

Thermal Analysis of Aspartic Acid and Its Polymers

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Aspartic acid is one of the proteinaceous acidic amino acids, has two carboxyl groups per molecule. Together, these two different carboxyl groups play an important role in thermal reactions. When aspartic acid is heated at elevated temperatures (up to 400 °C), it polymerizes to form polyaspartic acid. This reaction does not proceed on heating glutamic acid, which is another acidic amino acid. Present research used both thermal analysis and infrared measurements to examine the heating behaviour of aspartic acid over a given time period. The measurements reveal some aspects of the thermal reaction mechanism. During the heating reactions, amide bonds gradually form and then imide bond formation follows. This work may be useful for clarifying the mechanism of thermal polycondensation of aspartic acid and the structural features of the products.

Key Words: Thermal analysis, Aspartic acid, Polypeptide.

INTRODUCTION

Amino acids are known to be bifunctional organic compounds^{1,2} that form intramolecular ion pairs. In nature, amino acids form amide (peptide) bonds in proteins and peptides. *In vivo*, these peptide formation reactions^{1,2} are closely controlled by nucleic acids and enzymes acting as bioorganic peptide-making machinery. Such elaborate products, proteins and peptides require very sophisticated processes to be chemically synthesized *in vitro*. Although amino acids dissolve in water very well, most of the amino acids do not form linear peptides on simple heating under non-aqueous conditions and also under aqueous conditions if they release water. However, both asparagine³ in water and aspartic acid^{4,6} in the absence of water can form peptide bonds to become polypeptides⁵⁻¹¹ or co-polypeptides⁴ on heating.

Polyaspartic acid is a very useful compound in dishwashing detergents, superabsorbent materials, selective membranes, green-scale inhibitors and many other pharmaceutical applications¹². Polyaspartic acid can be synthesized from polymerization using N-carboxyanhydrides¹³, thermal reactions using only aspartic acid^{5,6}, aspartic acid and phosphoric acid¹⁴ or maleic acid and ammonia. The latter thermal reaction is well-known in simulation reactions of the chemical evolutionary

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synthesis of polypeptides^{4,5}. The proposed reaction mechanisms (Fig. 1) of the thermal polymerization of aspartic acid has been suggested to be *via* an anhydride formation pathway and the direct polymerization of aspartic acid. We have used both differential thermal analysis (DTA) and the infrared (IR) spectra of aspartic acid, polyaspartic acid and anhydro-polyaspartic acid to clarify the thermal reaction mechanism.

