Bull. Inst. Chem. Res., Kyoto Univ. Vol. 50, No. 3, 1972

Synthesis of L-β-Chloroalanine Derivative from L-Aspartic Acid[1]

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Received April 28, 1972

β-Haloalanine and its derivatives are more valuable and useful intermediates for the preparation of the other useful amino acids. In an example, from the chloroalanine derivative and the peptide, cysteine derivatives[3] and cysteine peptide[4] were recently prepared.

Though several reports on the synthesis of β-chloroalanine from alanine[5] by the action of chlorine and from serine[3] were demonstrated in more recent years, a method described by Yoneda *et al.*[6] from optically active N-tosyl aspargine which is the readily available and abundant as a material was unique. The method consisted of the Hofmann reaction of N-haloamide and subsequent diazotization gave a considerably high yield, although the method would not be practical because of a disadvantage due to the difficult cleavage of N-protecting tosyl group. In most recent, the convenient removal method of the tosyl group in amino acids by an electrolysis was found.[7] However, in the case of L-N-tosyl-β-chloroalanine, the resulting amino acid was observed as L-alanine.[8]

Accordingly, should a real practicable method of the preparation of the chloroalanine derivative be developed, it would possibly serve for the synthesis of a lot of intermediates of the other amino acid synthesis on a large scale.

In the present paper, we wish to report the practical method of the synthesis of L-β-chloroalanine derivative from L-aspartic acid *via* Curtius rearrangement and subsequent diazotization as shown in the following scheme.

Benzoylation of the β-ester of aspartic acid (I) was carried out in good yield by the use of magnesium oxide as the base. The hydrazide (III) was obtained from II and hydrazine hydrate at low temperature in high yield by the usual method.[9] The azide (XI) was given from the hydrazide and sodium nitrite at low temperature in usual procedure.[9] When the azide was dissolved in the various solvents, N-benzoyl aspartic acid anhydride (XII) which is unnecessary for the chloroalanine formation was obtained immediately as main product at any temperature. The anhydride was identical with a specimen derived from benzoyl aspartic acid and acetic anhydride. To avoid the formation of the anhydride, the hydrazide (III) was esterified with thionyl chloride and methanol at low temperature in high yield. The ester hydrazide (IV) was also readily converted to the ester azide (V) at low temperature in the presence of

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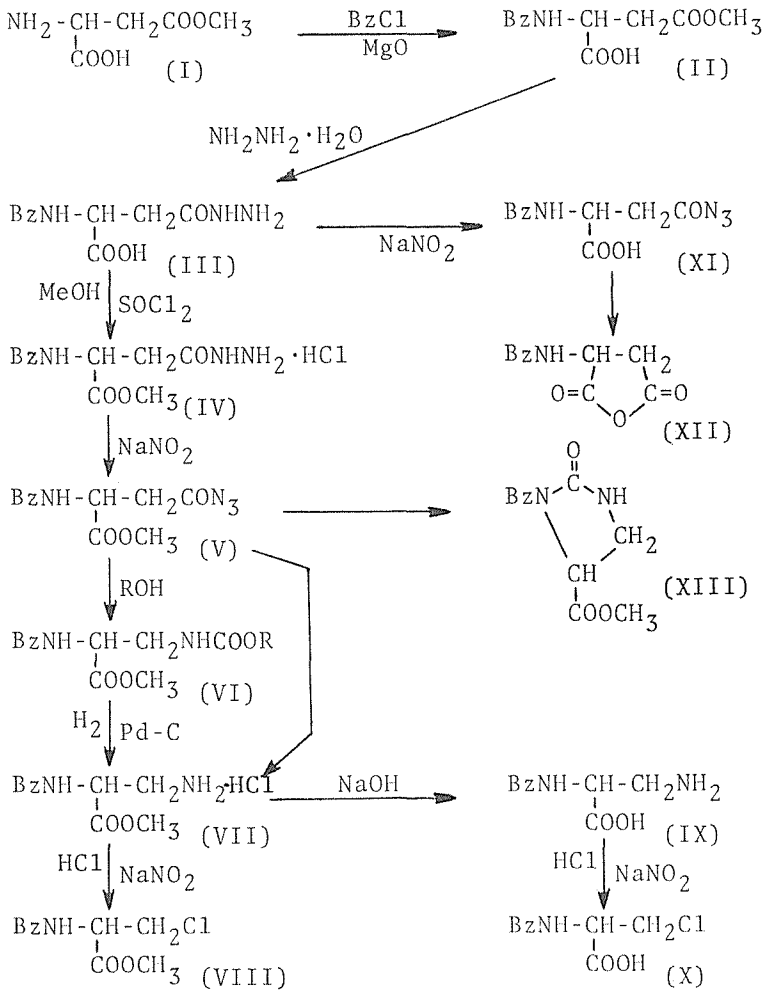

