Optical Resolution of Aspartic Acid by Using Copper Complexes of Optically Active Amino Acids*

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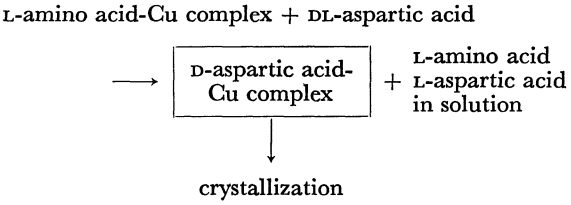
Optical resolution of DL-aspartic acid by using copper complexes of L-alanine, D-alanine, L-glutamic acid, D-glutamic acid, and L-proline in aqueous solution is described. In this investigation the effects of (1) the concentration of reactants and (2) the ratio of the optically active amino acid to racemic aspartic acid in the reaction mixture were studied.

Studies have been made[1,2] on the optical resolution of racemic aspartic acid by the use of enzymatic and also diastereomeric chemical methods. Several methods have been reported for the optical resolution of racemic aspartic acid or a racemic aspartic acid copper complex. Racemic aspartic acid was optically resolved by employing optically active amines[3] and by seeding a supersaturated solution of DL-aspartic acid with ammonium formate.[4,5] Racemic aspartic acid copper complex was resolved easily by seeding with either an L- or D-aspartic acid copper complex.[6] Racemic aspartic acid copper complex was partially resolved by using biopolymers such as cotton or wool.[7] It was found that a supersaturated solution of racemic aspartic acid copper complex was spontaneously resolved into partially optically active L-aspartic acid copper complex.[8] Several investigations of the utilization of metal chelate compounds for optical resolution of racemic amino acids have been reported.[9,10,11]

The optical resolution of DL-aspartic acid by adding a solution of DL-aspartic acid to solutions of the copper complexes of D- or L-alanine, D- or L-glutamic acid, or L-proline was reported.[12] The mixture was allowed to stand for 24 hr to crystallize the aspartic acid copper complex. The optically active aspartic acid copper complex that crystallized had a configuration opposite to the optically active amino acid used in the reaction. This simple procedure for the resolution of racemic aspartic acid yielded 90—100% optically active D- or L-aspartic acid. The phenomenon was explained as a stereoselective ligand exchange reaction.[12] This paper reports the effects of (1) the concentration of reactants and (2) the ratio of the optically active amino acid to racemic aspartic acid on the resolution of racemic aspartic acid.



L-Alanine (0.01, 0.02, and 0.03 mol) was treated with copper carbonate for preparation of the solution (50 m*l*) of L-alanine copper complex. Racemic aspartic acid (0.01, 0.02, and 0.03 mol) in 100 m*l* of water was added to each of the solutions of L-alanine copper complex. Since the crystallization of the aspartic acid copper complex depended on the composition and concentration of the reaction mixture, the crystallization was carried out from several hours to 24 hr. The crystals were then collected by filtration (1st crop). The filtrate was allowed to stand longer (24—72 hr) for the 2nd crop of crystals of the aspartic acid copper complex to grow. The results of resolution of racemic aspartic acid by using L-alanine copper complex are summarized in Table 1.

We see that in reactions A-1, B-2, and C-3, the total amount of aspartic acid copper complex increased as the concentration of the reaction mixture was increased. Figure 1 shows the amount of crystalline aspartic acid copper complex normalized to 0.01 mole racemic aspartic acid. For the 1st crop of crystals, the maximum crystallization occurred when the molar ratio of L-alanine to DL-aspartic acid was about 1.5. The total amount of crystalline complex (1st and 2nd crops) was almost constant when the molar ratio of L-alanine to DL-aspartic acid exceeded 1.5. Figure 2 shows the optical purity of aspartic acid obtained from the 1st and 2nd crops as a function of the molar ratio of L-alanine to DL-aspartic acid. The aspartic acid from the 1st crop was always of the D-configuration; whereas, the configuration of aspartic acid from the 2nd crop depended upon the molar ratio of L-alanine to DL-aspartic acid. When the molar ratio was less than 1, the 2nd crop of crystals had a D-configuration as in the 1st crop. On the other hand, when the molar ratio was greater than 1, the 2nd crop of crystals had an L-configuration. The sharp inversion of configuration in the 2nd crop of

