[CONTRIBUTION FROM THE DEPARTMENT OF ORGANIC CHEMISTRY, THE HEBREW UNIVERSITY]

Syntheses of Aspartic Acid Derivatives. I. Some N-Acetyl-N-benzyl Derivatives of *dl*-Aspartic Acid

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N-Benzyl-*dl*-aspartic acid is converted by acetic anhydride into N-acetyl-N-benzylaspartic anhydride which yields β-amides when acted upon by aqueous ammonia or benzylamine. Various N^2-acetyl-N^2-benzyl derivatives of asparagine and aspartimide have been prepared.

While exploring the possibility of using N-benzyl-*dl*-aspartic acid, which is readily synthesized,[1] for the preparation of aspartyl amides and peptides by converting it into an inner anhydride by means of acetic anhydride, we found that beside ring closure, the secondary α-amino group always suffered acetylation,[2] even at room temperature, with the formation of the anhydride I. If I is acted upon by aqueous ammonia or an aqueous solution of benzylamine, the respective N^2-acetyl-N^2-benzyl-β-amides II and III are formed (see Chart I).

In order to ascertain the structure of II and III which could conceivably be either α- or β-amides, we proceeded as follows. Removal of the N^2-benzyl group in II and III by catalytic hydrogenolysis should have yielded IV or V, which are known,[3,4] or their hitherto unknown α-isomers. All efforts, however, to achieve this end, using different catalysts, *e.g.*, palladium chloride-on-charcoal (30%), palladium black and platinum oxide, were unsuccessful and the unchanged starting materials were recovered. Hence it must be presumed that the presence of an acyl group prevents the hydrogenolysis of a benzyl group on the same nitrogen atom.

We then treated N,N^2-dibenzyl-*dl*-asparagine (VII) with acetic anhydride in order to achieve acylation of the secondary amino group, expecting III to be formed. Analyses of the substance (X^x) thus obtained indicated that the introduction of the acetyl group was accompanied by elimination of one molecule of water, and it should, therefore, be assigned the structure of N^2-acetyl-N,N^2-dibenzyl-*dl*-aspartimide. Since this latter compound (X), which was derived by another route (see below), differed from X^x in melting point and solubility, and as both X and X^x could be transformed into N,N^2-dibenzyl-*dl*-aspartimide hydrochloride (XIII)[4] by reflux with dilute hydrochloric acid, it seems that X^x represents one of the possible two forms of the isoimide.[5] Likewise, 2-imino-4-(acetylbenzyl)-aminofuran-5-one (IX^x) was obtained on treating N^2-benzyl-*dl*-asparagine (VI) with acetic anhydride. On hydrogenolysis of XIII the hydrochloride of N-benzyl-*dl*-aspartimide (XIV) resulted. When this was treated with dilute sodium hydroxide followed by acidification with hydrochloric acid, opening of the ring occurred and N-benzyl-*dl*-asparagine (XV)[4,6] was obtained which crystallized from water in hexagonal plates (m.p. 267°). Its melting point was not depressed on mixing the substance with an authentic sample of N-benzyl-*dl*-asparagine, and it moreover gave the same spot on paper chromatograms.

Treatment of N,N^2-dibenzylasparagine (VII) and N,N^2-dibenzyl-α-asparagine (XII) with acetyl chloride in the cold resulted in the formation of the hydrochloride of VII (VIII) and N,N^2-dibenzylaspartimide hydrochloride (XIII). This latter compound was also formed on refluxing VII with 2 *N* hydrochloric acid. On prolonged reaction with acetyl chloride both compounds finally yielded N^2-acetyl-N,N^2-dibenzylaspartimide (X). X could also be obtained from XII by reflux with glacial acetic acid. N^2-Acetyl-N^2-benzylaspartimide (IX) resulted from VI by similar reactions.

The Hofmann reaction with II, as a further means of identification, was of no avail; benzylamine was invariably split off and the succinic acid residue transformed into an unidentified product. The same splitting off reaction took place when trying to degrade VI. Since Karrer and Schlosser[3] were able to perform the reaction with N^2-acetyl-*l*-asparagine, the different course of the reaction in our case must be ascribed to the presence of the N^2-benzyl group.