Bz=Benzoyl
R=Benzyl, t-Butyl, t-Amyl

Reaction Scheme

hydrochloric acid. The ester azide did not give the anhydride under the same condition mentioned above. However, the azide was readily led to form methyl 1-benzoyl-2-oxo-imidazolidone-5-carboxylate (XIII) in non-reactive solvent such as benzene at any temperature shown in Table 1. The imidazolidone was identified by spectroscopy and elemental analysis. Namely, the compound showed an infrared absorption at 1750 cm^{-1} which is characteristic of the imidazolidone. NMR spectrum gave a reasonable pattern for the imidazolidone compound to show no proton at N-benzoyl group. In chemical evidence, the imidazolidone compound (XIII) obtained was hydrolyzed with hydrochloric acid to give diamino propionic acid which is identical with a sample prepared from aspartic acid by the Schmidt reaction.[10] In this reaction mechanism, the isocyanate which must be intermediate as soon as formed from the

azide would attack the nitrogen atom of the N-benzoyl residue because of the electron withdrawing effect of the carbon atom of the isocyanate group.

When an alcohol was added to the ester azide (V) at appropriate temperature, the urethane derivative (VI) was obtained as a predominant product as shown in Table 1. For example, benzyl alcohol afforded β-N-benzyloxycarbonyl (Z) compound which agreed with a sample[11] prepared from L-diamino propionic acid. Effect of the temperature in the Curtius reaction was also investigated as shown in Table 2. In the course

Table 1. Effect of the Solvent in the Curtius Rearrangement of Ester Azide (V) at 85–95°C for 30 min.

Solvent	Yields of products (%)[a]		
	(VI)	(VII)	(XIII)
H_2O	0	trace	80
1%-HCl	0	trace	82
5%-HCl	0	20	50
CH_3COOH	0	trace	72
38%-CH_3COOH	0	trace	70
MeOH	—	20	50
5%-MeOH-HCl	trace	41	trace
Benzene	0	trace	80
BzOH[b]	56	13	trace
t-BuOH[c]	26	21	23
t-AmOH[d]	18	32	12

a) Isolated yield. The separation of VI and XIII was carried out by column chromatography (silica gel-$CHCl_3$). b) Benzyl alcohol. c) *t*-Butyl alcohol. d) *t*-Amyl alcohol.

Table 2. Effect of Temperature in the Curtius Reaction of Azide (V) in Benzyl Alcohol (A) and *t*-Butyl Alcohol (B).[a]

A		B	
Temp. (°C)	Yield of VII (%)[b]	Temp. (°C)	Yield of VII (%)[b]
40–50	33	40–45	15
50–60	41	45–55	30
65–75	45	55–65	36
75–85	61	65–75	49
85–95	71	75–82	40
95–110	40	85–90	39

a). The reaction was carried out for 30 min. b). Reaction yield was represented as the product yield of VII, because of difficult isolation of the urethane derivative (VI). Z compound (A) and BOC compound (B) were hydrogenolyzed over Pd-C and hydrolyzed with MeOH-HCl to obtain VII respectively.