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TABLE 1. OPTICAL RESOLUTION OF DL-ASPARTIC ACID BY THE USE OF L-ALANINE COPPER COMPLEX

Reactions		L-Ala (mol) in 50 ml	DL-Asp (mol) in 100 ml	Optically active aspartic acid copper complex			Optically active aspartic acid	
				Crop	Weight (g)	Weight (g) normalized to 0.01 mol aspartic acid	Confign.	Optical purity (%)
A	1	0.01	0.01	1st	0.48	0.48	D	80
				2nd	0.51	0.51	L	33
				Total	0.99	0.99		
	2	0.01	0.02	1st	0.04	0.02	D	100
				2nd	0.62	0.31	D	96
				Total	0.66	0.33		
	3	0.01	0.03	1st	0.06	0.02	D	62
				2nd	0.60	0.20	D	31
				Total	0.66	0.22		
B	1	0.02	0.01	1st	0.87	0.87	D	84
				2nd	0.84	0.84	L	77
				Total	1.71	1.71		
	2	0.02	0.02	1st	1.06	0.53	D	80
				2nd	1.05	0.52	L	62
				Total	2.11	1.05		
	3	0.02	0.03	1st	0.24	0.12	D	74
				2nd	1.43	0.71	D	100
				Total	1.67	0.83		
C	1	0.03	0.01	1st	0.74	0.74	D	90
				2nd	1.13	1.13	L	40
				Total	1.87	1.87		
	2	0.03	0.02	1st	2.23	1.11	D	74
				2nd	1.11	0.56	L	97
				Total	3.34	1.67		
	3	0.03	0.03	1st	1.72	0.57	D	89
				2nd	1.40	0.47	L	98
				Total	3.12	1.04		

crystals is interesting and might be understood by taking into account the amount of DL-aspartic acid in the reaction mixture.

Figure 3 shows the results of a similar optical resolution of DL-aspartic acid by using L-proline copper complex. The amount of crystalline aspartic acid copper complex is normalized to 0.01 mole racemic aspartic acid. The curves are similar to those in Fig. 1. The 1st crop shows a maximum at a molar ratio of about 2. The total amount of crystalline complex reaches a plateau at a molar ratio of 2. Figure 4 shows the optical purity of aspartic acid obtained from the 1st and 2nd crops as a function of the molar ratio of L-proline to DL-aspartic acid. The aspartic acid from the 1st crop always had a D-configuration; whereas, the aspartic acid from the 2nd crop had a D-configura-

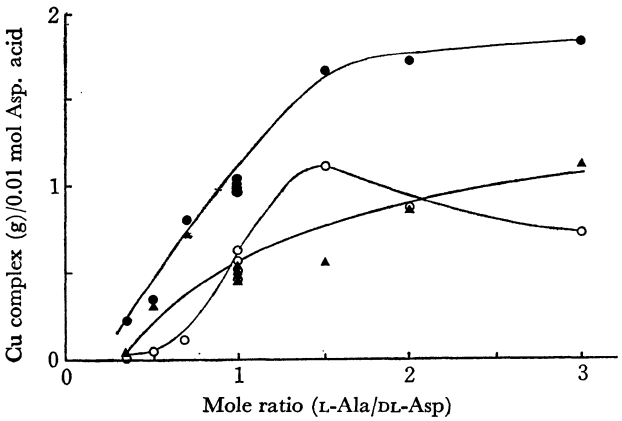

Fig. 1. Weight of crystallized aspartic acid copper complex *versus* molar ratio (L-Ala/DL-Asp).
▲ Aspartic acid copper complex (1st crop)
○ Aspartic acid copper complex (2nd crop)
● Total yield (1st+2nd)

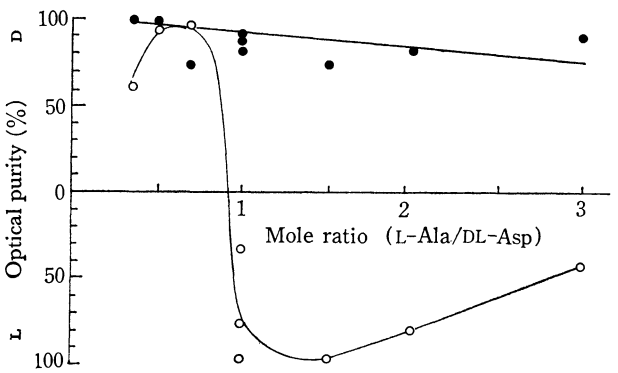

Fig. 2. Optical purity of aspartic acid *versus* molar ratio (L-Ala/DL-Asp).
●: Optical purity of aspartic acid (1st crop)
○: Optical purity of aspartic acid (2nd crop)

