586. *Some Derivatives of S-Carboxyalkyl-cysteines and -homocysteines.*

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The preparation of amides and hydrazides from some sulphur-containing amino-acids and their sulphones is described. Reduction of the amides by lithium aluminium hydride gave both hydroxy-amines and hydroxy-amides.

The use of *S*-ethylcysteine as a tuberculostatic agent *in vivo* has been claimed by an American group of workers.[1] Certain labile *S*-ethyl derivatives such as thiol esters have also been shown to possess this activity.[2] Although, in the main, derivatives of simple sulphur-containing amino-acids do not exhibit significant bacteriostatic activity,[3] it has been claimed that *S*-carboxymethyl-L-cysteine (I; R = H, R′ = R″ = OH) shows some activity against encephalitis-type viruses,[4] while the hydrazide [5] and substituted hydrazides [6] from methionine are bacteriostatic. Some derivatives of *S*-carboxyalkyl-cysteines and -homocysteines have therefore been prepared for microbiological examination.

(I) R′·CO·CH(NHR)·CH_2·S·CH_2·CO·R″ R′·CO·CH(NHR)·CH_2·SO_2·CH_2·CO·R′ (II)

S-Carboxymethyl-L-cysteine [7] was converted into the *N*-benzyloxycarbonyl derivative (I; R = CH_2Ph·O·CO, R′ = R″ = OH). Methylation with diazomethane afforded an oily dimethyl ester which was treated with ethanolic ammonia and with hydrazine hydrate to give the diamide (I; R = CH_2Ph·O·CO, R′ = R″ = NH_2) and the dihydrazide (I; R = CH_2Ph·O·CO, R′ = R″ = NH·NH_2) respectively. The same diamide was obtained when the dihydrazide was converted into the diazide and caused to react with ethanolic ammonia. After treatment with hydrogen bromide in acetic acid, the *N*-benzyloxycarbonyl-diamide was converted into the dihydrobromide of *S*-carbamoylmethyl-L-cysteine amide (I; R = H, R′ = R″ = NH_2).

[1] Solotorovsky, Winsten, Ironson, Brown, and Becker, *Amer. Rev. Tuberculosis*, 1954, **70**, 806; Brown, Matzuk, Becker, Conbere, Constanten, Solotorovsky, Winsten, Ironson, and Quastel, *J. Amer. Chem. Soc.*, 1954, **76**, 3860.

[2] Kushner, Dalalian, Bach, Centola, Sanjurio, and Williams, *J. Amer. Chem. Soc.*, 1955, **77**, 1152.

[3] Karrer and Aman, *Helv. Chim. Acta*, 1950, **33**, 302; Youmans, Doub, and Youmans, *Chem. Biol. Coordination Centre Review No.* 4, p. 249, Nat. Res. Council, Washington, D.C., 1953.

[4] Foye and Vederame, *J. Amer. Pharm. Assoc.*, 1957, **46**, 273.

[5] Harii, *J. Pharm. Soc. Japan*, 1956, **76**, 1319.

[6] Winterstein, Hegedüs, Fust, Böhni, and Studer, *Helv. Chim. Acta*, 1956, **39**, 229.

[7] Du Vigneaud, Audrieth, and Loring, *J. Amer. Chem. Soc.*, 1930, **52**, 4500.

The benzoyl dihydrazide (I; R = Bz, R′ = R″ = NH·NH_2) was similarly prepared and converted into the diazide which with aniline afforded two products, only one of which was the expected dianilide (I; R = Bz, R′ = R″ = NHPh): a second product gave analyses for $C_{17}H_{19}O_2N_3S$, which is not consistent with a half-amide structure such as (I; R = Bz, R′ = NH_2, R″ = NHPh) that might have arisen by replacement of an azide group by an amide as has been observed by some workers.[8] The related sulphone (II; R = Bz, R′ = OH) underwent a similar series of reactions to give the dihydrazide (II; R = Bz, R′ = NH·NH_2).

(III) R′·CO·CH(NHR)·CH_2·S·CH_2·CH_2·CO·R″ R′·CO·CH(NHR)·CH_2·SO_2·CH_2·CH_2·CO·R′ (IV)

S-2-Carboxyethyl-L-cysteine (III; R = H, R′ = R″ = OH) formed a crystalline *N*-benzyloxycarbonyl derivative from which was obtained the sulphone (IV; R = CH_2Ph·O·CO, R′ = OH) and the amide (III; R = CH_2Ph·O·CO, R′ = R″ = NH_2). *N*-Benzoyl-*S*-2-carboxyethyl-L-cysteine (III; R = Bz, R′ = R″ = OH) gave a crystalline dimethyl ester which furnished the dihydrazide with hydrazine hydrate, but with ethanolic ammonia gave only a half amide (III; R = Bz, R′ = OMe, R″ = NH_2 or its isomer). The diamide (III; R = Bz, R′ = R″ = NH_2) was, however, readily obtained from the diazide. The acid sulphone (IV; R = H, R′ = OH) was prepared by acid hydrolysis of its *N*-benzoyl derivative: a dihydrazide was also obtained from the latter compound.

R′·CO·CH(NHR)·CH_2·S·CHMe·CO·R″ (V)

Karrer and Aman [3] described the reaction of L-cysteine with L(−)-α-bromopropionic and with DL-α-iodopropionic acid in the presence of aqueous barium hydroxide to give *S*-(L-1-carboxyethyl)-L-cysteine (V; R = H, R′ = R″ = OH) and the diastereoisomeric mixture with the *S*-(DL-1-carboxyethyl) group, respectively, which had m. p. 178—179° and 169—171°. We have treated L-cysteine with DL-α-chloropropionic acid in liquid ammonia and obtained the above diastereoisomeric mixture with m. p. 183°. Benzoylation and esterification of the mixture gave an oily dimethyl ester which on treatment with hydrazine hydrate gave two crystalline dihydrazides (V; R = Bz, R′ = R″ = NH·NH_2). The first, m. p. 197—199°, was arbitrarily termed the α-, and the second, m. p. 157—159°, the β-dihydrazide. When the dimethyl ester was set aside with ethanolic ammonia, only one diamide (V; R = Bz, R′ = R″ = NH_2), m. p. 258—259°, could be isolated in crystalline form and was shown to be of the α-series since it was also obtained from the α-dihydrazide. The β-diamide, m. p. 219—220°, was obtained from the β-dihydrazide. *N*-Benzyloxycarbonyl-*S*-1-carboxyethyl-L-cysteine (V; R = CH_2Ph·O·CO, R′ = R″ = OH) was usually obtained as a solid, m. p. 104—106°, although in one experiment a product with m. p. 140° was isolated which gave correct analytical figures. The lower-melting product was probably a mixture and afforded an oily dimethyl ester from which only one pure dihydrazide could be obtained. Treatment of the oily ester with aqueous ammonia gave a half amide in poor yield (V; R = CH_2Ph·O·CO, R′ = OMe, R″ = NH_2, or its isomer), a reaction paralleled in the *S*-2-carboxyethyl-L-cysteine series.