of the reaction, β-amino compound (VII) was also partly observed because of the fission of the urethane derivative by heat. The β-amino compound was concordant with the product derived from the β-N-Z compound (VI) by the hydrogenolysis over palladium on charcoal. When *t*-butyl alcohol and *t*-amyl alcohol were used, β-N-*t*-butyloxycarbonyl (BOC) and β-N-*t*-amyloxycarbonyl (AOC) as urethane type com-

pound were introduced under same condition as above, in which the BOC and AOC groups were readily eliminated to give the β-amino acid derivative (VII) in the presence of methanolic hydrochloric acid. The β-amino compound was converted to the corresponding β-chloro compound by the diazotization with sodium nitrite in the presence of potassium chloride in hydrochloric acid solution. In the case of α-N-benzoyl-β-amino propionic acid methyl ester (VII), satisfactory result was not given in the diazotization as shown in Table 3. However, in the case of the diazotization of α-N-benzoyl-β-amino propionic acid (IX), α-N-benzoyl-β-chloroalanine was obtained in reasonable yield even in the same condition as above. (Table 4). These compounds were easily hydrolyzed with hydrochloric acid or hydrobromic acid to afford L-serine according to a method reported by Baganz and Dransch.[12] Therefore, this synthetic method is also employed for the preparation of L-serine.

Table 3. Diazotization of α-N-Benzoyl-β-Amino Propionic Acid Methyl Ester (VII)[a]

VII	HCl		$NaNO_2$	KCl	Temp.	Yield[b]
(g)	(%)	(ml)	(g)	(g)	(°C)	(%)
2.58	25	20	1.0	2.0	5–9	21
2.58	20	20	1.0	2.0	5–10	18
2.58	20	20	1.5	1.0	15–23	29
2.58	20	20	1.0	2.0	25–30	43
2.58	20	20	2.0	2.0	30–46	28

a) Reaction time; 45 min.
b) The product was separated from the serine derivative by the use of silica gel column.

Table 4. Diazotization of α-N-Benzoyl-β-Amino Propionic Acid (IX)[a]

IX	HCl		$NaNO_2$	KCl	Temp.	Yield[b]
(g)	(%)	(ml)	(g)	(g)	(°C)	(%)
1.0	25	25	1.0	2.0	10–20	50
1.0	25	25	1.0	2.0	25–30	57
1.3	20	20	3.0	1.0	30–40	70
2.1	20	20	5.0	3.5	35–40	76
3.0	20	30	5.0	5.0	40–50	55

a) Reaction time; 45 min.
b) The product was isolated by column chromatography using silica gel $-CHCl_3$

EXPERIMENTAL

All melting points are uncorrected. The IR spectra were recorded on a Hitachi EPI-S2 spectrophotometer. The NMR spectra were determined with a Hitachi-Perkin-Elmer R-20 at 60 MHz, using tetramethylsilane as reference. Optical rotation was measured by the use of Parkin-Elmer 141 polarimeter.

L-Aspartic Acid β-Methyl Ester Hydrochloride (I). β-Methyl ester was prepared from L-aspartic acid and thionyl chloride in methanol according to the method described by Schwarz *et al.*[13] in 87% yield. mp 195–197°C, $[\alpha]_D^{20}$ +16.8° (*c* 3.5, EtOH–H_2O=1:3) (reported mp 191–193°C, $[\alpha]_D^{20}$+21.4° (EtOH–H_2O=1:3).

α-N-Benzoyl-L-Aspartic Acid β-Methyl Ester (II). An 80 g of benzoyl chloride was

added dropwise at 0–5 °C for 1 hr to a vigorously stirring mixture of 92 g of I and 80 g of magnesium oxide in water of 900 ml. After the addition of the reagent was complete, the reaction mixture was warmed to room temperature with continuous stirring for 3 hr. The undissolved substances were filtered off, and the filtrate was washed with ether to remove excess benzoyl chloride and then was acidified with hydrochloric acid. The resulting precipitate was extracted twice with ethyl acetate. The combined ethyl acetate extracts were washed with water and dried over sodium sulfate. The solution was evaporated *in vacuo* to afford colourless crystal which was recrystallized from ethyl acetate-hexane. Yield, 99 g (83%), mp 125–126 °C, $[\alpha]_D^{20}$ −13.3° (*c* 1.6, MeOH). IR (Nujol cm^{-1}): 3280, 1750, 1685, 1640. NMR (d_6-DMSO, ppm): 8.76 (d, 1H, NH), 7.96–7.40 (m, 5H, arom), 4.84 (q, 1H, –CH–), 3.62 (s, 3H, $-OCH_3$), 2.20–2.96 (m, 2H, $-CH_2-$). Found: C, 57.09: H, 5.18; N, 5.53%. Calcd for $C_{12}H_{13}O_5N$: C, 57.37; H, H, 5.22; N, 5.58%.