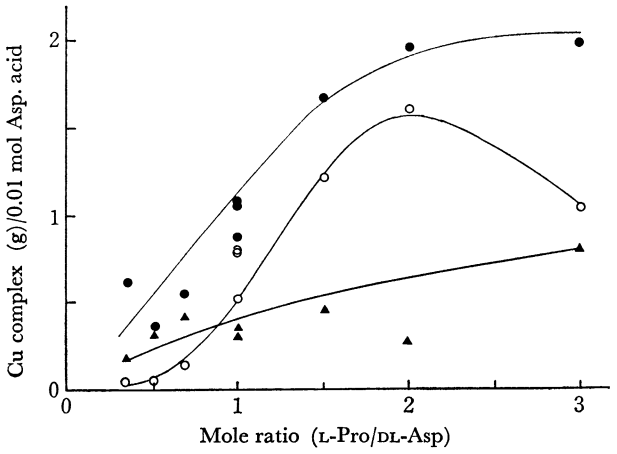

Fig. 3. Weight of crystallized aspartic acid copper complex *versus* molar ratio (L-Pro/DL-Asp).
▲: Aspartic acid copper complex (1st crop)
○: Aspartic acid copper complex (2nd crop)
●: Total yield (1st+2nd)

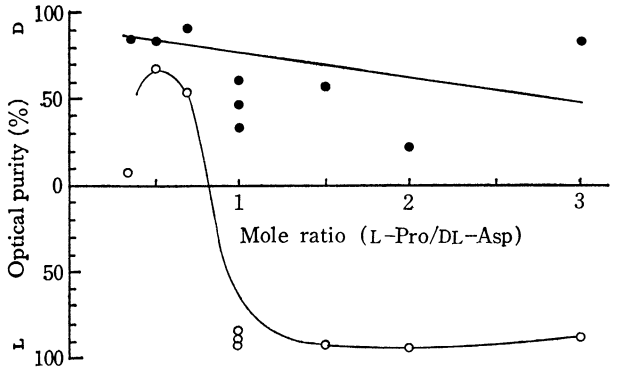

Fig. 4. Optical purity of aspartic acid *versus* molar ratio (L-Pro/DL-Asp).
●: Optical purity of aspartic acid (1st crop)
○: Optical purity of aspartic acid (2nd crop)

tion when the molar ratio of L-proline to DL-aspartic acid was less than 0.9. The aspartic acid had an L-configuration when the molar ratio was greater than 1. The curves are similar to those in Fig. 2. Figure 5 shows the amount of crystalline aspartic acid copper complex and optical purity of aspartic acid obtained by prolonged crystallization for up to 7 days. In this experiment, the molar ratio of L-proline to DL-aspartic acid was 1. As the degree of crystallization increased, the optical purity of the aspartic acid decreased rapidly.

Figure 6 shows the results of the optical resolution of DL-aspartic acid by using L- or D-glutamic acid copper complex. Optically active glutamic acid copper complex gave the highest stereospecific resolution of DL-aspartic acid. Figure 6a shows the optical purity of aspartic acid obtained from the 1st and 2nd crops as a function of the molar ratio of L-glutamic acid to DL-aspartic acid. The aspartic acid from the first crop shows a maximum optical purity when the molar ratio of L-glutamic acid to DL-aspartic acid was 1. In comparison, the optical purity of the aspartic acid from the 2nd crop is high throughout. Figure 6b shows almost the same results by using D-glutamic acid.

The optical resolution of racemic aspartic acid using several copper complexes of optically active amino acids was explained by a stereoselective ligand exchange mechanism and subsequent preferential crystallization.[9] However, attempts to observe the possible stereoselective ligand exchange by the absorption spectrum and the mutarotation of the reaction mixture were unsuccessful. These observations together with the rapid rate of ligand exchange in copper complexes suggest that the proposed stereoselective ligand exchange may not represent the mechanism of these reactions.