CO_2H·CH(NH_2)·CH_2·CH_2·S·[CH_2]$_n$·CO_2H (VI)

When DL-methionine in liquid ammonia was treated with sodium, followed by β-chloropropionic acid, *S*-2-carboxyethyl-DL-homocysteine (VI; n = 2) was obtained. Armstrong and Lewis [9] reported that it was preferable to use *S*-benzyl-DL-homocysteine in this synthesis: we encountered no particular difficulty using DL-methionine. The same product was also conveniently prepared by heating DL-methionine and β-chloropropionic acid in hydrochloric acid.[9,10] From this starting material *N*-benzoyl-*S*-2-hydrazinocarbonylethyl-DL-homocysteine hydrazide and the corresponding diamide were prepared

[8] Hegedüs, *Helv. Chim. Acta*, 1948, **31**, 737; Roberts, *J. Amer. Chem. Soc.*, 1954, **76**, 6203; Roeske, Stewart, Stedman, and Du Vigneaud, *J. Amer. Chem. Soc.*, 1956, **78**, 5883; MacLaren, Savige, and Swan, *Austral. J. Chem.*, 1958, **2**, 1345.

[9] Armstrong and Lewis, *J. Org. Chem.*, 1951, **16**, 749.

[10] Dekker and Fruton, *J. Biol. Chem.*, 1948, **173**, 471.

in the usual way. A corresponding series of derivatives was prepared from *S*-carboxymethyl-DL-homocysteine (VI; $n = 1$).

$$R'\cdot CO\cdot CH(NHR)\cdot CH_2\cdot CH_2\cdot S\cdot CH_2\cdot CH(NHR)\cdot CO\cdot R' \quad (VII)$$

$$R'\cdot CO\cdot CH(NHR)\cdot CH_2\cdot CH_2\cdot SO_2\cdot CH_2\cdot CH(NHR)\cdot CO\cdot R' \quad (VIII)$$

Some derivatives of L-allocystathionine (VII; R = H, R′ = OH) and of the corresponding sulphone (VIII; R = H, R′ = OH) were prepared by similar methods: most of the allocystathionines prepared tended to form gels.

Many of our products were examined for activity *in vitro* against strains of *S. aureus* and *E. coli*: no bacteriostatic action was detected. Activity against *M. tuberculosis* H37Rv *in vivo* in mice was also absent.

(IX) $HO\cdot CH_2\cdot CH(NHR)\cdot CH_2\cdot S\cdot CH_2\cdot CH_2\cdot OH$ $HO\cdot CH_2\cdot CH(NHR)\cdot CH_2\cdot S\cdot CH_2\cdot CH_2\cdot CH_2\cdot OH$ (X)

A few of our compounds were reduced with lithium aluminium hydride, by Karrer and Aman's method,[3] in the hope of obtaining amino-substituted diols of types (IX) and (X) of microbiological interest. *N*-Benzoyl-*S*-methoxycarbonylmethyl-L-cysteine methyl ester (I; R = Bz, R′ = R″ = OMe), at room temperature, afforded a neutral solid and a basic oil. The solid gave correct analyses for the partially reduced product (IX; R = Bz), while the oil was characterised as the crystalline oxalate of the diol (IX; R = CH_2·Ph). Similarly reduction of the *S*-2-methoxycarbonylethyl derivative (III; R = Bz, R′ = R″ = OMe) afforded a crystalline amide (X; R = Bz) and benzyl derivative. Reduction of *S*-2-methoxycarbonylmethyl-L-cysteine methyl ester (III; R = H, R′ = R″ = OMe) under the same conditions gave an oily amine forming a crystalline hydrogen oxalate. Like the amino-acid derivatives, the reduced products had insignificant bacteriostatic action.

Experimental

S-Carboxymethyl-L-cysteine (I; R = H, R′ = R″ = OH).—L-Cystine (12·0 g.), stirred in liquid ammonia (200 ml.), was treated portionwise with sodium (4·7 g.) until the blue colour persisted for *ca.* 10 min. The colour was discharged by addition of solid ammonium chloride (2 g.), and chloroacetic acid (9·6 g.) was added in 10 min. After further stirring, the ammonia was allowed to evaporate, and the residual solid dissolved in warm water (120 ml.), treated with sodium cyanide (2·0 g.) and left for 15 min. The solution was brought to pH 3 with concentrated hydrochloric acid and cooled, a white solid separating [12·0 g; m. p. 194·5—196° (decomp.)]. A second crop was obtained from the mother-liquors [4·7 g.; m. p. 190—191° (decomp.)], both crops being free from thiol and disulphide. Crystallisation from water afforded leaflets (15·5 g.), m. p. 194·5° (decomp.) (Found: C, 34·0; H, 5·0; N, 7·7. Calc. for $C_5H_9O_4NS$: C, 33·6; H, 5·1; N, 7·8%). Armstrong and Lewis [9] give m. p. 204—207° (decomp.); Berger *et al.*[11] give m. p. 192° (decomp.).

S-Methoxycarbonylmethyl-L-cysteine Methyl Ester Hydrochloride (I; R = H, R′ = R″ = OMe).—*S*-Carboxymethyl-L-cysteine (3·0 g.) was left overnight with saturated methanolic hydrogen chloride. The *ester* (3·1 g.) formed needles, m. p. 98—100° (from ethanol–ether) (Found: C, 34·6; H, 5·6; N, 5·9. $C_7H_{14}O_4NSCl$ requires C, 34·4; H, 5·8; N, 5·8%).