α-N-Benzoyl-L-Aspartic Acid β-Hydrazide Hydrazine Salt (III). A 50 g of II was dissolved in 150 ml of methanol. To this solution was added 35 g of 99% hydrazine monohydrate below 10 °C. The mixture was allowed to stand at room temperature for 12 hr, and the solvent was evaporated under reduced pressure. To this residue was added ethyl acetate (100 ml) to crystallize. The precipitate was collected by filtration and was recrystallized from methanol to afford the hydrazine salt (III). Yield, 45 g (80%), mp 155–157 °C (dec), $[\alpha]_D^{20}$ +3.3° (c 1.6, H_2O). IR (Nujol, cm^{-1}): 3320, 3260, 3190, 1640, 1605, 1550, 1530, NMR (d_6-DMSO, ppm): 8.18 (d, 1H, HN), 7.90–7.38 (m, 5H, arom), 6.38 (bs, 8H, NH, COOH), 4.5 (q, 1H, –CH–), 2.55 (m, 2H, $-CH_2-$).

α-N-Benzoyl-L-Aspartic Acid Methyl Ester β-Hydrazide Hydrochloride (IV). A 23 g of thionyl chloride was added to a suspension of 28.3 g of III in 300 ml methanol at 0–5 °C for 1 hr under vigorous stirring. After the addition of the reagent was over, the stirring was continued for 2 hr at the same temperature. Hydrazine dihydrochloride separated as white precipitate was filtered off, and ether was added to the filtrate until crystallization occured. The crystals were collected by filtration and washed thoroughly with ether. Yield, 27.1 g (90%), mp 174–176 °C (methanol-ether), $[\alpha]_D^{20}$ −37.9° (c 1.38, MeOH). IR (Nujol, cm^{-1}): 3300, 3150, 2580, 1930, 1755, 1966, 1645. NMR (d_6-DMSO, ppm): 9.02 (d, 1H, HN), 8.2–7.4 (m, 8H, arom, NH), 4.93 (q, 1H, –CH–), 3.68 (s, 3H, OCH_3), 2.9 (d, 2H, $-CH_2-$). Found: C, 47.76; H, 5.30; N, 14.26%. Calcd for $C_{12}H_{16}O_4N_3Cl$: C, 47.38; H, 5.21; N, 14.05%.

α-N-Benzoyl-L-Aspartic Acid Methyl Ester β-Azide (V). IV (15 g) was dissolved in a mixture of 200 ml water containing 15 ml of conc. hydrochloric acid and ether of 100 ml. To this solution was added 5.3 g of sodium nitrite dissolved in water (15 ml) at a moderate rate at −5—0 °C for 30 min with vigorous stirring. The stirring was continued for 1 hr at the same temperature. Crystals appeared were collected by filtration and recrystallized from ethyl acetate-hexane. Yield, 12 g (85%), mp 84–85 °C (dec). IR (Nujol, cm^{-1}): 3280, 2150, 1739, 1725, 1640. Found: C, 51.93; H, 4.55%. Calcd for $C_{12}H_{12}O_4N_4$: C, 52.17; H, 4.38%. Satisfactory assignment was not obtained in the determination of NMR, because the azide would decompose. This compound should be stored below −10 °C to avoid the decomposition.

The azide (V) was easily converted to XIII in non-reactive solvent as listed in

Table 1. A 2.76 g of V was added portionwise to 30 ml of benzene which had already been refluxed. After the evolution of gas ceased, the solvent was evaporated under reduced pressure. The residue was dissolved in ethyl acetate of 10 ml and the allowed to stand overnight. Crystals appeared and were collected by filtration. Yield of the XIII, 1.98 g (80%), mp 154–155°C. IR (Nujol, cm^{-1}): 3260, 1750, 1642, 1600. NMR ($CDCl_3$, ppm): 7.71–7.24 (m, 5H, arom), 6.32 (bs, 1H, NH), 4.92 (q, 1H, –CH–), 3.76 (s, 3H, $-OCH_3$), 3.59–3.10 (m, 2H, $-CH_2-$). Found: C, 58.03; H, 4.88; N, 11.20%. Calcd for $C_{12}H_{12}O_4N_2$: C, 58.03; H, 4.87; N, 11.29%.