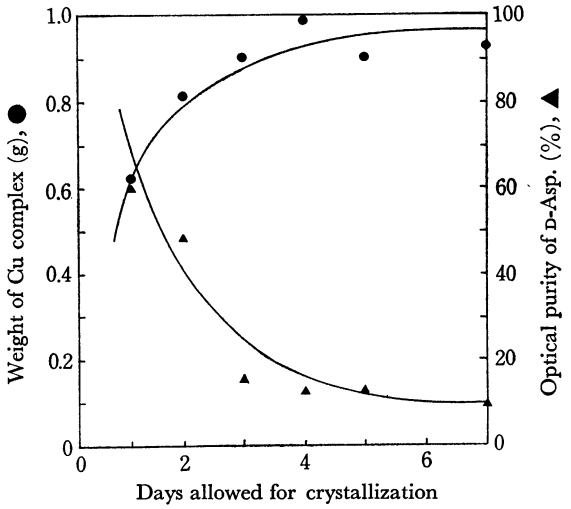

Fig. 5. Weight of aspartic acid copper complex and optical purity of aspartic acid *versus* days allowed for crystallization. (Cu complex prepared from 0.01 mol L-Pro and 0.01 mol DL-Asp in 150 m*l* H_2O)

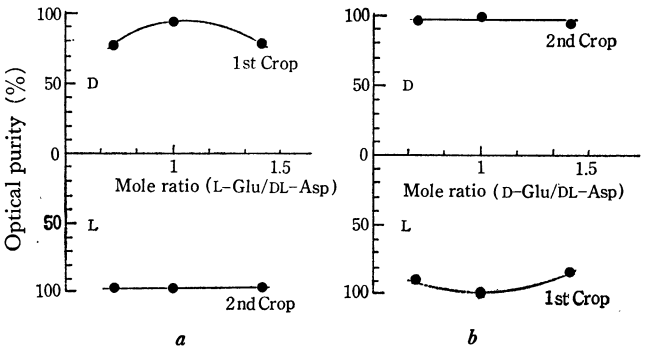

Fig. 6. Optical purity of aspartic acid *versus* mole ratio; a: L-glutamic acid copper complex was used; b: D-glutamic acid copper complex was used.

Experimental

Copper Complex of Optically Active Aspartic Acid (Reaction A-1). L-Alanine (0.89 g, 0.01 mol; Mann Research Laboratories, Inc.; $[\alpha]_D^{25}=+14.3°$ in 6N HCl) was boiled gently with 5.0 g of copper carbonate (Baker Analytical Grade) in 50 m*l* of water for five minutes. The excess of copper carbonate was removed by filtration under reduced pressure. DL-Aspartic acid (1.33 g, 0.01 mol; Mann assayed grade) in 100 m*l* of hot water was added through filter paper to the filtrate. The solution was kept at room temperature for 1 day without agitation. The crystallized D-aspartic acid copper complex, 0.48 g, was collected by filtration. The filtrate was allowed

to stand an additional two days for further crystallization. L-Rich aspartic acid copper complex, 0.51 g, was obtained.

Other amino acids used were L-proline ($[\alpha]_D^{20}=-51.8°$, 0.5N HCl), L-glutamic acid ($[\alpha]_D^{20}=+31.5°$, 6N HCl), and D-glutamic acid ($[\alpha]_{20}^{D}=-31.5°$, 6N HCl).

Preparation of Free Aspartic Acid. The optically active aspartic acid copper complexes were dissolved by heating in 30 to 50 m*l* of 7% acetic acid. The copper ion in each solution was removed by precipitation with hydrogen sulfide. After the precipitation was complete, a small amount of activated charcoal was added to the solution and the solution was cooled to room temperature. The mixture was then filtered and the filtrate was evaporated to dryness under reduced pressure. The aspartic acid was weighed and its specific rotation was measured without further purification.

Some of the optically active preparations of aspartic acid contained a trace to a small amount of the other optically active amino acid used in the resolution reaction (L-ala, L-pro, L- and D-glu). The impurities were detected by paper chromatography. The small amounts of impurities could be eliminated easily by one recrystallization from 25 volumes of water. The optical purity of aspartic acid was measured without recrystallization, since such a purification procedure would cause considerable fractionation of partially optically active aspartic acid. All the optical rotation measurements were carried out by the use of a Rudolph Model 80 polarimeter with a PEC-101 photometer at 25°C using a 10 cm cell.

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