On treating X with aqueous sodium carbonate under reflux and acidifying with hydrochloric acid III was obtained. This constitutes evidence that III is a β-amide, since McMillan and Albertson[4] have shown that N^2-acetyl-N-benzylasparagine is obtained when opening N^2-acetyl-N-benzylaspartimide in a similar manner.

Moreover, comparison of the infrared spectra of II and III with those of substances lacking the acetyl group, *i.e.*, VI and XI on the one and VII and XII on the other hand, also confirms the β-nature of these two compounds. Both III and II possess a characteristic band (due to the amido group) at 6.2 μ which corresponds much more closely to the bands of the two β-derivatives at 6.1 and 6.15 μ, respectively, the α-isomers having bands at 6.0 and 6.05 μ.[7]

Finally, it should be remarked that when trying

(1) Max Frankel, Y. Liwschitz and Y. Amiel, THIS JOURNAL, **75**, 330 (1953).

(2) Y. Liwschitz and A. Zilkha, *ibid.*, **76**, 3698 (1954).

(3) P. Karrer and A. Schlosser, *Helv. Chim. Acta*, **6**, 411 (1923).

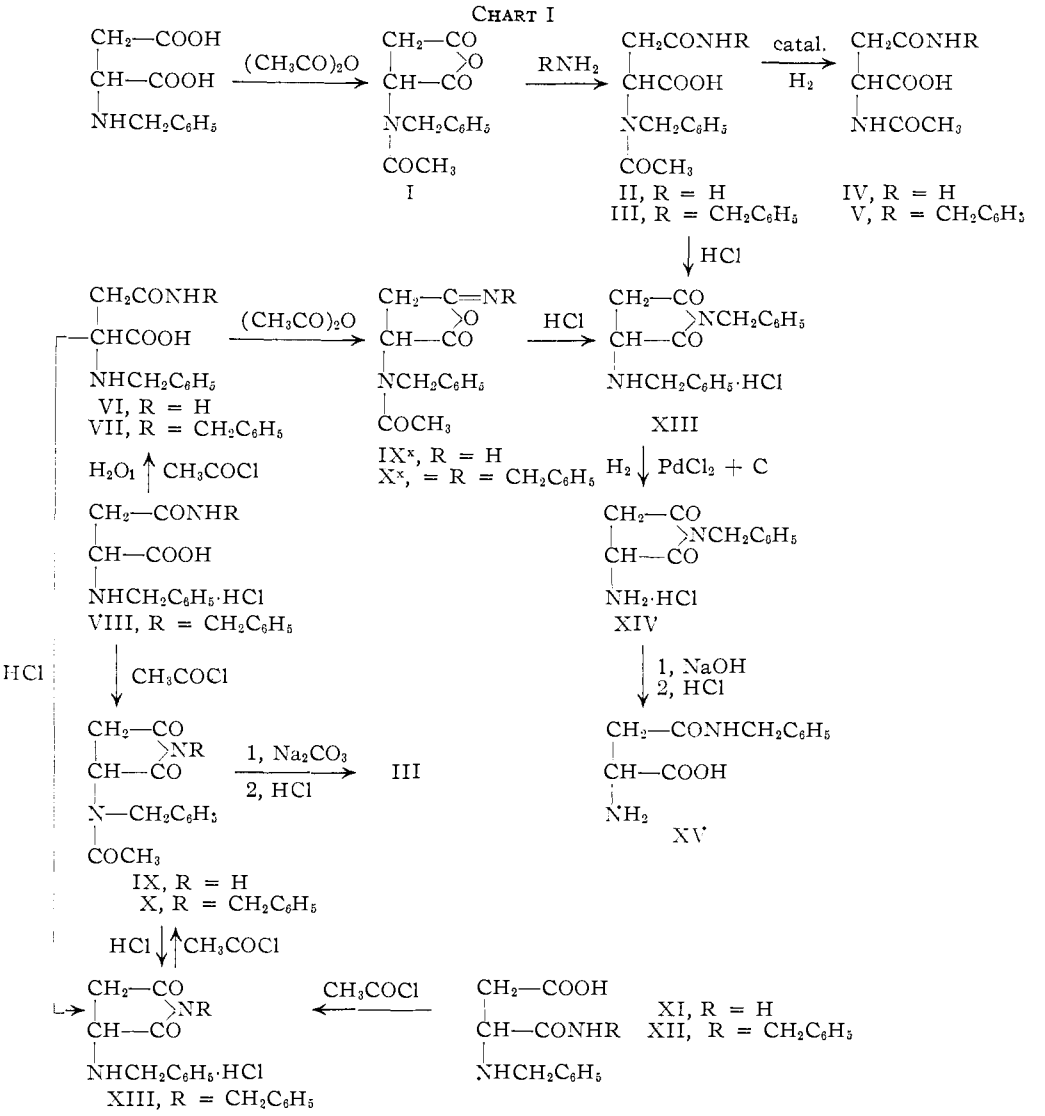
(4) F. H. McMillan and N. F. Albertson, THIS JOURNAL, **70**, 3778 (1948).

