Although α-amino acids are readily available substances, many of their transformations and, in particular their polycondensations, have been very little studied. The literature contains only a few reports of experiments on the heating of the amino acids themselves or of their esters with formation of the corresponding polyamides and diketopiperazines.


Polyakova and Vereshchagin [5] observed the influence of pressure on the formation of polypeptide chains. Bresler and Glikin [6] showed that alanine ester in aqueous solution under a pressure of 5000 atm, forms a diketopiperazine, while trialanine ester forms a polymer with a polymerization coefficient of 12-18.

We have studied the polycondensation of the ethyl ester of D,L-alanine in various temperature conditions with special reference to the clarification of the effect of catalysts on this reaction.

We prepared the starting ethyl ester of D,L-alanine by Fischer's method [7] and finally purified it by vacuum distillation in carbon dioxide-free air in the special apparatus described below.

**EXPERIMENTAL PROCEDURE**

We established that carbon dioxide is an extremely active catalyst for the polycondensation of alanine ester, and this necessitated the preparation of the starting material and the performance of the polycondensation reaction itself in conditions ensuring complete absence of carbon dioxide; it was necessary to have a method of exact proportioning of the minute amounts of carbon dioxide employed as catalyst. The polycondensation reaction was therefore performed in sealed ampoules heated in a thermostat at 55 ± 0.1°. The ampoules were filled in the apparatus sketched in Fig. 1 which permitted preparation of carbon dioxide-free ester and the proportioning of small additions of carbon dioxide and other catalysts.

The apparatus consists of a flask A, equipped with a condenser joined through a ground-glass connection with a chamber B, whose side tubes are joined by ground-glass connections to a round-bottomed flask O and special receivers for weighed amounts of ester; the shape of these receivers varies with the nature of the catalyst.

When the catalyst is carbon dioxide the receiver sketched in more detail in Fig. 2 is used, while in the case of other catalysts the simpler receiver illustrated in Fig 3 is used.

When carrying out the polycondensation reaction in presence of carbon dioxide, the gas buret E is first filled with carbon dioxide via the limb F while the tap K is closed and tap L is open. The apparatus is then pumped out with a vacuum pump to a residual pressure of 2-3 mm and again filled with dry carbon dioxide which had been dried over sulfuric acid. The carbon dioxide is derived from chemically pure sodium bicarbonate and sulfuric acid. These operations are repeated six times and dry nitrogen is passed through after filling of the buret with carbon dioxide and closing tap L and limb F. After this the apparatus for proportioning the carbon dioxide is weighed on an analytical balance.

To the difference between the weights is added the weight of the air in the ampoule, and the result is the weight of the ester (usually 0.2-0.4 g). The branch F is then filled with sulfuric acid, with tap K opened,
through tap K is introduced the required amount of carbon dioxide. Tap K is closed and ampoule D is sealed
from the buret E at the point N. A sealed ampoule G is thus obtained which contains a weighed amount of est-
er and a known amount of carbon dioxide as catalyst. The ampoule is at once placed in the thermostat and
heated for the required period.

In experiments carried out with other catalysts, the fitment shown in Fig. 3 is used; into the latter is prev-
iously introduced the required amount of catalyst; then the required amount of ester is distilled into it and the
vessel is sealed off under vacuum at point M.

After the heating, the ampoule is withdrawn from the thermostat, rapidly cooled, and crushed in a small,
strong porcelain beaker to which is previously added 10-20 ml of 3% aqueous acetic acid. The resultant solu-
tion is poured into a measuring flask; the beaker is rinsed out with fresh portions of acetic acid which are also
transferred to the flask; the volume is then made up to the mark with additional 3% acetic acid solution. The
content of free amino groups in the solution is then determined by the Van Slyke method [8].

A series of experiments in which the reaction products were dissolved in water showed that the polyconden-
sation reaction also takes place in aqueous solution, whereas dissolution in acetic dilute acid leads to complete
stoppage of the polycondensation reaction and ensures more correct results. The polyalanine and diketopiperazine
resulting from the reaction dissolve completely in 3% acetic acid as well as in water at any degree of complete-
ness of the process, whereas the polymers obtained by heating D,L-valine, D,L-norleucine or glycine only dissolve
up to the stage of 30-50% completion of the reaction. Their further heating leads to products with ever decrea-
ing solubility in water and dilute acetic acid; their investigation therefore calls for a special technique whereas in the
case of alanine such difficulties do not arise.

Water soluble products can be obtained by simultaneous polycondensation of two amino acid esters. Thus
the separate heating of the ethyl esters of D,L-phenylalanine and glycine leads to water-insoluble polymers, where-
as their joint polycondensation leads to formation of water-soluble products.

EXPERIMENTAL RESULTS

Investigation of the action of various additives on the reaction of polycondensation of the ethyl ester of D,L-
alanine showed that considerable catalytic activity is manifested by acids such as carbonic, hydrochloric, sulfuric,
acetic and propionic acids, as well as by polyalanine and amino-epanthetic acid. Neutral and alkaline substances,
such as diketopiperazine (lactimide), polyalanine ester, water, D,L-alanine, ammonia, diethylamine and KOH, do
not bring about an appreciable increase in reaction velocity. Bearing in mind that esters of amino acids are ther-

Fig. 1

Fig. 2

Fig. 3