XIII was hydrolyzed with 6N hydrochloric acid under reflux to yield diamino propionic acid hydrochloride which was concordant with the authentic specimen.[10]

α-N-Benzoyl-L-Aspartic Acid β-Azide (XI). To a solution of 12.5 g of the hydrazide (III) dissolved in 240 ml of water containing 10 ml of conc. hydrochloric acid and 140 ml of chloroform was added a solution of 3.0 g of sodium nitrite dissolved in 10 ml of water below 0°C in the same way as above. Resulting crystals were collected and immediately dried up *in vacuo* at low temperature. Yield, 11.2 g (85%), mp 155–157°C (dec), IR (Nujol, cm^{-1}): 3300, 2920, 2700, 2300, 2130, 1735, 1715, 1640. The azide dissolved in dioxane was readily converted at 50°C to the anhydride (XII) which showed mp 199–201°C and 1866, 1790 cm^{-1} (C=O) in IR.

α-N-Benzoyl-β-Aminopropionic Acid Methyl Ester Hydrochloride (VI, R=Benzyl). Benzyl alcohol (20 ml) was heated at 80°C. To this was carefully added crystalline azide (V) of 5.52 g maintaining the reaction temperature at 85°C to 95°C for 30 min. After the evolution of gas ceased, the solvent was evaporated under reduced pressure. The residue was dissolved in ethyl acetate. The solution was washed with 5% hydrochloric acid and water. The ethyl acetate layer was dried over sodium sulfate and was evaporated under reduced pressure. The resulting oil was well washed with petroleum ether to solidify. The solid residue was crystallized from ether and petroleum ether to afford crystalline VI. Yield, 4.0 g (56%), mp 101–102°C (from CCl_4) $[\alpha]_D^{25}$ −18.0° (*c* 1.6, MeOH). (reported[11] mp 98.5–99.5°C, $[\alpha]_D^{28}$ −17.3° (*c* 1.2, MeOH)). IR (Nujol, cm^{-1}): 1735, 1690, 1635.

This compound (VI) was hydrogenolyzed over palladium on charcoal in methanol containing a small amount of hydrochloric acid to yield α-N-benzoyl-L-diamino propionic acid methyl ester hydrochloride (VII) in quantitative yield. mp 174–176°C (dec), $[\alpha]_D^{25}$ −45.0° (*c* 1.3, MeOH). (reported,[11] mp 171–174°C (dec), $[\alpha]_D^{30}$ −48.3° (MeOH). On the other hand, hydrochloric acid solution separated from ethyl acetate extraction was dried up *in vacuo*. The residue was dissolved in a small amount of methanol. To the methanol solution was added ether in order to crystallize the β-amino compound (VII). Yield, 0.64 g (13%), mp 174–177°C (dec) (from methanol–ether). This agreed with that obtained by the method mentioned above. When *t*-butyl alcohol and *t*-amyl alcohol instead of benzyl alcohol were used, α-N-benzoyl-β-N-*t*-butyloxycarbonyl- and α-N-benzoyl-β-N-*t*-amyloxycarbonyl diamino propionic acid methyl esters which were both oil, isolated by silica gel column chromatography and identified by IR spectra showing 1730, 1687, 1640 cm^{-1} (C=O), were obtained as shown in Table 1.

VII was hydrolyzed with 1N sodium hydroxide solution at 15–20°C overnight to

obtain IX in 85% yield. mp 200–202°C, IR (Nujol, cm^{-1}): 3250, 2050, 1660, 1635. Found: C, 57.48; H, 5.90; N, 13.18%. Calcd for $C^{10}H_{12}O_3N_2$: C, 57.69: H, 5.77; N, 13.46%.

