Tetrahedron Letters, Vol.22, No.32, pp 3101 - 3104, 1981
Printed in Great Britain

0040-4039/81/323101-04$02.00/0

PROOF OF DELOCALIZATION-STABILIZATION BY SULFUR IN ENOLATE FORMATION DURING RACEMISATION OF SULFUR CONTAINING AMINO ACID RESIDUES.

M.J.O. Anteunis*, F. Borremans, D. Wante and R. Schrooten
Laboratory of Organic Chemistry, Rijksuniversiteit-Gent, Krijgslaan 271 S4bis, 9000 GENT (BELGIUM)

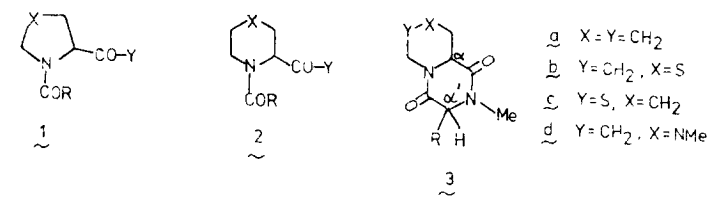
The present kinetic data on the accelerating action of sulfur in the base catalyzed racemisation of cyclic dipeptides (3) support the explanation of Barber and Jones (4) for the α center and suggest further a 1,3- and 1,4-interaction of -S- with the amide N-atom (8) for the α' center

The problem of ease of racemisation is paramount for the success of peptide synthesis whereby often active esters of amino acids that are optically labile have to be used. Among these (S-benzyl)-cysteine is especially prone to racemisation (1) in the presence of base. It has been shown that the pathway is via C_α-deprotonation (enolization) (1, 3) and not via β-elimination (4) or the otherwise favoured 5(4H)oxazolone route (5, 6). This particular case [that is equalled by the racemisation of cystine in acidic medium (7)] is in contrast to the homolog methionine that behaves normally (8). On the other hand the cyclic corgeners N-acyl-thiazolidine-4-carboxylic esters (1; X = S) and the thiapipecolic analog (2; X = S) resist racemisation, unless R is sterically uncongested (R = H) or a strongly electron-withdrawing group (9, 10). Therefore it could be expected that the latter cyclic imino acid residues, once incorporated in a cyclic dipeptide system (diketopiperazine, DKP, e.g. 3), would also racemise easily. This parallels the fact that proline resists racemisation when incorporated in linear peptides (11-13) but does not in DKP's (14-18) being moreover involved in a preceeding cis-peptide bond, favouring C_α-deprotonation (19). This is borne out by our present kinetic studies of epimerization and deuteration on some DKP's 3. By following the rate of deuteration by ^1H nmr spectroscopy and the rate of epimerization at both α-carbon centers for some DKP's (3a-d; with R = $CH_2C_6H_5$ and R = H) by a procedure developed at these laboratories (20), the relative rates were obtained, see table. In the case of c/Pip,Phe/ the α-Pip center epimerized faster than the α-Phe center. Preliminary results for α' deuteration of c/γ(S)Pip,Sarc/ (3b; R = H) show that deuterium incorporation in alkaline medium is faster

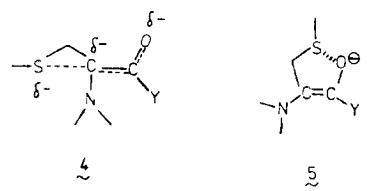
TABLE. Relative rates for racemisation (k_{rel}) at respectively α(Pip) and α'(Phe) centers in H_2O:t.BuOH (75:25 v:v) at 35°C, starting from the least stable (14, 21, 28) trans-c/Pip,Phe/(structures 3: R = $CH_2C_6H_5$).

	(R = $CH_2C_6H_5$)			
	3a	3b	3c	3d
k_{rel} (α-Pip)	1	42	31	0
k_{rel} (α'-Phe)	1	29	21	∿0.1