N-Benzyloxycarbonyl-S-carboxymethyl-L-cysteine (I; R = CH_2Ph·O·CO, R′ = R″ = OH).—To a cooled stirred solution of *S*-carboxymethyl-L-cysteine (3·6 g.) in aqueous 4N-sodium hydroxide (5·8 ml.) were simultaneously added benzyl chloroformate (2·4 g.) and aqueous 4N-sodium hydroxide (12·5 ml.) during 15 min. After further stirring (1 hr.), water (20 ml.) was added and the mixture extracted with ether. The ether washings were rejected and the aqueous layer, after acidification, was extracted with ethyl acetate: evaporation of the extracts gave a *product* (3·1 g.) that formed needles, m. p. 116—117° from ethyl acetate–ether (Found: C, 50·1; H, 4·7; N, 4·4. $C_{13}H_{15}O_6NS$ requires C, 49·8; H, 4·5; N, 4·5%). Use of magnesium oxide in place of sodium hydroxide did not improve the yield.

[11] Berger, Noguchi, and Katchalski, *J. Amer. Chem. Soc.*, 1956, **78**, 4483.

N-Benzyloxycarbonyl-S-hydrazinocarbonylmethyl-L-cysteine Hydrazide (I; R = $CH_2Ph{\cdot}O{\cdot}CO$, R′ = R″ = $NH{\cdot}NH_2$).—The foregoing acid (3·0 g.), suspended in ether (30 ml.), was treated with excess of ethereal diazomethane; a clear solution was obtained. Evaporation afforded an almost colourless oil which was taken up in methanol (20 ml.) and treated with 95% hydrazine hydrate (3·0 ml.). Next morning the solution was filtered and concentrated; cream-coloured needles (2·1 g.), m. p. 153—154°, separated. Crystallisation from methanol containing a little water gave the pure *dihydrazide*, m. p. 153—154° (Found: C, 45·9; H, 6·0; N, 20·3. $C_{13}H_{19}O_4N_5$ requires C, 45·8; H, 5·6; N, 20·5%).

N-Benzyloxycarbonyl-S-carbamoylmethyl-L-cysteine Amide (I; R = $CH_2Ph{\cdot}O{\cdot}CO$, R′ = R″ = NH_2).—(*a*) Treatment of the foregoing dimethyl ester with methanolic ammonia at room temperature for 24 hr. yielded the *diamide* (2·5 g.), m. p. 182—183° (from methanol containing a few drops of water) (Found: C, 49·7; H, 5·6; N, 13·7. $C_{13}H_{17}O_4N_3S$ requires C, 50·1; H, 5·5; N, 13·5%). (*b*) The foregoing dihydrazide (0·2 g.) in N-hydrochloric acid (1·7 ml.) was stirred with ethyl acetate (2·0 ml.) and slowly treated with sodium nitrite (0·1 g.) in water (1·0 ml.). The organic layer was separated, washed with water, and dried. Ethanolic 8% ammonia (1·0 ml.) was added; a white solid (75 mg.), m. p. 171—172°, separated; from methanol this formed needles, m. p. 179—181° not depressed on admixture with the sample prepared as in method (*a*) (Found: C, 49·8; H, 5·5; N, 13·0%).

S-Carbamoylmethyl-L-cysteine Amide Dihydrobromide (as I; R = H, R′ = R″ = NH_2).—*N*-Benzyloxycarbonyl-*S*-carboxyamido-L-cysteine amide (0·5 g.) and a 50% solution (5·0 ml.) of hydrogen bromide in acetic acid were kept for 1 hr. Dry ether was added, and the *product* collected and washed with ether (0·52 g.), m. p. 160° (decomp.) (Found: C, 18·1; H, 4·1; N, 12·1. $C_5H_{13}O_2N_3SBr_2$ requires C, 17·7; H, 3·9; N, 12·4%). This salt was very soluble in water and did not crystallise satisfactorily.

N-Benzoyl-S-carboxymethyl-L-cysteine (I; R = Bz, R′ = R″ = OH).—*S*-Carboxymethyl-L-cysteine (15 g.) in 2N-sodium hydroxide (90 ml.) was stirred at 0° while benzoyl chloride (18·8 ml.) and 4N-sodium hydroxide (105 ml.) were added (15 min.). Stirring was continued for 1 hr., the mixture extracted once with ether, and the aqueous layer acidified to give an oil. After isolation with ethyl acetate the resulting oil was stirred with ether (200 ml.) to give a white solid (23·6 g.), m. p. 123—130°. Crystallisation from ethyl acetate gave white needles, m. p. 140·5—141·5° (19·1 g.) (Berger *et al.*[11] and Harris *et al.*[12] give m. p. 139—140°). When prepared in 8% aqueous sodium hydrogen carbonate or N-sodium hydroxide, a *bis-S-benzylisothiouronium salt sesquihydrate* was obtained, having m. p. 118—120° after drying at 70°/15 mm. (Found: C, 52·4; H, 5·7; N, 11·0; S, 15·2. $C_{28}H_{33}O_5N_5S_3,1{\cdot}5H_2O$ requires C, 52·3; H, 5·7; N, 10·9; S, 15·0%). In 2% aqueous sodium hydrogen carbonate a *mono-S-benzylisothiouronium salt* was formed as needles, m. p. 167—168° (Found: C, 52·9; H, 5·0; S, 13·6. $C_{20}H_{23}O_5N_3S_2$ requires C, 53·3; H, 5·2; S, 14·2%). Treatment of the bis-salt (0·56 g.) with 2N-hydrochloric acid yielded *N*-benzoyl-*S*-carboxymethyl-L-cysteine (0·165 g.), m. p. 136—138°, while the monosalt (0·57 g.) gave the same product (0·245 g.), m. p. 136—138°.