(5) M. L. Sherrill, F. L. Schaeffer and E. P. Shoyer, *ibid.*, **50**, 474 (1928).

(6) Max Frankel, Y. Liwschitz and A. Zilkha, *ibid.*, **75**, 3270 (1953).

(7) Infrared spectra of all these compounds have been prepared by Messrs. Samuel P. Sadtler and Son, Inc., to whom our thanks are due, and have been deposited as Document number 4783 with the ADI Auxiliary Publications Project, Photoduplication Service, Library of Congress, Washington 25, D. C. A copy may be secured by citing the Document number and by remitting in advance $1.25 for photoprints or $1.25 for 35 mm. microfilm payable to: Chief, Photoduplication Service, Library of Congress.

CHART I



to prepare ethyl N,N^2-dibenzyl-*dl*-asparaginate according to the method of McMillan and Albertson,[4] we invariably obtained N,N^2-dibenzyl-*dl*-asparagine hydrochloride (VIII). This compound in aqueous solution reverts to the starting material VII. Since the analysis for nitrogen in the above work (N, 7.64) could also account for VIII (calcd. N, 8.0) and no further analytical values or characterization of the base (obtained from the "ester" by treatment with dilute alkali) are given, we must conclude that these authors were dealing with the same substance we obtained according to their procedure, namely, VIII and not the ester.

Experimental[8]

N-Acetyl-N-benzyl-*dl*-aspartic Anhydride (I).—Thirty ml. of acetic anhydride was heated to boiling and N-benzyl-*dl*-aspartic acid (10 g.) added with stirring. The substance dissolved quickly and the solution was cooled in an ice-bath. On addition of ether–petroleum ether, a heavy oil separated which solidified on scratching and cooling in a freezing mixture; yield 7 g. (63%), m.p. 116–117°, m.p. 118° after recrystallization from ethyl acetate–petroleum ether (needles).

(8) Micro-combustion analyses were made by Drs. Weiler and Strauss. Melting points were determined in a Fisher–Johns apparatus.

Anal. Calcd. for $C_{13}H_{13}O_4N$: C, 63.2 H, 5.3; N, 5.7. Found: C, 63.3; H, 5.5; N, 5.7.

N^2-Acetyl-N^2-benzyl-*dl*-asparagine (II).—Twenty ml. of a 25% ammonium hydroxide solution was cooled in an ice-bath and compound I (2.7 g.) added with stirring. On acidification with hydrochloric acid white crystals separated. The substance was filtered and washed with water and ethanol; yield 2.1 g. (73%), m.p. 144–146°, m.p. after recrystallization from water 148°.

Anal. Calcd. for $C_{13}H_{16}O_4N_2$: C, 59.1; H, 6.1; N, 10.6. Found: C, 59.1; H, 6.0; N, 10.5.

N^2-Acetyl-N,N^2-dibenzyl-*dl*-asparagine (III).—A solution of benzylamine (4 g.) in 15 ml. of water was cooled in an ice-bath and compound I (2.5 g.) was added with stirring. On acidification with hydrochloric acid an oil separated which solidified on scratching and cooling; yield of crude product quantitative; recrystallization from ethanol (irregular octahedra) gave 2.9 g. (82%) of substance melting at 174°.

Anal. Calcd. for $C_{20}H_{22}O_4N_2$: C, 67.7; H, 6.3; N, 7.9. Found: C, 67.7; H, 6.2; N, 7.9.

2-Imino-4-(acetylbenzyl)-aminofuran-5-one (IX^x).—VI (4 g.) in 20 ml. of acetic anhydride was heated to boiling until it dissolved. The solution was then cooled in an ice-bath and ether and petroleum ether was added to precipitate a white substance; m.p. after recrystallization from ethyl acetate–petroleum ether 143°, yield 2.7 g. (61%).