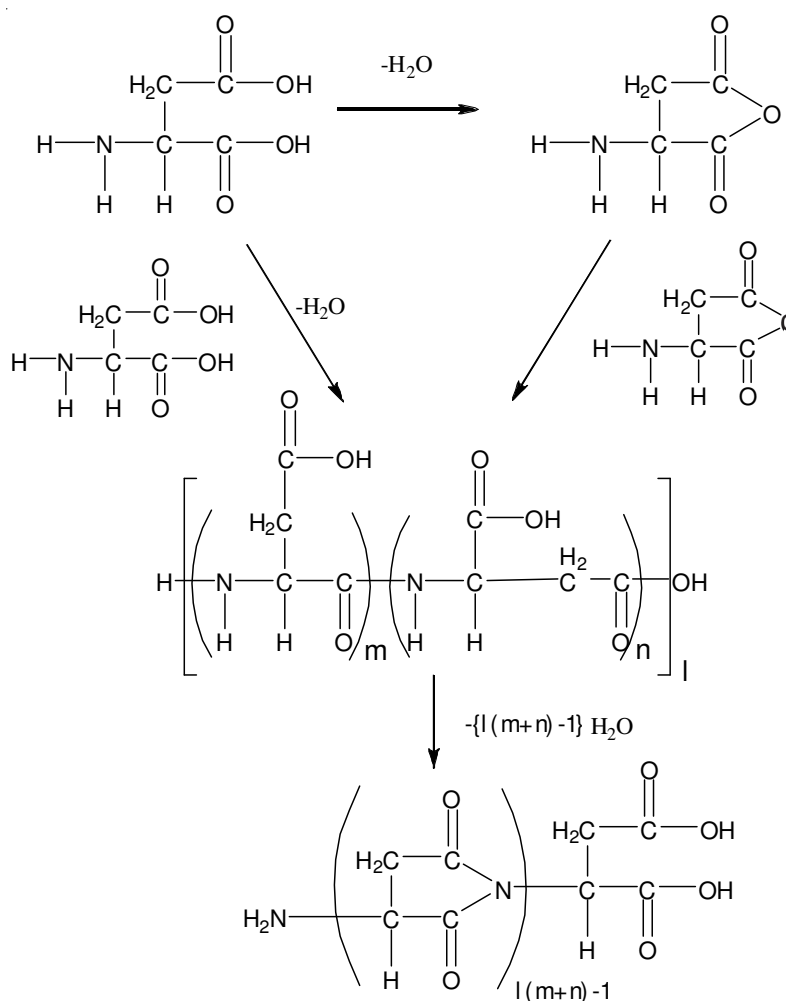


Fig. 1. A proposed mechanism of the thermal reaction of aspartic acid

EXPERIMENTAL

The thermal analyzer used was a Shimadzu DT-40 thermal analyzer (Shimadzu, Kyoto, Japan) and the infrared spectra were recorded using a Hitachi Model 260-50 infrared spectrophotometer (Hitachi, Tokyo, Japan). The DL-aspartic acid used was purchased from Wako Pure Chemical Industries (Japan).

About 5.0 to 15.7 mg samples of DL-aspartic acid were measured to within 0.1 mg and these were placed in the aluminum sample containers of the DTA. The γ -aluminum reference disk was placed in the center of the second aluminum container. The temperature of the heating chamber was programmed from 25-400 °C using a heating rate of 5 °C/min under the flowing nitrogen gas (35 mL/min). The IR spectra of the samples were recorded after each 20 min heating period.

RESULTS AND DISCUSSION

Fig. 2 shows the thermal analysis data of a 15.7 mg DL-aspartic acid sample. Although no change was observed in both the differential thermal analysis and thermogravimetric data at temperatures up to 200 °C [point (a)], the weight loss of the sample proceeded at a very fast rate at temperatures up to 260.1 °C. During this weight loss period, a shoulder was observed at 226.1 °C and this point is consistent with the end point of the first endothermic peak [point (b)]. The end points of two endothermic peaks were detected. It can be seen that a weight loss of 12.8 % of the total weight of the sample occurred from points (a)-(b) and a weight loss of 14.8 % occurred from points (b)-(c). The weight loss of 12.7 % is close to the theoretical weight loss (13.5 %) predicted from the loss of one water molecule from DL aspartic acid. Further, the second weight loss (14.7 %) is also close to the theoretical weight loss expected for the generation of a water molecule. Therefore, the total weight loss (27.5 %) is consistent with the weight loss from two molecules of water (27.1 %).

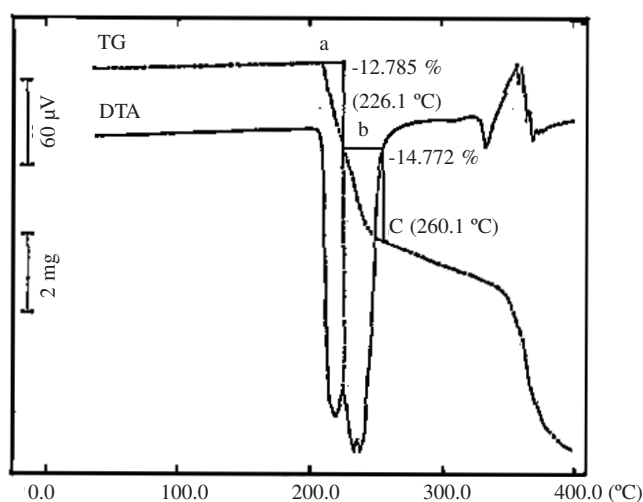


Fig. 2. Thermal analysis of DL-aspartic acid from 25-400 °C using a heating rate of 5 °C/min. DTA: differential thermal analysis, TG: thermogravimetry

Fig. 3 shows the IR spectra of unheated and heated DL-aspartic acid samples. The absorbance in Fig. 3 corresponds to IR spectra of the samples taken from the DTA containers at each temperature. The curves from samples heated to 200 and

220 °C do not show any amide I and II absorbance, but have a similar absorbance to DL-aspartic acid, including the absorbance from the OH group (3200 cm^{-1}). However, an absorbance characteristic of the amide I (1650 cm^{-1}) and amide II (1550 cm^{-1}) groups was observed from a sample heated to 230 °C. This observation suggests that peptide bonds had formed at this temperature. This is consistent with the desorption of a water molecule from an aspartic acid molecule, as shown in Fig. 2. At higher temperatures (240-260 °C), an absorbance due to imide bonds was more pronounced. Such an absorbance arising from imide groups suggests the formation of anhydro-polyaspartic acid. The IR spectra shown in Fig. 3 do not show the formation of the anhydride of aspartic acid. Although aspartic anhydrides can form on heating, these groups would readily react to form a peptide bond. Therefore, no absorbance from the anhydride groups appears in Fig. 3.