N-Benzoyl-S-methoxycarbonylmethyl-L-cysteine Methyl Ester (I; R = Bz, R′ = R″ = OMe)—Prepared from the corresponding acid (3·0 g.) and ethereal diazomethane, the diester was obtained as needles (2·8 g.), m. p. 59—60°, from aqueous methanol (Found: C, 53·9; H, 5·4; N, 4·4. Calc. for $C_{14}H_{17}O_5NS$: C, 54·0; H, 5·5; N, 4·5%), $[\alpha]_D^{13}$ −76·5° (*c* 1 in EtOH) (lit.,[11,12] m. p. 63—64°). The diester (1·0 g.), methanol (10 ml.), and aqueous ammonia (4 ml.; *d* 0·88) gave needles of the *diamide* (0·6 g.), m. p. 219—220° (from aqueous methanol) (Found: C, 51·2; H, 5·6; N, 14·8. $C_{12}H_{15}O_3N_3S$ requires C, 51·2; H, 5·4; N, 14·9%). The *dihydrazide hemihydrate* formed needles, m. p. 166—167°, from aqueous methanol (Found: C, 46·5; H, 5·6; N, 22·6; S, 10·2. $C_{12}H_{17}O_3N_5S,0{\cdot}5H_2O$ requires C, 46·5; H, 5·9; N, 22·6; S, 10·3%).

Reactions of N-Benzoyl-S-azidocarbonylmethyl-L-cysteine Azide.—The foregoing dihydrazide (3·0 g.), N-hydrochloric acid (25 ml.), and ethyl acetate (25 ml.) were stirred at 5° while sodium nitrite (1·4 g.) in water (3 ml.) was added during 10 min. After separation, the organic layer was washed with water, dried, and divided into two equal portions. (*a*) One portion was treated with 8% ethanolic ammonia (6 ml.), a white solid rapidly separating; this formed needles (1·1 g.), m. p. 225—226°, from aqueous methanol, not depressed on admixture with the above diamide. (*b*) To the other portion of azide solution was added aniline (2·0 ml.). After 17 hr., a solid was deposited (0·25 g.; m. p. 198—201°). Two crystallisations from aqueous dimethylformamide afforded needles of a substance, m. p. 216—217° (Found: C, 62·8;

[12] Harris, Easton, Heyl, and Folkers, *J. Amer. Chem. Soc.*, 1944, **66**, 1757.

H, 5·5; N, 12·3. Calc. for $C_{17}H_{19}O_2N_3S$: C, 62·1; H, 5·8; N, 12·7%). The substance was soluble in concentrated hydrochloric acid on slight warming. The original ethyl acetate mother-liquors were concentrated to give a second product (0·65 g.), m. p. 188—190°; the N-*benzoyldianilide* (I; R = Bz, R′ = R″ = NHPh) formed needles (from methanol), m. p. 200—201° (Found: C, 66·4; H, 5·2; N, 10·2. $C_{24}H_{23}O_3N_3S$ requires C, 66·5; H, 5·4; N, 9·7%).

N-*Benzoyl*-S-*carboxymethyl*-L-*cysteine* SS-*Dioxide* (II; R = Bz, R′ = OH).—*N*-Benzoyl-*S*-carboxymethyl-L-cysteine (3·3 g.), acetic acid (15 ml.), and 30% aqueous hydrogen peroxide (10 ml.) were warmed on the steam-bath for 30 min. Evaporation afforded the *sulphone* which separated from acetic acid–ethyl acetate as needles (2·0 g.), m. p. 177—178° (decomp.) (Found: C, 44·9; H, 4·2; N, 4·3; S, 9·5. $C_{12}H_{13}O_7NS,0{\cdot}5H_2O$ requires C, 44·5; H, 4·4; N, 4·3; S, 9·9%). Both a *mono*-S-*benzylisothiouronium salt*, needles (from ethanol–ether), m. p. 144—145° (decomp.) (Found: C, 49·9; H, 4·9; N, 8·7. $C_{20}H_{23}O_7N_3S_2$ requires C, 49·8; H, 4·8; N, 8·8%), and a *bis*-S-*benzylisothiouronium salt*, needles (from water), m. p. 167° (decomp.) (Found: C, 51·7; H, 5·1; N, 10·3. $C_{28}H_{33}O_7N_5S_3$ requires C, 51·8; H, 5·1; N, 10·8%), were obtained. The *dimethyl ester* (II; R = Bz, R′ = R″ = OMe), prepared from the acid with diazomethane, formed needles (from ethyl acetate), m. p. 156—157° (Found: C, 48·6; H, 5·2; N, 3·8; S, 9·0. $C_{14}H_{17}O_7NS$ requires C, 49·0; H, 5·0; N, 4·1; S, 9·3%). Treatment of the dimethyl ester (0·5 g.) in methanol (5 ml.) with 95% hydrazine hydrate (1·0 ml.) yielded the *sulphone dihydrazide* (II; R = Bz, R′ = R″ = NH·NH_2) (0·35 g.), needles (from methanol–water), m. p. 206—207° (decomp.) (Found: C, 41·8; H, 4·8; N, 20·6. $C_{12}H_{17}O_5N_5S$ requires C, 42·0; H, 5·0; N, 20·4%).

S-2-Carboxyethyl-L-cysteine (III; R = H, R′ = R″ = OH), prepared either by the action of β-chloropropionic acid on the sodium salt of L-cysteine in liquid ammonia (82% yield) or from L-cysteine and α-acetamidoacrylic acid (81% yield) according to Schöberl and Wagner,[13] had m. p. 218° (decomp.) (lit., m. p. 214—216° [13] and m. p. 227—230° [9]). Esterification with methanolic hydrogen chloride gave the *dimethyl ester hydrochloride*, needles (from methanol–ether), m. p. 90—91° (Found: C, 36·7; H, 6·4; N, 5·1. $C_8H_{16}O_4NSCl$ requires C, 37·2; H, 6·3; N, 5·4%). The *monohydroxamic acid* (III; R = H, R′ = OMe, R″ = NH·OH, or its isomer), prepared by treatment of the dimethyl ester with hydroxylamine (2 mol.) and potassium hydroxide in methanol, formed needles (from aqueous methanol), m. p. 153—154° (Found: C, 38·0; H, 6·1; N, 12·7. $C_7H_{14}O_4N_2S$ requires C, 37·8; H, 6·3; N, 12·6%).

