primarily in the fumaric acid and succinic acid, in particular the carboxyl carbon atoms. Although it is not possible to directly correlate the present work and the previous findings with fruit, the intimate relationship between ethylene and the Krebs cycle acids is, nevertheless, indicated by the results in both experiments.

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Occurrence of *Bis*(β-Amino-β-Carboxyethyl) Trisulphide in the Hydrolysates of Wool and other Proteins

Investigations of acid hydrolysates of wool labelled with sulphur-35, and of non-radioactive wool hydrolysed with ^{35}S-cystine, have shown the presence of unidentified sulphur compounds derived from cystine[1,2]. The most abundant of these (I) increased in amount with increasing time of hydrolysis up to 100 hr., when about one-tenth of the original sulphur in wool was present as (I). Somewhat smaller amounts of (I), representing about one-fiftieth of the protein sulphur, were found in acid hydrolysates of insulin, ribonuclease and bovine serum albumin, but none could be detected in hydrolysates of lysozyme and β-lactoglobulin.

We have recently obtained 1·4 gm. of (I) as a stable, white, microcrystalline powder from 150 gm. of wool by the following procedure. The wool was hydrolysed in 1·5 litres of 5·7 *N* hydrochloric acid for 100 hr. under reflux, and the hydrolysate fractionated by chromatography on a 9-cm. × 40-cm. column of 'Zeo-Karb 225' (× 4) resin in the H^+ form by elution with 2·5 *N* hydrochloric acid[3]. (I) was eluted with phenylalanine and tyrosine, and was separated from these by subsequent chromatography on a 4-cm. × 15-cm. column of starch by elution with 2 : 1 (v/v) *n*-propanol/0·5 *N* hydrochloric acid[4]. Traces of contaminating carbohydrate were removed by re-chromatography of (I) on a 2·5-cm. × 5-cm. column of 'Zeo-Karb 225' (× 4) in the H^+ form eluted with 2·5 *N* hydrochloric acid. (I) was isolated from the aqueous solution of its hydrochloride by the addition of pyridine until the *p*H was 4·6.

The properties of (I) have led us to conclude that it is the previously unknown trisulphide analogue of cystine, *bis*(β-amino-β-carboxyethyl) trisulphide, $[HOOC{\cdot}CH(NH_2){\cdot}CH_2]_2S_3$.

Thus, when (I) is oxidized by bromine, with the theoretical uptake of oxygen, cysteic and sulphuric acids are obtained in the molecular proportions of 2 : 1. On treatment with cyanide under the conditions of Sörbo[5] (omitting Cu^{++} ion), one-third of the sulphur of (I) is converted to thiocyanate. Approximately one-third of the sulphur of (I) is released as hydrogen sulphide during reduction with tin or zinc, and hydrochloric acid, and the reduced solution contains cysteine and no other thiol. By 'formol' titration (I) has an equivalent weight of 138 (theoretical 136). Elementary analysis gave C, 27·1; H, 4·4; N, 10·0; S, 36·4 per cent ($C_6H_{12}O_4S_3N_2$ requires C, 26·5; H, 4·4; N, 10·3; S, 35·3 per cent).

The compound is stable in the dry state or in strongly acid solutions, but in neutral or alkaline solutions dismutates to give cystine and other products. When chromatographed on paper in *n*-butanol/acetic acid/water (4 : 1 : 5 v/v), (I) moves as a single spot with R_F 0·09, but analytical amounts (< 0·5 mgm.) chromatographed on 0·9-cm. × 40-cm. columns of 'Zeo-Karb 225' (× 4) resin by the hydrochloric acid elution method are resolved into a double peak, indicating the presence of two stereoisomers.

By refluxing a suspension of *N*-acetylcysteine in chloroform containing sulphur dichloride with constant stirring for 40 hr., distilling off the chloroform, and hydrolysing the residue for 4 hr. in 5 *N* hydrochloric acid we have obtained, in addition to cysteine, cystine and one other unidentified minor product, a substance the chromatographic behaviour of which is identical with that of (I), but which has not yet been further characterized.

The formation of the trisulphide must involve the scission of a carbon–sulphur bond in cystine. We have, therefore, examined cystine solutions that had been subjected to a 4×10^7 r. dose of γ-irradiation from a multicurie cobalt-60 source. Traces of (I), about 3 per cent of the initial cystine sulphur, were found, but not sufficient to make this a practicable source of the compound.

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An Interpretation of the Transition in Composition of Soluble Lipoproteins

A recent examination[1] of a wide range of soluble lipoproteins has shown that a transition in the proportions of protein, phospholipid and neutral lipid occurs in the region where these three components are present in about equal amounts. This transition is clearly evident when the percentage of neutral lipid (Y) is plotted against the proportion of phospholipid in the protein–phospholipid complex (X). Curves fitted to the experimental points for 42 lipoproteins by least squares showed that two linear equations:

$$Y = 8{\cdot}54 + 51{\cdot}6\,X \text{ (for } Y < 40 \text{ per cent)} \quad (1)$$

$$Y = 8{\cdot}73 + 99{\cdot}5\,X \text{ (for } Y > 40 \text{ per cent)} \quad (2)$$

represented the data more adequately than a cubic equation fitted to all the points. However, both types of equation show evidence of the transition since the discontinuity between the linear equations