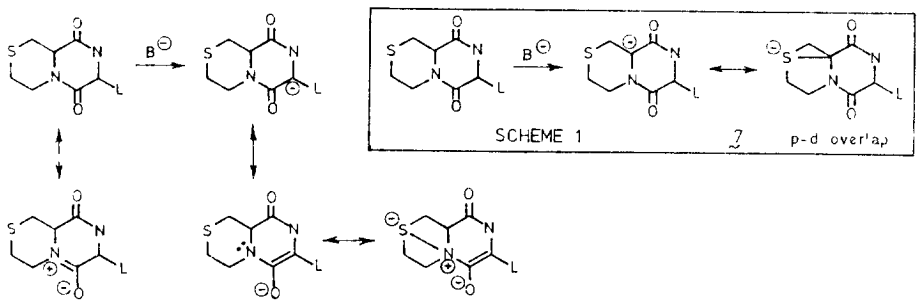
for H_{ax}-α' by a factor of 60 and for H_{eq}-α' by a factor of 43 than for the α-center. These α'-protons exchange for deuterium at a rate that is ten times faster than in the parent Pip-derivatives (3a; R = H), while in the latter case the α-proton completely resists exchange under the same conditions. Rates of deuteration for c/Pip,Phe/ compounds (3a-3c) run parallel with the epimerization, disclosing the carbanion-enolate nature of the process and excluding a substantial participation of an isoracemisation process (3) as observed for cysteine fragments by triethylamine catalysis in chloroform-methanol mixtures.



1 2 3



4 5 6



8

SCHEME 2

Two mechanisms have been proposed in order to explain the accelerating action of a sulfur atom at a position beta to the deprotonation site, in addition to its inductive electronegative contribution (9, 10). The one proposed by Barber and Jones (9, 10) invokes an overlap of the p orbital of the α-carbanion, or alternatively of the π orbital of the delocalized enolate with the d orbital of sulfur (d-p or d-π overlap) as shown in 4. This possibility is rejected by Kovacs et al (6, 8) arguing that such overlap would even be more favourable in the case of methionine. Their mechanism makes use of an oxygen anion-sulfur attractive interaction that would be favourable in a five-membered (5) but not in a six-membered (6) configuration. Our present findings strongly

support the Barber-Jones mechanism, e.g. as represented in SCHEME 1. This follows from the observations that (a) the cyclic intermediates by S-$O^{\ominus}$ interaction are constitutionally impossible in the present case of the DKP's;
(b) it seems unlikely that a hard ($O^{\ominus}$)-soft (S) interaction is favourable in comparison with a soft ($C^{\ominus}_{\alpha}$ or π-system)-soft (S) interaction;
(c) a statistical treatment of long-range atomic interactions from solid state data of 21 representative proteins has shown (22) that sulfur atoms avoid negatively charged oxygen atoms;
(d) the hexacyclic intermediate $\underset{\sim}{6}$ would be (much) less favourable than the pentacyclic $\underset{\sim}{5}$ proposal;
(e) in contrast to the favourable two-electron delocalisation p-d, the resistance towards C_{α}-epimerization of the aza congener ($\underset{\sim}{3}$d, R = $CH_2C_6H_5$) is noticeable. It is clear that the unfavourable four-electron delocalization that would operate in the Barber-Jones mechanism is now avoided;
(f) Morgan et al. have proposed an attractive interaction to be possible between an aromatic moiety and sulfur (24-26); an interaction that has been corroborated by our own findings (21, 27, 28). This exemplifies the generality of soft sulfur-p(or π)interactions.

Further, it is obvious that the absence of the effect in methionine is simply due to the unfavourable 1-4 approach in the Barber-Jones concept for acyclic side chains which is overcome in a transannular disposition ($\underset{\sim}{3}$c).

Perhaps the most remarkable result is the fact that the effect of S-acceleration/N-deceleration is also felt in the α'-position, viz. at the opposite site of the DKP-ring. This can, in analogy to the foregoing statements, be explained by an interaction of S with the amide nitrogen atom in the same ring when the latter has received sp^3 character while this occurs only in the α'-deprotonated carbanion ($\underset{\sim}{8}$). The fact that these combined effects are met both in the γ-S and δ-S Pip congeners illustrates that both 1-3 and 1-4 dispositions of S versus carbanion or sp^3-N are operative especially in favourable configurations (e.g. cyclic fragments)(30).

REFERENCES AND NOTES.