N-*Benzyloxycarbonyl*-S-2-*carboxyethyl*-L-*cysteine* (III; R = CH_2Ph·O·CO, R′ = R″ = OH).—*S*-2-Carboxyethyl-L-cysteine (6·0 g.), magnesium oxide (4·6 g.), and water (60 ml.) were cooled in ice and stirred vigorously while benzyl chloroformate (6·0 g.) in toluene (40 ml.) was added during 30 min. The mixture was further stirred overnight at room temperature and filtered and the aqueous layer separated and extracted with ether. The ether washings were rejected, and the aqueous layer was acidified and extracted with ethyl acetate. Concentration afforded the *product* which separated from ethyl acetate–light petroleum as needles (7·3 g.), m. p. 92—94° (Found: C, 51·2; H, 5·4; N, 4·3. $C_{14}H_{17}O_6NS$ requires C, 51·5; H, 5·2; N, 4·3%). The *bis*-S-*benzylisothiouronium salt* formed needles (from ethanol–ether), m. p. 117—119° (Found: C, 54·6; H, 5·8; N, 10·2. $C_{30}H_{27}O_6N_5S_3$ requires C, 54·5; H, 5·7; N, 10·6%). The *sulphone* (IV; R = CH_2Ph·O·CO, R′ = OH) was obtained by oxidation of the sulphide (needles from ethyl acetate) and had m. p. 152·5—154° (Found: C, 47·1; H, 5·2; N, 3·9. $C_{14}H_{17}O_8NS$ requires C, 46·8; H, 4·8; N, 3·9%).

N-*Benzyloxycarbonyl*-S-2-*carbamoylethyl*-L-*cysteine Amide* (III; R = CH_2Ph·O·CO, R′ = R″ = NH_2).—The *N*-benzyloxycarbonyl acid (III; R = CH_2Ph·O·CO, R′ = R″ = OH) (6·0 g.) with diazomethane gave an oily dimethyl ester which was left with methanol (15 ml.) and aqueous ammonia (15 ml.; *d* 0·88) for 24 hr. The *diamide* (3·8 g.) formed needles, m. p. 160—162° (Found: C, 51·6; H, 5·9; N, 12·5. $C_{14}H_{19}O_4N_3S$ requires C, 51·7; H, 5·9; N, 12·9%).

S-2-*Carboxyethyl*-N-*toluene*-p-*sulphonyl*-L-*cysteine* (III; R = *p*-Me·C_6H_4·SO_2, R′ = R″ = OH).—Prepared under Schotten–Baumann conditions, the hemihydrate of this *product* formed needles, m. p. 142—144° (Found: C, 43·4; H, 5·2; N, 4·2. $C_{13}H_{17}O_6NS_2,0{\cdot}5H_2O$ requires C, 43·9; H, 5·1; N, 4·0%). Oxidation with peracetic acid yielded the *sulphone* (needles from water), m. p. 184—185° (Found: C, 39·5; H, 4·8; N, 3·9. $C_{13}H_{17}O_8NS_2,H_2O$ requires C, 39·3; H, 4·8; N, 3·5%).

N-*Benzoyl*-S-2-*carboxyethyl*-L-*cysteine* (III; R = Bz, R′ = R″ = OH).—Also prepared

[13] Schöberl and Wagner, *Chem. Ber.*, 1947, **80**, 379.

under Schotten–Baumann conditions (71% yield) this *product* crystallised from ethyl acetate–light petroleum as needles, m. p. 106·5—108° (Found: C, 52·5; H, 5·2; N, 4·4. $C_{13}H_{15}O_5NS$ requires C, 52·8; H, 5·1; N, 4·7%). The corresponding *sulphone* (IV; R = Bz, R′ = OH) formed needles, m. p. 178—179° (decomp.), from aqueous acetic acid (98% yield) (Found: C, 47·5; H, 4·4; N, 3·9; S, 9·3. $C_{13}H_{15}O_7NS$ requires C, 47·4; H, 4·6; N, 4·3; S, 9·7%).

S-2-*Carboxyethyl*-L-*cysteine* SS-*Dioxide* (IV; R = H, R′ = OH).—The foregoing sulphone (2·5 g.) and 17% hydrochloric acid (40 ml.) were heated under reflux for 5 hr. After cooling, the benzoic acid was removed (0·8 g.) and the filtrate evaporated to a gum which crystallised when scratched with chloroform. Crystallisation from water gave the *amino-sulphone* as needles (1·4 g.), m. p. 184—185° (decomp.) (Found: C, 32·0; H, 5·3; N, 6·0; S, 14·0. $C_6H_{11}O_6NS$ requires C, 32·0; H, 4·9; N, 6·2; S, 14·2%).

N-*Benzoyl*-S-2-*hydrazinocarbonylethyl*-L-*cysteine Hydrazide* SS-*Dioxide* (IV; R = Bz, R′ = R″ = NH·NH_2).—The *N*-benzoylcysteine dioxide (IV; R = Bz, R′ = OH) (2·0 g.) with diazomethane gave a *dimethyl ester* (1·8 g.), needles (from ethyl acetate), m. p. 154—156° (Found: C, 50·1; H, 5·3; N, 3·6. $C_{15}H_{19}O_7NS$ requires C, 50·4; H, 5·4; N, 3·9%). The ester (0·65 g.) in methanol with hydrazine hydrate afforded the *sulphone hydrazide* (0·5 g.) as needles, m. p. 200° (from aqueous methanol) (Found: C, 44·3; H, 5·4; N, 19·5. $C_{13}H_{19}O_5N_5S$ requires C, 43·7; H, 5·4; N, 19·6%).

Reactions of Dimethyl Ester (III; R = Bz, R′ = R″ = OMe).—The *N*-benzoyl acid (III; R = Bz, R′ = R″ = OH) (5·0 g.) was esterified with diazomethane to give the oily dimethyl ester, which was taken up in methanol (30 ml.) and divided into two equal portions. (*a*) To one portion was added aqueous ammonia (6 ml.; *d* 0·88) and the solution left overnight. Evaporation of the solvent and recrystallisation of the residue from aqueous methanol gave the *monoamide ester* (III; R = Bz, R′ = OMe, R″ = NH_2, or its isomer) as needles (1·0 g.), m. p. 138—139° (Found: C, 54·2; H, 5·5; N, 8·6. $C_{14}H_{18}O_4N_2S$ requires C, 54·2; H, 5·8; N, 9·0%). (*b*) From the other portion was obtained the *dihydrazide* (III; R = Bz, R′ = R″ = NH·NH_2) which formed needles (from aqueous methanol) (1·5 g.), m. p. 135—136° (Found: C, 48·0; H, 6·1; N, 21·4. $C_{13}H_{19}O_3N_5S$ requires C, 48·0; H, 5·9; N, 21·5%).

