Tetrahedron Letters, Vol.22, No.35, pp 3359 - 3360, 1981
Printed in Great Britain

0040-4039/81/353359-02$02.00/0

THE RELATIVE SUSCEPTIBILITY TO RACEMIZATION OF L- AND D-RESIDUES IN PEPTIDE SYNTHESIS

N. Leo Benoiton*, K. Kuroda and Francis M.F. Chen

Department of Biochemistry, University of Ottawa,
Ottawa, Canada K1N 9A9

ABSTRACT: An activated D-residue of a protected dipeptide when coupled racemizes twice as much as an activated L-residue, which is contrary to the case for couplings of acylamino acids.

The extent of racemization in peptide synthesis depends on many factors[1] including the coupling method, solvent polarity, and the nature of the residue undergoing activation.[2,3] Moreover, it depends in particular on the type of N-substituent on the activated residue, namely alkoxycarbonyl [ROCO-] << acyl [RCO-] < acylaminoacyl [R'CONHCHRCO-].[1,3] Only recently has the influence of the configuration of the activated residue been demonstrated. Arendt et al.[4] showed that Ac-L-Leu racemizes three times as much when coupled with L-Leu-OMe than when coupled with D-Leu-OMe [Table]. The inference is that more racemization will be observed when coupling an L-residue with an L-isomer than when coupling a D-residue with an L-isomer. We have confirmed this hypothesis using benzoylamino acids,[5] the L-residues racemizing more than the D-residues when coupled with an ester of L-lysine [Table]. However, we now find that this phenomenon holds for acylamino acids only. When protected dipeptides [Z-Gly-Xxx] were coupled, the activated D-residues [Xxx] underwent twice as much stereomutation as did the L-residues [Table]. This was observed for three different residues [Leu, Phe, Val] coupled using DCC alone in dichloromethane or DMF, or in the presence of HOBt[6] in aqueous DMF. In the latter case, water was added in order to increase the racemization[3] so that a clear difference would be observed. The conclusion to be drawn from the data is that unless an adjacent chiral residue instead of glycyl in this case would drastically change the course of events, the danger of racemization is twice as great when a peptide bond is formed between residues of opposite configuration than in normal peptide synthesis.

We have already described[3] cases where conclusions based on model tests implicating activated benzoylamino acids did not apply to the couplings of activated dipeptides [Z-Gly-Xxx]. The additional example presented here serves to emphasize the caution which should be exercised in interpreting racemization data obtained using acylamino acids.

Experimental: To a mixture containing Z-Gly-Xxx[7] [0.50 mmol], L-Lys(Z)-OBzl.HCl[8] [0.50 mmol] and N-methylmorpholine [0.45 mmol] in 25 ml of solvent was added HOBt[6] [0.50 mmol] where pertinent followed by DCC [0.50 mmol]. The diastereomeric Gly-D/L-Xxx-Lys peptides were analyzed with an amino acid analyzer[7] after appropriate work-up and deprotection of the coupling mixtures.[3]

Abbreviations: Ac, acetyl; Bz, benzoyl; Bzl, benzyl, DCC, N,N'-dicyclohexylcarbodiimide; DMF, N,N-dimethylformamide; HOBt, 1-hydroxybenzotriazole; Z, benzyloxycarbonyl.

Table. Racemization during coupling with an amino acid ester[a]

Substrate	Coupling method	Xxx					
		L-Leu	D-Leu	L-Phe	D-Phe	L-Val	D-Val
Z-Gly-Xxx[b]	DCC/CH_2Cl_2	24	56	28	58	16	34
	DCC/DMF	35	70	42	62	64	106
	DCC-HOBt/DMF:H_2O (2:1)	12	23	16	36	34	64
Bz-Xxx[c]	DCC/CH_2Cl_2	104	65	115	71	72	31
Ac-Xxx[d]	DCC/dioxane	80	26[+]				

a) Data given as % racemization = 2 x [D-L or L-L]/[L-L + D-L].

b) Coupling of Z-Gly-Xxx with L-Lys(Z)-OBzl.

c) Coupling of Bz-Xxx with L-Lys(Z)-OMe. Data of Benoiton et al.[5]

d) Coupling of Ac-L-Leu with L- and D-Leu-OMe[+]. Data of Arendt et al.[4]

This work was financially supported by the Medical Research Council.

References

1. D.S. Kemp in: The Peptides Vol. 1, Major Methods of Peptide Bond Formation (E. Gross and J. Meienhofer, eds.), Academic Press Inc., New York, 1979. pp. 315-383.
2. N.L. Benoiton, K. Kuroda and F.M.F. Chen, Int. J. Peptide Protein Res. 13, 403-408 (1979)
3. N.L. Benoiton and K. Kuroda, Int. J. Peptide Protein Res. 17, 197-204 (1981).
4. A. Arendt, A.M. Kolodziejczyk and T. Sokolowska, Tetrahedron Lett. 2711-2712 (1978).
5. N.L. Benoiton, K. Kuroda and F.M.F. Chen, Int. J. Peptide Protein Res. 15, 475-479 (1980)
6. W. König and R. Geiger, Chem. Ber. 103, 788-798 (1970).
7. N.L. Benoiton, K. Kuroda, S.T. Cheung and F.M.F. Chen. Can. J. Biochem. 57, 776-781 (1979).
8. S.T.Cheung and N.L. Benoiton, Can J. Chem. 55, 916-921 (1977).

(Received in USA 26 March 1981)