α-N-Benzoyl-β-L-Chloroalanine Methyl Ester (VIII). A typical procedure of the diazotization was as follow: VII (2.58 g) and potassium chloride (2.0 g) were dissolved in 20 ml of 20% hydrochloric acid. To this mixture was added a solution of sodium nitrite (1.5 g) dissolved in 10 ml of water at 25–30°C for 45 min under vigorous stirring in usual diazotization. After addition of nitrite solution, stirring was continued for 1 hr at the same temperature. Reaction mixture was cooled and extracted with ethyl acetate. The ethyl acetate solution was washed with saturated sodium bicarbonate solution and water, and dried over magnesium sulfate. The solvent was evaporated to dryness *in vacuo*. The residue was treated with silica gel (Kieselgel) column and was eluted with chloroform to separate from by-products such as serine derivatives. The fractions including chloro compound were evaporated under reduced pressure. To the residue was added ether to crystallize. The crude crystals were recrystallization from ethanol to obtain pure VIII. Yield, 1.0 g (43%), mp 115–116°C, $[\alpha]_D^{25}$ −12.8° (*c* 1.73, MeOH) (reported[12] mp 114°C), IR (Nujol, cm^{-1}): 3300, 1738, 1625.

VIII was easily converted to L-serine according to a method described by Baganz and Dransch.[12] $[\alpha]_D^{25}$ +14.0° (*c* 3.37, 1N-HCl). (reported, $[\alpha]_D^{20}$ +14.3°).

In the same way, the diazotization of IX was carried out in order to obtain α-N-benzoyl-β-chloroalanine (IX) in various yield shown in Table 4. Mp 134–136°C (H_2O), $[\alpha]_D^{25}$ +14.9° (*c* 2.05, MeOH) IR (Nujol, cm^{-1}); 3290, 1720, 1638, 1610, NMR (d_6-DMSO, ppm): 8.73 (d, 1H, –NH–), 8.0–7.3 (m, 5H, arom), 4.8 (q, 1H, –CH–), 4.05 (d, 2H, $-CH_2-$), Found C, 52.65; H, 4.45; N, 6.21; Cl, 15.82%. Calcd for $C_{10}H_{10}NO_3Cl$: C, 52.74; H, 4.39; N, 6.15; Cl, 15.60%.

ACKNOWLEDGMENT

We wish to express our thanks to Drs. I. Chibata and M. Miyoshi for their encouragement in this series.

REFERENCES

(1) Formation of optically active amino acid. 3. Part 2; This Bulletin, **50**, 220 (1972).
(2) To whom inquiries regarding this paper should be addressed.
(3) T. Furuta and Y. Ishimaru, *Nippon Kagaku Zasshi*, **89**, 716 (1968); **90**, 936 (1969).
(4) M. Wilchek, C. Zioudrou and A. Patchornik, *J. Org. Chem.*, **31**, 2865 (1966).
(5) T. Zaima, K. Mitsuhashi, I. Sasaji and T. Asahara, *Kogyo Kagaku Zasshi*, **73**, 319 (1970).
(6) N. Yoneda, T. Fujii, A. Umeda, H. Yasuo, Y. Taguchi and K. Okumura, *Yakugaku Zasshi*, **89**, 98 (1969).
(7) K. Okumura, T. Iwasaki, M. Matsuoka and K. Matsumoto, *Chem. and Ind.*, 929 (1971).
(8) T. Iwasaki, K. Matsumoto, M. Matsuoka and K. Okumura, in publication.
(9) "Organic Reactions" Vol III. John Wiley & Sons, Inc., New York (1956), p. 337.
(10) T. Kitagawa, T. Ozasa and H. Taniyama, *Yakugaku Zasshi*, **89**, 285 (1969).
(11) A. Kjaer and P. O. Larsen, *Acta. Chem. Scand.* **13**, 1565 (1959).
(12) H. Baganz and G. Dransch, *Chem. Ber.*, **94**, 2597 (1961).
(13) H. Schwarz, F. M. Bumpus and I. H. Pagl, *J. Amer. Chem. Soc.*, **79**, 5697 (1957).