N-*Benzoyl*-S-2-*carbamoylethyl*-L-*cysteine Amide* (III; R = Bz, R′ = R″ = NH_2).—The foregoing hydrazide (1·65 g.) was converted *via* the diazide into the *diamide*, separating as needles (0·85 g.), m. p. 164—165°, from ethanol (Found: C, 52·6; H, 5·5; N, 13·8. $C_{13}H_{17}O_3N_3S$ requires C, 52·9; H, 5·8; N, 14·2%).

N-*Benzoyl*-S-2-N-*phenylcarbamoylethyl*-L-*cysteine Anilide* (III; R = Bz, R′ = R″ = NHPh).—Prepared from the dihydrazide in the same way as the diamide, the *dianilide* formed needles (0·95 g.), m. p. 178—179° (Found: C, 66·9; H, 5·6; N, 9·0. $C_{25}H_{25}O_3N_3S$ requires C, 67·1; H, 5·6; N, 9·4%).

S-1-Carboxyethyl-L-cysteine (V; R = H, R′ = R″ = OH) was prepared from L-cysteine and α-chloropropionic acid (in the same way as *S*-carboxymethyl-L-cysteine) in 79% yield and formed prisms (from water), m. p. 177—178° (decomp.) (Found: C, 37·2; H, 5·8; N, 7·4. Calc. for $C_6H_{11}O_4NS$: C, 37·3; H, 5·7; N, 7·3%). Karrer and Aman [3] give m. p. 169—171°. The N-*benzoyl derivative* formed prisms, m. p. 139—140°, from ethyl acetate—light petroleum (Found: C, 52·6; H, 5·2; N, 4·7. $C_{13}H_{15}O_5NS$ requires C, 52·8; H, 5·1; N, 4·7%). When esterified with diazomethane, the *N*-benzoyl dimethyl ester was obtained as an oil which did not solidify and was used without purification.

Reactions of N-*Benzoyl*-S-2-*methoxycarbonylethyl*-L-*cysteine Methyl Ester*.—(*a*) The oily ester (4·5 g.) in methanol (25 ml.) was treated with hydrazine hydrate (3·5 ml.) and left for 24 hr. at room temperature, giving a solid (4·6 g.), m. p. 153—170°: from the mother-liquors was obtained a second solid (0·25 g.), m. p. 143—155°. Fractional crystallisation from methanol gave the α-*dihydrazide* (V; R = Bz, R′ = R″ = NH·NH_2) as needles (1·8 g.), m. p. 197—198° (Found: C, 47·6; H, 6·2; N, 20·9. $C_{13}H_{19}O_3N_5S$ requires C, 48·0; H, 5·9; N, 21·5%). The more soluble β-*dihydrazide* formed needles (1·2 g.), m. p. 153—154°, from ethyl acetate–methanol (Found: C, 47·8; H, 6·0; N, 21·6%). (*b*) Aqueous ammonia (5 ml.; *d* 0·88) was added to the dimethyl ester (1·5 g.) in methanol and left for 24 hr., to give a solid (0·2 g.), m. p. 216—220° (decomp.): evaporation of the mother-liquor afforded only gum. The α-*diamide* (V; R = Bz, R′ = R″ = NH_2) formed needles (from aqueous methanol), m. p. 251—252° (decomp.) (Found: C, 53·4; H, 5·7; N, 14·0. $C_{13}H_{17}O_3N_3S$ requires C, 52·9; H, 5·8, N, 14·2%). Authentic α-diamide (V; R = Bz, R′ = R″ = NH_2) identical with the foregoing diamide was prepared in the usual manner from the α-dihydrazide (2·0 g.) as needles (1·0 g.), m. p. 258—259° (decomp.),

showing no depression of mixed m. p. (Found: C, 52·3; H, 5·5; N, 13·9%). The corresponding β-*diamide* (0·8 g.) was similarly prepared from the β-dihydrazide (2·0 g.) and formed rosettes of needles (from water), m. p. 219—220° (Found: C, 52·7; H, 5·9; N, 14·3%).

N-*Benzyloxycarbonyl*-S-1-*carboxymethyl*-L-*cysteine* (V; R = $CH_2Ph{\cdot}O{\cdot}CO$, R′ = R″ = OH), obtained by the Schotten–Baumann method, formed needles, m. p. 104—106°, from ether–light petroleum (Found: C, 51·3; H, 5·2; N, 4·3. $C_{14}H_{17}O_6NS$ requires C, 51·5; H, 5·2; N, 4·3%). In one experiment a small quantity of an isomer, m. p. 140°, was isolated (Found: C, 52·1; H, 4·9; N, 4·2%). Esterification of the lower-melting material yielded an oily dimethyl ester which was used with purification.

N-*Benzyloxycarbonyl*-S-1-*hydrazinocarbonylethyl*-L-*cysteine Hydrazide* (V; R = $CH_2Ph{\cdot}O{\cdot}CO$, R′ = R″ = $NH{\cdot}NH_2$).—The foregoing oily ester (2·5 g.) gave, by the usual method, needles of a *dihydrazide* (1·5 g.) (from methanol), m. p. 180—181° (Found: C, 47·5; H, 5·5; N, 19·9. $C_{14}H_{21}O_4N_5S$ requires C, 47·3; H, 5·7; N, 19·7%). When the same ester (4·0 g.) was treated in methanol (20 ml.) with aqueous ammonia (15 ml.; *d* 0·88) reaction was slow. Crystallisation of the crude solid (0·75 g.) from water gave needles of the *monoamide methyl ester* (0·6 g.) (V; R = $CH_2Ph{\cdot}O{\cdot}CO$, R′ = OMe, R″ = NH_2, or an isomer), m. p. 146—148° (Found: C, 53·3; H, 6·0; N, 7·7. $C_{15}H_{20}O_5N_2S$ requires C, 53·0; H, 5·9; N, 8·2%).