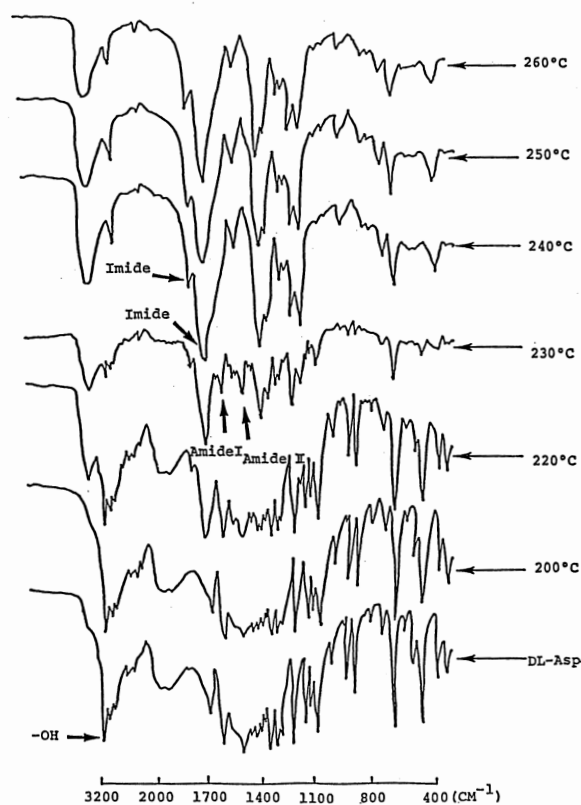


Fig. 3. IR spectra of DL-aspartic acid and heated DL-aspartic acid polymer

Fig. 4 shows the TG and DTA data of DL-aspartic acid heated at a rate of 2 °C/min in the temperature range 25-400 °C. The curve shows two main endothermic peaks that correspond to the first and second desorption of a water molecule. The form of this curve is similar to that obtained using a heating rate of 5 °C/min, as shown in

Fig. 2. However, the two endothermic peaks can be distinguished from each other more clearly than those shown in Fig. 2. This means that the desorption reactions of the two water molecules proceeds in stages.

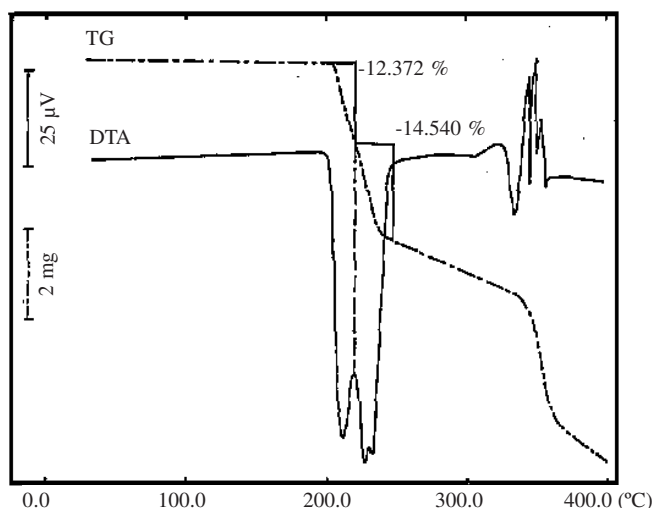


Fig. 4. Thermal analysis of DL-aspartic acid from 25-400 °C using a heating rate of 2 °C/min

Fig. 5 shows the TG and DTA data of DL-aspartic acid heated using a heating rate of 20 °C/min from room temperature to 400 °C. The two endothermic peaks shown in Figs. 3 and 4 were not observed, but a single endothermic peak due to the overlapping of the two peaks was observed. The weight loss until the end point of the endothermic peak was 27.1 %, which corresponds to the loss of two water molecules from a single aspartic acid molecule. Anhydro-polyaspartic acid prepared by heating DL-aspartic acid for a period of 6 h at 200 °C was placed in a measuring cell and reheated from room temperature to 400 °C at a heating rate of 5 °C/min. The first endothermic peak observed was assigned to the release of water molecules absorbed in the crystals of the sample. After this peak, no endothermic peaks were observed in the DTA curve. This observation suggests that neither more water molecules were removed, nor a dehydration reaction proceeded in the anhydro-polyaspartic acid sample used.

The same anhydro-polyaspartic acid sample used in thermal analysis experiments was partly hydrolyzed in sodium bicarbonate to form polyaspartic acid. The resulting sample was lyophilized to give dry polyaspartic acid, which has free carboxyl groups. The free polyaspartic acid was then subjected to thermal analysis, as shown in Fig. 7, which shows a single large endothermic peak, corresponding to the desorption of a water molecule formed when a carboxyl group reacts with an amide NH group to give an imide group. No further endothermic peaks were observed after this event.

