 10.9 spectrophotometrically [as for *N*-benzoylaziridine (**8**)[13b]], the data being collected in Table 2. It is seen that **7b** hydrolyses 5.3 times as fast as **8:** although this seems to suggest NH participation, it is intriguing that the acceleration is entropic rather than enthalpic in origin. (The enthalpy data may indicate conjugative stabilisation of the reacting carbonyl group in **7b.**) The entropy data indicate substantial pre-ordering in the case of **7b:** it is possible that the chelation by the pre-positioned NH moiety obviates the need for solvation, which presumably dominates the reaction of **8.** (These conclusions are predicated on a negligible inductive effect of the phenanthridone moiety: this is difficult to estimate quantitatively, but it would appear that the entropy data indicate N–H participation.)

All the above model compounds were characterised spectrally and analytically (HRMS in the case of **7b**); the terminal *N*-acetyl derivatives of **2b** and **2c** possessed the reported[8a] characteristics. Interestingly, the X-ray crystal structure[12] of the alanylalanine **6b** shows each β-alanine unit in a *gauche* conformation, indicating chelation in the crystal.

General conclusions

The kinetic effect of amide N–H was not clearly manifest in the above model systems (except possibly in the phenanthridone). The results may be interpreted as evidence for N–H participation, but by involving complicated explanations.

We thank U.G.C. (New Delhi) for financial support.

Received 15 June 2000; accepted 17 September 2000
Paper 00/375

References cited in this synopsis

1 S. Srinivasa Gopalan, Ph. D. Thesis, Indian Institute of Science, April 1998.
2 H. Guo and D.R. Salahub, *Angew. Chem., Int. Ed. Engl.,* 1998, **37,** 2985-2989.
3 A.R. Fersht, *Enzyme Structure and Mechanism,* W. H. Freeman, New York, 2nd edn, 1985.
4 (a) B. Capon, *Chem. Soc.Rev.,* 1964, **18,** 45-111.
5 T. Cohen and J. Lipowitz, *J. Am. Chem. Soc.,* 1967, **86,** 5611–5616.
6 F.M. Menger and C. L. Johnson, *Tetrahedron,* 1967, **23,** 19–27.
7 (a) W. J. Hickinbottom, in *Rodd's Chemistry of Carbon Compounds,* ed. E. H. Rodd, Elsevier, Amsterdam, 1954, vol. III A, ch. 10, pp. 577–584.
8 (a) Y. Hamuro, J. S. Geib and A. D. Hamilton, *J. Am. Chem. Soc.,* 1996, **118,** 7529–7541.
9 A. R. Fersht and A. J. Kirby, *J. Am. Chem. Soc.,* 1967, **89,** 4853–4857; 4857–4863.
11 (a) N. Mori, M. Aihara, Y. Asabe and Y. Tsuzuki, *Bull. Chem. Soc. Jpn.,* 1972, **45,** 1786–1790.
12 S. Chandrasekhar, T. N. Guru Row, S. Srinivasa Gopalan and K. Vishnumurthy, unpublished work.
13 (b) H. Slebocka-Tilk and R. S. Brown, *J. Org. Chem.,* 1987, **52,** 805–808, and references therein.
14 A. Resplandy and P. Le Roux, *Bull. Soc. Chim. Fr.,* 1968, 4975–4981.