BULLETIN OF THE CHEMICAL SOCIETY OF JAPAN, VOL. 46, 2611—2612 (1973)

The Synthesis of the Salts of L-Aspartic Anhydride with Alkylsulfuric Acid and Their Use in the Preparation of Aspartylphenylalanine Methyl Ester

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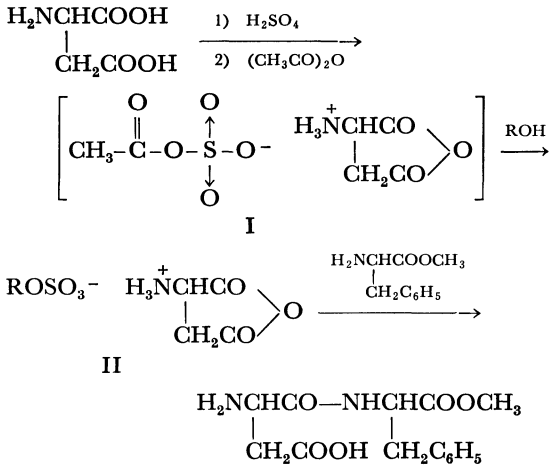
(Received January 25, 1973)

In our previous paper,[1] α-L-aspartyl-L-phenylalanine methyl ester (α-APM), which has a sweet taste,[2] has been conveniently prepared by the condensation of L-aspartic anhydride hydrochloride with methyl L-phenylalaninate, followed by the separation of α-APM from a mixture of α- and β-APM. In the present paper, we will describe the synthesis of the salts of L-aspartic anhydride with alkylsulfuric acid and their use in the preparation of α-APM.

By bringing L-aspartic acid into contact with an excess molar amount of concentrated sulfuric acid in ethyl acetate, we obtained L-aspartic acid sulfate, which, on dehydration with two molecular equivalents of acetic anhydride in ethyl acetate, gave the corresponding anhydride (I). Although the anhydride (I) could not be isolated, its structure was assumed to be the salt of L-aspartic anhydride with mixed acetic-sulfuric anhydride (I). The treatment of the anhydride (I) with an equimolar amount of an alcohol, such as methanol, 2-propanol, or cyclohexanol, gave the salts of L-aspartic anhydride with alkylsulfuric acid (II) (Table 1). The reaction of the anhydride (I) with an alcohol took place exclusively at the sulfuric acid moiety of I. The results of the elemental analyses of the reaction products agreed with the values calculated for the anhydrides (II) (Table 1). All of the salts of L-aspartic anhydride with alkylsulfuric acid were practically stable and were used successfully in the preparation of α-APM. In order to increase the formation of the desired α-isomer, the reaction conditions, such as the temperature, the solvent effects, and the molar ratios of both reactants, were investigated. It was found that the reaction could be effected by adding carbon dioxide, by employing an excess of methyl L-phenylalaninate, and by conducting it at a low temperature.



The treatment of L-aspartic anhydride isopropylsulfate with 4 molecular equivalents of methyl L-phenylalaninate in the presence of carbon dioxide in ethylene dichloride at −20 °C gave a mixture of α- and β-APM, from which α-APM was isolated in a 45% yield by separation through its hydrochloride salt, followed by neutralization and subsequent column chromatography.

Experimental

All the melting points are uncorrected. The determination of α- and β-APM by paper electrophoresis was carried out according to the method described in a previous paper.[3] The IR spectra were recorded in Nujol mull with a Jasco IR-S spectrometer. The NMR spectra were obtained with a Varian T-60 spectrometer at 60 MHz; the chemical shifts are given from sodium 2,2-dimethyl-2-silapentane-5-sulfonate, used as the internal reference.

L-Aspartic Acid Sulfate. A mixture of L-aspartic acid (60 g), concentrated sulfuric acid (52.5 ml), and ethyl acetate (200 ml) was stirred for 48 hr at room temperature. The crystals were then collected by filtration and dried *in vacuo* over phosphorus pentoxide; yield, 99 g (95%), mp 151—152 °C. Found: C, 20.59; H, 3.87; N, 6.00; S, 13.71%. Calcd for $C_4H_7O_4N \cdot H_2SO_4$: C, 20.78; H, 3.80; N, 6.06; S, 13.85%.

