Tetrahedron Letters No.46, pp. 4733–4734, 1968. Pergamon Press. Printed in Great Britain.

THREO- AND ERYTHRO-β-HYDROXY-L-ASPARAGINES

A. Singerman and Y. Liwschitz

Department of Organic Chemistry, The Hebrew University of Jerusalem, Israel.

(Received in UK 5 August 1968; accepted for publication 12 August 1968)

The recent findings of Campbell et al. (1) regarding the action of L-asparaginase on various asparagine-requiring tumours have prompted the syntheses and investigation of analogues of this amino acid (2).

We wish to report the preparation of the β-amides of threo- and erythro-β-hydroxy-L-aspartic acids by ammonolysis of β-benzyl threo-β-hydroxy-L-aspartate (3) and β-methyl erythro-β-hydroxy-L-aspartate respectively. The latter ester was prepared by direct esterification of erythro-β-hydroxy-L-aspartic acid obtained by resolution of its racemic N-benzyl derivative by means of L-histidine and subsequent hydrogenolysis (4).

The amides differ in their stability in acidic medium. The one belonging to the threo- series when left for several hours in N HCl is partly hydrolysed to threo-β-hydroxy-L-aspartic acid, detected on chromatograms, which caused a sharp drop in optical rotation. Erythro-β-hydroxy-L-asparagine, however, was stable under the same conditions.

The "β-hydroxy asparagine" isolated from normal human urine (5) could not be identified with either of the isomers synthesised by us, having different properties (R_f values and IR spectrum).

Threo-β-hydroxy-L-asparagine - β-Benzyl threo-β-hydroxy-L-aspartate (3) (1 g) was dissolved in 25% aqueous ammonia (10 ml). After 7 days at room temperature the solution was evaporated to dryness in vacuo. The solid residue was dissolved in hot water and the pH adjusted to 5 by addition of 6N HCl. The substance precipitated and was recrystallised from water (0.6 g, yield quantitative); m.p. 278° (decomp.); $[\alpha]_D^{22}$ -30.6° (c 2.45 in N HCl) (Found: C, 32.2; H, 5.9; N, 18.9; N (Van Slyke), 9.6. $C_4H_8N_2O_4$ requires: C, 32.4; H, 5.4; N, 18.9; N (Van Slyke), 9.4%).

β-Methyl erythro-β-hydroxy-L-aspartate - To a suspension of erythro-β-hydroxy-L-aspartic acid (4) (5 g) in anhydrous methanol (50 ml) was added concentrated HCl (6 ml). The mixture was heated under reflux for 3 hrs. The

solvents were removed in vacuo, the residue was dissolved in ethanol and the pH adjusted to 8 by addition of pyridine. The resulting precipitate was recrystallised from 50% aqueous ethanol. (3.5 g, 65%); m.p. 229° (decomp.); $[\alpha]_D^{22}$+65.5° (c, 1.8 in N HCl) (Found: C, 36.8; H, 5.8; N, 8.5; N (Van Slyke), 8.7. $C_5H_9NO_5$ requires: C, 36.8; H, 5.5; N, 8.6; N (Van Slyke), 8.6%).

Erythro-β-hydroxy-L-asparagine was prepared in an analogous manner to the threo isomer by ammonolysis of the above ester; m.p. 260° (decomp.); $[\alpha]_D^{22}$+63.4° (c 1.9 in N HCl) (Found: C, 32.4; H, 5.9; N, 18.9; N (Van Slyke), 9.2. $C_4H_8N_2O_4$ requires: C, 32.4; H, 5.4; N, 18.9; N (Van Slyke), 9.4%).

Paper chromatography in 80% phenol-water (light-brown colour with ninhydrin) gave R_f values 0.18 and 0.13 for threo- and erythro-β-hydroxy-L-asparagine respectively. On TLC (silica gel) in n-butanol-acetic acid-pyridine-water (2:1:1:4) they were 0.39 and 0.23 respectively.

REFERENCES

1) H.A. Campbell, L.T. Mashburn, E.A. Boyse and L.J. Old, Biochemistry, 6, 721 (1967).
2) E. Falco and G.B. Brown, J. Medicinal Chem., 11, 142 (1968).
3) Y. Liwschitz, A. Singerman and S. Sokoloff, J. Chem. Soc. (C), 1968 in press.
4) Y. Liwschitz, A. Singerman and Y. Wiesel, Israel J. Chem., 1968 in press.
5) F. Tominaga, C. Hiwaki, T. Maekawa and H. Yoshida, J. Biochem. (Japan) 53, 227 (1963).