XLIII. A NOTE ON THE COMBINATION OF CYSTEINE WITH ALLYL ISOTHIOCYANATE

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IN an earlier paper [Walker, 1925] it is recorded that the sulphydryl group of tissues and muscle protein disappears on treatment with allyl isothiocyanate. The reaction described below was observed in an attempt to discover what happens in these circumstances. For reasons mentioned in the paper quoted it was considered that a direct combination took place between allyl isothiocyanate and the tissue sulphydryl group. Consequently we have examined the reaction between allyl isothiocyanate and cysteine.

EXPERIMENTAL

0.895 g. of pure cysteine hydrochloride was dissolved in a little water and neutralized with 52.05 ml. of *N*/10 NaOH. The equivalent quantity of allyl isothiocyanate (0.554 ml.) was run in and gentle shaking maintained for 10 min., whereupon a white crystalline solid started to separate. The mixture was kept for 2 hours, then filtered and the solid washed with alcohol, followed by ether. 0.565 g. of soft, white, odourless crystals was obtained; yield, 45 %. The material obtained was insoluble in the usual organic solvents and slightly soluble in hot water, but in this case we were suspicious of decomposition. It was soluble in hot 90 % methyl alcohol, but again the high temperature seemed to cause a certain amount of decomposition. The material was never satisfactorily re-crystallized. In the dry state it decomposed in the neighbourhood of 200°.

Another preparation was made in which the reaction was carried out at greater dilution and the product purified by washing thoroughly with water, alcohol and ether respectively. (Found after drying *in vacuo*: C, 38.24, H, 5.44, N, 12.6, S, 25.58 %. Cysteine + allyl isothiocyanate = $C_7H_{12}N_2O_2S_2$, which requires C, 38.11, H, 5.49, N, 12.7, S, 29.11 %.) The N was determined by ourselves, using the micro-Kjeldahl method, and was kindly confirmed by Mr J. R. P. O'Brien by the micro-Dumas method. The remaining analyses were by Schoeller. The substance formed is clearly an addition compound. It is readily soluble in alkali and therefore probably an acid. By analogy with the reaction between isocyanates and amino-acids, addition with rearrangement probably takes place at the amino-group, giving:

The compound can thus be described as α -*N*-allylthiocarbamido- β -thiolpropionic acid. We confirmed the structure by

(1) demonstrating the free sulphydryl group by the nitroprusside test,

(2) showing that no reaction occurs with cysteine in which the amino-group has been blocked by formaldehyde.

We have satisfied ourselves that an analogous reaction occurs with cystine, in alkaline solution; but we have not obtained an adequately pure product.

DISCUSSION

The reaction does not appear to have been described before, although allylthiohydantoin of glycine and alanine [Marckwald *et al.*, 1891] and simple thiohydantoin of cystine and cysteine [Nicolet, 1930] have been described. Aschan [1884] reported failure of reaction between amino-acids and alkyl isothiocyanate; and reactions of a quite different type appear to occur between simple thioacids and isothiocyanates [Wheeler & Merriam, 1901].

It is obvious that the reaction described throws no light on the effect of allyl isothiocyanate on protein sulphydryl, which must be left for further work to solve. We have always borne in mind that the inhibiting influence of allyl isothiocyanate may have been on the mechanism of the nitroprusside reaction rather than on the substrate. In trying to test this view, we found that the nitroprusside reaction for acetone was not affected by allyl isothiocyanate, whilst on the other hand we did observe that the magenta colour obtained in performing the nitroprusside test with sulphydryl compounds was discharged on the addition of allyl isothiocyanate.

SUMMARY

Combination takes place readily between cysteine and allyl isothiocyanate with the formation of α -*N*-allylthiocarbamido- β -thiolpropionic acid.

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