The Salts of L-Aspartic Anhydride with Alkylsulfuric Acid. In a typical procedure (No 1 in Table 1), a mixture of L-aspartic acid sulfate (11.55 g), ethyl acetate (10 ml), acetic

TABLE 1. THE SALTS OF L-ASPARTIC ANHYDRIDE WITH ALKYLSULFURIC ACID(II)

No.	R	Yield (%)	Mp °C	Found (%)				Calcd (%)				IR cm^{-1}	
				C	H	N	S	C	H	N	S		
1	CH_3–	93	105—109[a]	26.13	4.24	6.13	13.91	26.43	3.96	6.17	14.10	1820	1890
2	$(CH_3)_2CH$–	82	98—101.5	32.64	5.32	5.23	12.42	32.94	5.10	5.49	12.55	1800	1885
3	C_6H_{11}–	90	95—100[b]	40.52	5.94	4.80	10.84	40.68	5.76	4.75	10.85	1810	1890

a) Partly melted at 96°C. b) Partly melted at 67°C.

1) Y. Ariyoshi, T. Yamatani, N. Uchiyama, Y. Adachi, and N. Sato, This Bulletin, **46**, 1893 (1973).

2) R. H. Mazur, J. M. Schlatter, and A. H. Goldkamp, *J. Amer. Chem. Soc.*, **91**, 2684 (1969).

3) Y. Ariyoshi and N. Sato, This Bulletin, **45**, 942 (1972).

anhydride (12.8 g), and concentrated sulfuric acid (0.5 g) was stirred for 4 hr at room temperature. Anhydrous methanol (24 ml) was then added to the solution with stirring and cooling in an ice-bath. After stirring for 30 min in an ice-bath, the crystals thus formed were collected by filtration, washed with anhydrous ethyl acetate, and dried *in vacuo* over phosphorus pentoxide and sodium hydroxide pellets; yield, 10.5 g (93%); $[\alpha]_D^{26}$ +15.3° (*c* 2, 6 M HCl); NMR (D_2O, DSS): δ 3.22(d, 2H, J=5.5 Hz), 3.80(s, 3H), 4.50 (t, 1H, J=5.5 Hz).

The data are given in Table 1.

Synthesis of α-APM. Carbon dioxide (8.8 g) was introduced into a solution of methyl L-phenylalaninate (71.6 g) in ethylene dichloride (750 ml) containing 8 ml of methanol at −10 °C. To the stirred solution, L-aspartic anhydride isopropylsulfate (22.7 g) was added, portion by portion, at −20 °C. After stirring for 30 min at this temperature, hot water (300 ml, 70—80 °C) and sodium carbonate (6.3 g) were successively added to the solution. After the removal of the excess of methyl L-phenylalaninate by two extractions with 150-ml portions of ethylene dichloride, the aqueous layer was acidified to pH 3 with dilute hydrochloric acid. The solution was found to contain 20.3 g (61%) of α-APM and 5.8 g (19%) of β-APM by paper electrophoresis. The solution was concentrated *in vacuo* to 135 ml and kept in a refrigerator overnight to give crystals (α-APM·HCl, 20 g, 55%), which were then dissolved in water (150 ml). The solution was adjusted to pH 4.8 with a 5% aqueous sodium carbonate solution while being stirred at 50 °C, and then it was kept in a refrigerator overnight to give crystals (α-APM, 14 g, 46%). The crystals were dissolved in water (500 ml) and passed through a column (1×20 cm) of Dowex 1×4 (acetate form) while being maintained at 45 °C, and then the column was washed with water (20 ml). The eluate and washings were concentrated *in vacuo* to give α-APM, which was recrystallized from water; yield, 12.6 g (42%); mp 235—236 °C (decomp.); $[\alpha]_D^{26}$ +31.5° (*c* 1, acetic acid). lit,[1] mp 235—236 °C; $[\alpha]_D$ +32° (*c*, 1, acetic acid); lit,[2] mp 190 °C and 246—247 °C. lit,[4] mp 245—247 °C. Found: C, 55.46; H, 6.24; N, 8.96%. Calcd for $C_{14}H_{18}O_5N_2 \cdot 1/2H_2O$: C, 55.44; H, 6.31; N, 9.24%.

4) J. M. Davey, A. H. Laird, and J. S. Morley, *J. Chem. Soc., C*, **1966**, 555.