(1) J. Kovacs, G.L. Mayers, R.H. Johnson, R.E. Cover and U.R. Ghatak, J. Org. Chem., 35, 1810 (1970).
(2) J. Kovacs, G.L. Mayers, R.H. Johnson and U.R. Ghatak, J. Chem. Soc., Chem. Commun., 1066 (1968).
(3) J. Kovacs, Y. Hsich, E.M. Holleran and Y.F. Ting, Proceedings of the 15th Eur. Pept. Symp., (September, Gdansk - Poland) 159 (1978).
(4) M. Bodansky and A. Bodansky, J. Chem. Soc., Chem. Commun., 591 (1967).
(5) See e.g. D.S. Kemp, "Racemisation in Peptide Synthesis, 'The Peptides'" Vol. I, (Editors E. Gross and J. Meienhofer) Acad. Press N.Y., 1979, p. 332.
(6) J. Kovacs, Y. Hsich, K.Y. Hui and S.E. Kim, "Peptides, Proceedings of the 5th Amer. Peptide Symp." (Editors M. Goodman and J. Meienhofer) Wiley, N.Y., 1977, p. 461.
(7) S.J. Jacobson, C.G. Wilson and H. Rapoport, J. Org. Chem., 39, 1074 (1974).
(8) J. Kovacs, E.M. Holleran and K.Y. Hiu, J. Org. Chem., 45, 1060 (1980).

(9) M. Barber and J.H. Jones, in "Peptides 1976" Proceedings of the 14th Eur. Pept. Symp., Wépion (Editor A. Loffet) Edit. Univ. Bruxelles, Brussels (1976), 109.

(10) M. Barber, J.H. Jones and M.J. Wittig, J. Chem. Soc. Perkin I, 2425 (1979).

(11) M.W. Williams, G.T. Young, J. Chem. Soc., 3701 (1964).

(12) Cf. J.R. Mc.Dermott and L. Benoiton, Canad. J. Chem., 51, 2555, 2562 (1973).

(13) This is also less the case if Pro is at the penultimate carboxylic end position by a 5(4H) oxazolon mechanism via exocyclic enolization (5, 11).

(14) J. Vičar and K. Bláha, Coll. Czechosl. Chem. Commun., 38, 3307 (1973).

(15) J. Vičar, M. Buděšinský and K. Bláha, Coll. Czechosl. Chem. Commun., 38, 1940 (1973).

(16) H. Ott, A.J. Frey and A. Hofman, Tetrahedron, 19, 1675 (1963).

(17) C. Eguchi and A. Kakuta, J. Am. Chem. Soc., 96, 3985 (1974) and Bull. Chem. Soc. Japan 47, 2277 (1974).

(18) U. Schmidt and A. Nikiforov, Monatsh. f. Chemie, 106, 313 (1975).

(19) M. Goodman and M. Fried, J. Am. Chem. Soc., 89, 1264 (1967) Cf. J.E. Mark and M. Goodman, J. Am. Chem. Soc., 89, 1267 (1967).

(20) F. Borremans, M.J. Anteunis and R. Schrooten, submitted J. Chem. Soc., Transact. Faraday Soc.,

(21) R. Schrooten, F. Borremans and M.J. Anteunis, in preparation.

(22) P.K. Warme and R.S. Morgan, J. Molec. Biology, 118, 273 (1978).

(23) In linear fragments, the presence of a ß-nitrogen accelerates epimerization in acidic medium (7), but the mechanism of preference is the 5(4H)oxazolone.

(24) We have observed however also appreciable amounts of racemisation during peptide synthesis with γ-thiapipecolic esters, where slow-coupling active esters of it require days to achieve reasonable completion, and where carefull control of activating species, concentration and amount of base are needed to avoid these difficulties (cf. 8).

(25) B.L. Bodner, L.M. Jackman and R.S. Morgan, Abstr. Am. Meeting Biophys. Soc.; Biophys. J., 17, 57a (1977).

(26) R.S. Morgan and J. McAdon, Intern. Journ. Pept. Protein Res., 15, 177 (1980).

(27) B.L. Bodner, L.M. Jackman and R.S. Morgan, Biochem. Biophys. Res. Commun., 94, 807 (1980).

(28) M.J. Anteunis, R. Schrooten and W. Nachtergaele, Bull. Soc. Chim. Belges, 89, 289 (1980).

(29) R. Schrooten, S. Joshi and M.J. Anteunis, Bull. Soc. Chim. Belges, 89, 615 (1980).

(30) The depicted interactions are not necessarily acting through-space, cf. R. Sarneel, C.W. Worrell, P. Pasman, J.W. Verhoeven and G.F. Mes, Tetrahedron, 36, 3241 (1980) and references cited there.

(Received in UK 5 June 1981)