S-2-*Carboxyethyl*-DL-*homocysteine* (VI; *n* = 2).—Methionine (15 g.) in liquid ammonia (200 ml.) was stirred while sodium (*ca.* 6 g.) was added in small portions. Addition of β-chloropropionic acid (11·5 g.) and working up in the usual way afforded white needles (18·9 g.), m. p. 215—217°. Two crystallisations from water gave the pure product (12·5 g.), m. p. 232° (decomp.) (lit.,[9] m. p. 227—230°) (Found: C, 40·0; H, 6·6; N, 7·1. Calc. for $C_7H_{13}O_4NS$: C, 40·5; H, 6·4; N, 6·8%). The same product was obtained (10·1 g.) when DL-methionine (14·9 g.), concentrated hydrochloric acid (200 ml.), and β-chloropropionic acid (11·5 g.) were heated under reflux for 24 hr. The N-*benzoyl derivative* formed needles (from ethyl acetate–light petroleum), m. p. 122—124° (Found: C, 53·5; H, 5·5; N, 3·9. $C_{14}H_{17}O_5NS$ requires C, 54·0; H, 5·5; N, 4·5%).

N-*Benzoyl*-S-2-*hydrazinocarbonyl*-DL-*homocysteine Hydrazide*.—The foregoing acid was converted *via* the oily dimethyl ester into the *dihydrazide* which separated from methanol as a gelatinous solid containing varying amounts of water. After drying at 50°/15 mm., the hemihydrate had m. p. 98—99° (cloudy melt, clear at 152°) (Found: C, 48·3; H, 6·2; N, 19·7. $C_{14}H_{21}O_3N_5S,0{\cdot}5H_2O$ requires C, 48·0; H, 6·3; N, 20·1%). From the dihydrazide (1·7 g.) N-*benzoyl*-S-2-*carbamoylethyl*-DL-*homocysteine amide* was obtained as needles (from dimethylformamide–ethanol) (1·0 g.), m. p. 185—187° (Found: C, 54·6; H, 6·3; N, 13·6. $C_{14}H_{19}O_3N_3S$ requires C, 54·4; N, 6·2; N, 13·6%).

S-Carboxymethyl-DL-homocysteine (VI; *n* = 1), prepared as the carboxyethyl homologue, formed needles, m. p. 225—226° (decomp.) (lit.,[9] m. p. 224—226°). Its N-*benzoyl derivative* formed needles (from ethyl acetate–light petroleum), m. p. 155—156° (Found: C, 52·6; H, 5·2; N, 4·5. $C_{13}H_{15}O_5NS$ requires C, 52·5; H, 5·1; N, 4·7%). Treatment of the *N*-benzoyl compound with diazomethane afforded an oily dimethyl ester which with hydrazine hydrate was converted into N-*benzoyl*-S-*hydrazinocarbonylmethyl*-DL-*homocysteine hydrazide* (50% yield), needles (from aqueous methanol), m. p. 121—123° (Found: C, 48·4; H, 6·6; N, 21·3. $C_{13}H_{19}O_3N_5S$ requires C, 48·1; H, 6·4; N, 21·6%). The corresponding *diamide* prepared from the diester by the action of aqueous ammonia in methanol formed needles (55% yield), m. p. 184—185° (Found: C, 52·7; H, 5·8; N, 14·1. $C_{13}H_{17}O_3N_3S$ requires C, 52·9; H, 5·8; N, 14·2%). The same diamide was obtained from the diazide corresponding to the foregoing dihydrazide by treatment with methanolic ammonia, having m. p. and mixed m. p. 183—184°.

N-*Benzoyl*-S-N-*phenylcarbamoylmethyl*-DL-*homocysteine anilide*, also prepared from the above dihydrazide, crystallised from aqueous dimethylformamide as needles of the hemihydrate, m. p. 185—187° (Found: C, 65·5; H, 5·7; N, 9·2. $C_{25}H_{25}O_3N_3S,0{\cdot}5H_2O$ requires C, 65·5; H, 5·7; N, 9·2%).

S-2-*Carboxypropyl*-L-*cysteine*.—L-Cysteine hydrochloride (15·8 g.) and methacrylic acid (10·6 g., 90% material) were brought to pH 7—8 under nitrogen with 2N-sodium hydroxide, and the solution was heated for 8 hr. on the steam-bath until the test for thiol was negative (nitroprusside). The cooled solution was brought to pH 2 with hydrochloric acid, giving a solid (16·4 g.), m. p. 188—190° (decomp.). Two crystallisations from water gave the *product* as needles, m. p. 194° (decomp.) (Found: C, 40·7; H, 6·3; N, 6·6. $C_7H_{13}O_4NS$ requires C, 40·5; H, 6·3; N, 6·8%).

NN′-*Dibenzoyl*-L(−)-*allocystathionine Dihydrazide* (VII; R = Bz, R′ = NH·NH_2).—Dibenzoyl-L(−)-allocystathionine [14] (1 g.) with ethereal diazomethane gave the *dimethyl ester* (VII; R = Bz, R′ = OMe) (0·95 g.), m. p. 155—156° (from ethanol) (Found: C, 60·1; H, 5·7; N, 5·8. $C_{23}H_{26}O_6N_2S$ requires C, 60·2; H, 5·7; N, 6·1%), which was converted into the *dihydrazide* (0·7 g.) (needles from ethanol), m. p. 225—226° (Found: C, 55·7; H, 6·0; N, 17·7. $C_{21}H_{26}O_4N_6S$ requires C, 55·0; H, 5·7; N, 18·4%).