Anal. Calcd. for $C_{13}H_{14}O_3N_2$: C, 63.3; H, 5.7; N, 11.4. Found: C, 63.9; H, 5.7; N, 11.2.

2-Benzylimino-4-(acetylbenzyl)-aminofuran-5-one (X^x).—VII (4 g.) was heated with 20 ml. of acetic anhydride until it dissolved completely. After cooling and on addition of ether and petroleum ether, a white substance was precipitated; m.p. after recrystallization from ethyl acetate–petroleum ether 118°, yield 3 g. (70%).

Anal. Calcd. for $C_{20}H_{20}O_3N_2$: C, 71.4; H, 6.0; N, 8.3. Found: C, 70.6; H, 6.2; N, 8.5.

N,N^2-Dibenzyl-*dl*-asparagine Hydrochloride (VIII).—VII (4 g.) was suspended in 20 ml. of glacial acetic acid and 20 ml. of acetyl chloride added. The substance dissolved at once and after a short time, the hydrochloride precipitated. This was filtered off five minutes after the start of the reaction and washed with acetyl chloride and ether; yield 2.7 g. (60%), m.p. 170–171°; on recrystallization from ethanol (hexagonal plates) the m.p. was raised to 172°.

Anal. Calcd. for $C_{18}H_{21}O_3N_2Cl$: C, 62.0; H, 6.1; N, 8.0; Cl, 10.2. Found: C, 61.6; H, 6.0; N, 8.0; Cl, 10.2.

N^2-Acetyl-N^2-benzyl-*dl*-aspartimide (IX).—VI (2.5 g.) was suspended in 15 ml. of acetyl chloride and on addition of 10 ml. of glacial acetic acid it dissolved completely. The reaction mixture was left for 6 hr. and finally petroleum ether was added which precipitated an oil. This solidified on recrystallization from water (rhombs), m.p. 144–145°. On mixing IX with IX^x the m.p. was considerably lowered; yield 1.6 g. (58%).

Anal. Calcd. for $C_{13}H_{14}O_3N_2$: C, 63.3; H, 5.7; N, 11.4. Found: C, 63.2; H, 5.7; N, 11.2.

N^2-Acetyl-N,N^2-dibenzyl-*dl*-aspartimide (X). (A) From VII.—VII (2 g.) was suspended in 10 ml. of glacial acetic acid and 10 ml. of acetyl chloride added. The hydrochloride which precipitated almost immediately was not separated and it dissolved in the reaction mixture after having been left for some hours at room temperature. On addition of ether, X was obtained; m.p. 140° on recrystallization from ethanol (triangular prisms), yield 1.3 g. (60%).

Anal. Calcd. for $C_{20}H_{20}O_3N_2$: C, 71.4; H, 6.0; N, 8.3. Found: C, 72.1; H, 5.9; N, 8.3.

(B) From XII.—XII (4 g.) dissolved in acetyl chloride (100 ml.) was refluxed for 0.5 hr. After cooling, ether and petroleum ether was added and an oil obtained. This on trituration with ethanol gave a solid which, when crystallized from the same solvent, melted at 140°.

(C) From III.—III (1 g.) suspended in 10 ml. of acetyl chloride was refluxed for 90 minutes, when it dissolved completely. After cooling, the solution was poured into 50 ml. of water. A white oil separated which solidified on scratching with a glass rod; m.p. after recrystallization from ethanol, 140°.

Hydrolysis of X.—X (1 g.) suspended in 20 ml. of 10% sodium carbonate solution was refluxed for 2 hr. After filtration from undissolved residue, the solution was acidified with hydrochloric acid. The precipitated substance melted at 172–173°. On recrystallization from ethanol the typical irregular octahedra of III, m.p. 174°, were obtained.

N,N^2-Dibenzyl-*dl*-aspartimide Hydrochloride (XIII). (A) From XII.—XII (4 g.) was dissolved in 110 ml. of acetyl chloride and left for 6 hr., when precipitation of white crystals commenced. After an additional 30 minutes, the substance was filtered and washed with acetyl chloride; yield 2.6 g. (59%), m.p. 191°, m.p. after recrystallization from ethanol (short needles), 195°.