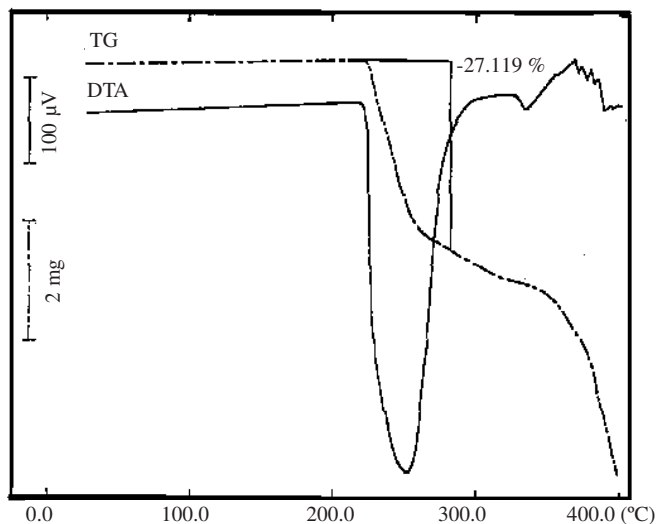


Fig. 5. Thermal analysis of DL-aspartic acid from 25-400 °C using a heating rate of 20 °C/min

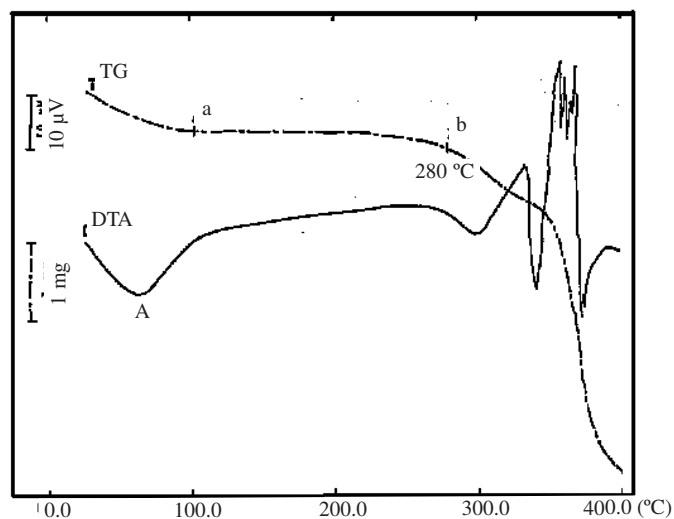


Fig. 6. Thermal analysis of imide-type polyaspartic acid from 25-400 °C using a heating rate of 20 °C/min

The data in Figs. 6 and 7 show that the anhydro-polyaspartic acid passed through a stepwise dehydration of its water molecules. The thermal analysis demonstrates that the dehydration of water molecules proceeded gradually. Present results show the fundamental relationship between the decrease in weight and the appearance of endothermic peaks in the thermal analysis during the thermal reactions. In addition, it also gave us significant information on the mechanism of reactions other than those of aspartic acid.

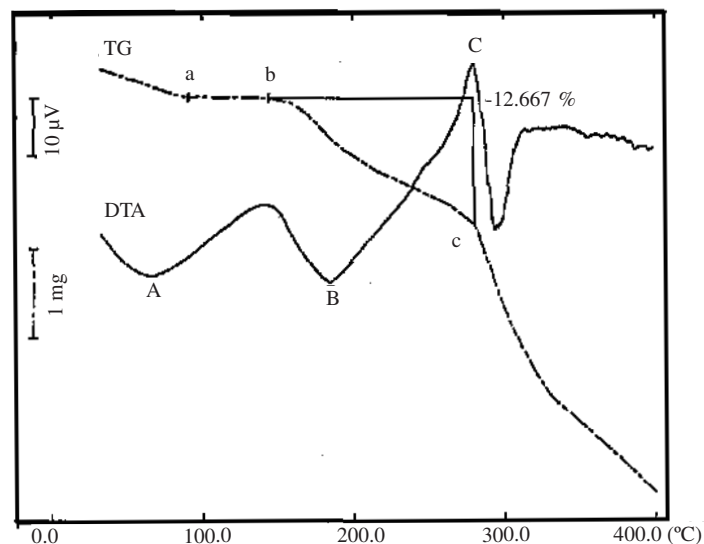


Fig. 7. Thermal analysis of peptide-type polyaspartic acid from 25-400 °C using a heating rate of 20 °C/min

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