Anal. Calcd. for $C_{18}H_{19}O_2N_2Cl$: C, 65.2; H, 5.7; N, 8.5; Cl, 10.7. Found: C, 64.9; H, 5.9; N, 8.7; Cl, 10.7.

(B) From X^x or X.—X^x or X (4 g.) suspended in 30 ml. of 5 *N* hydrochloric acid was heated at 130° (oil-bath) for 1 hr.; on cooling, crystals of XIII separated.

Anal. (for substance obtained from X) Found: C, 65.2; H, 5.8; N, 8.4; Cl, 10.7.

(C) From III.—As in B.

(D) From VII.—VII (2 g.) in 25 ml. of 2 *N* hydrochloric acid was refluxed for 30 minutes; on cooling, XIII crystallized; yield 1.5 g. (73%), m.p. 194°.

N-Benzyl-*dl*-aspartimide Hydrochloride (XIV).—XIII (2 g.) was dissolved in glacial acetic acid (25 ml.) and 0.2 g. of $PdCl_2$-on-charcoal (30%) added. Hydrogenolysis was carried out for 4 hr. at 70°. After separation of the catalyst by filtration the solvent was evaporated *in vacuo;* yield almost quantitative. The residue was recrystallized from ethanol (needles), m.p. 210°.

Anal. Calcd. for $C_{11}H_{13}O_2N_2Cl$: C, 54.9; H, 5.4; N, 11.6; Cl, 14.7. Found: C, 54.8; H, 5.3; N, 11.6; Cl, 14.6.

N-Benzyl-*dl*-asparagine (XV).—XIV was dissolved in 2 *N* sodium hydroxide and the solution was neutralized by addition of 2 *N* hydrochloric acid. XV precipitated and was recrystallized from water (hexagonal plates); m.p. 265° was not depressed by mixing with an authentic sample.[4]

Anal. Calcd. for $C_{11}H_{14}O_3N_2$: C, 59.4; H, 6.3; N, 12.6. Found: C, 59.4; H, 6.3; N, 12.2.

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[CONTRIBUTION FROM THE DEPARTMENT OF ORGANIC CHEMISTRY, THE HEBREW UNIVERSITY]

Syntheses of Aspartic Acid Derivatives. II. N-Alkylated α- and β-Asparagines

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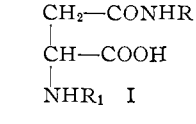
N-Alkylated-β-asparagines have been prepared by reaction of N-alkyl-maleamic acids with benzylamine and hydrogenolysis of the intermediate N^2-benzyl derivatives. In this last step, which had to be carried out in acid medium, partial ring closure to the aspartimide derivative occurred, depending on the nature of the alkyl group. N^2-Alkylated-β-asparagines were obtained when maleamic acid reacted with alkyl amines. N-Alkylated-α-asparagines have been synthesized by the reaction of the mixed anhydride of N-benzyl-*dl*-aspartic acid and chloroformic acid with primary amines.

The synthesis of derivatives of α- or β-asparagines in which either the α-amino, the amido or both groups are alkylated presents difficulties and only relatively few substances of this class have been recorded in the literature.[1]

We, therefore, sought to exploit the methods communicated in our previous papers for the preparation of such compounds.

The method, permitting the synthesis of aspartyl peptides,[2] has now been extended to the preparation of aspartyl amides, treated in the present paper. It has also been demonstrated that by variations of this method any asparagine derivative of type I (R and R_1 being either alkyl groups or one of them hydrogen), may be prepared.[3]



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(1) (a) G. Piutti, *Gazz. chim. ital.*, **18**, 480 (1888); (b) O. Lutz, *Ber.*, **62B**, 1879 (1929); (c) D. C. Carpenter, *et al.*, THIS JOURNAL, **64**, 2899 (1942); (d) F. E. King and D. A. A. Kidd, *J. Chem. Soc.*, 2976 (1951); (e) P. Desnuelle and G. Bonjour, *Biochim. Biophys. Acta*, **9**, 356 (1952).

(2) Y. Liwschitz and A. Zilkha, THIS JOURNAL, **77**, 1265 (1955).

(3) Substances for which R = H and R_1 = alkyl are dealt with in this paper, those for which R = R_1 = alkyl in a forthcoming one.