NN′-*Dibenzoyl*-L(−)-*allocystathionine*-SS-*Dioxide* (VIII; R = Bz, R′ = OH).—Oxidation of the dibenzoyl acid (1 g.) with peracetic acid gave the *sulphone hydrate* (1 g.), m. p. 131—132° (from water) (Found: C, 53·0; H, 4·8; N, 5·8. $C_{21}H_{22}O_8N_2S,H_2O$ requires C, 52·5; H, 5·0; N, 5·8%); esterification with ethereal diazomethane gave NN′-*dibenzoyl*-L(−)-*allocystathionine dioxide dimethyl ester* (VIII; R = Bz, R′ = OMe) (0·9 g.), m. p. 190—191°, as needles from ethyl acetate–methanol (Found: C, 56·0; H, 5·4; N, 5·7. $C_{23}H_{26}O_8N_2S$ requires C, 56·3; H, 5·3; N, 5·7%). Treatment of the ester with hydrazine hydrate yielded a compound soluble in aqueous sodium hydroxide which was probably NN′-*dibenzoyl*-L(−)-*allocystathionine dioxide monohydrazide*, m. p. 190—192° (from aqueous dimethylformamide) (Found: C, 52·8; H, 5·1; N, 12·2. $C_{21}H_{24}O_7N_4S$ requires C, 52·9; H, 5·1; N, 11·8%).

NN′-*Dibenzyloxycarbonyl*-L(−)-*allocystathionine* (VII; R = CH_2Ph·O·CO, R′ = OH).—Benzyl chloroformate (3·5 ml.) in toluene (25 ml.) was added, dropwise with stirring, to a mixture of L(−)-allocystathionine [14] (2 g.), magnesium oxide (3·6 g.), and water at 0°. After being stirred overnight, the solid was filtered off and the filtrate extracted with ether. The solid was dissolved in dilute hydrochloric acid, combined with the filtrate, and extracted with ethyl acetate. Evaporation of the ethyl acetate and crystallisation of the resulting solid from ethanol–light petroleum (b. p. 40—60°) gave the *dibenzyloxycarbonyl compound* (2·35 g.), m. p. 138—140° (Found: C, 56·2; H, 5·3; N, 5·4. $C_{23}H_{26}O_8N_2S$ requires C, 56·3; H, 5·3; N, 5·7%).

NN′-*Dibenzyloxycarbonyl*-L(−)-*allocystathionine Dihydrazide* (VII; R= CH_2Ph·O·CO, R′ = NH·NH_2).—The dibenzyloxycarbonyl acid (1 g.) with ethereal diazomethane gave an oily dimethyl ester from which was obtained the *dihydrazide* (0·7 g.), m. p. 148—150° (from aqueous methanol) (Found: C, 52·5; H, 6·1; N, 16·1. $C_{23}H_{30}O_6N_6S$ requires C, 53·2; H, 5·8; N, 16·2%).

Reduction of N-*Benzoyl*-S-*methoxycarbonylmethyl*-L-*cysteine Methyl Ester* (I; R = Bz, R′ = R″ = OMe).—The dimethyl ester (3 g.) in dry ether (100 ml.) was added dropwise with stirring to a suspension of lithium aluminium hydride (1·3 g.) in dry ether (150 ml.). The mixture was stirred for 4 hr. and then hydrolysed by ether saturated with water. The inorganic solid was filtered off and extracted with ether; the filtrate and ether extract were combined and gradually deposited fine needles. Crystallisation from ethyl acetate gave 2-*benzamido*-3-*hydroxypropyl* 2-*hydroxyethyl sulphide* (IX; R = Bz) (0·5 g.), m. p. 117—118° (Found: C, 55·8; H, 6·5; N, 5·2. $C_{12}H_{17}O_3NS$ requires C, 56·4; H, 6·7; N, 5·5%). Evaporation of the ether filtrate gave an oil (0·7 g.) which was treated in ethanol with oxalic acid dihydrate (0·35 g.). The resulting solution, on treatment with ethyl acetate and light petroleum (b. p. 40—60°), deposited crystals (0·2 g.), m. p. 124—127° (decomp.). Recrystallisation of this solid from methanol–ethyl acetate gave *di*-(2-*benzylamino*-3-*hydroxypropyl* 2-*hydroxyethyl sulphide*) *oxalate*, m. p. 166—167° (decomp.) (Found: C, 55·0; H, 7·0; N, 4·4. $C_{26}H_{40}O_8N_2S_2$ requires C, 54·5; H, 7·0; N, 4·9%).

Reduction of N-*Benzoyl*-S-2-*methoxycarboxyethyl*-L-*cysteine Methyl Ester* (III; R = Bz, R′ = R″ = OMe).—The dimethyl ester (5 g.), reduced as above, gave 2-*benzamido*-3-*hydroxypropyl* 3-*hydroxypropyl sulphide* (X; R = Bz) (1·7 g.), m. p. 94—95°, as needles from ethyl acetate (Found: C, 58·1; H, 7·6; N, 5·2. $C_{13}H_{19}O_3NS$ requires C, 58·0; H, 7·1; N, 5·2%), together with (2-*benzylamino*-3-*hydroxylpropyl* 3-*hydroxypropyl sulphide*) *hydrogen oxalate*, m. p. 130—132° [from methanol–ethyl acetate–light petroleum (b. p. 40—60°)] (Found: C, 51·8; H, 6·7; N, 4·0. $C_{15}H_{23}O_6NS$ requires C, 52·2; H, 6·7; N, 4·1%).

The acid oxalate (2·5 g.) was suspended in dry methanol (10 ml.) and treated with methanolic 0·42N-potassium hydroxide (33·7 ml.). Potassium oxalate was filtered off and the filtrate evaporated. The *amino-diol* (1·5 g.) crystallised from ethyl acetate–light petroleum (b. p. 40—60°) as needles, m. p. 58·5—60° (Found: C, 60·7; H, 8·5; N, 5·8. $C_{13}H_{21}O_2NS$ requires C, 61·1; H, 8·4; N, 5·5%).

Reduction of S-2-*Methoxycarbonylethyl*-L-*cysteine Methyl Ester* (III; R = H, R′ = R″ = OMe).—The dimethyl ester hydrochloride (5·1 g.) in chloroform (20 ml.) was treated with triethylamine (2·0 g.) and dry ether. The precipitated triethylamine hydrochloride was

[14] McHale, Mamalis, and Green, 2847.

collected and the filtrate evaporated at normal pressure. The resulting oil in dry ether (100 ml.) was added, dropwise with stirring, to lithium aluminium hydride (2·3 g.) in dry ether (100 ml.). The mixture was stirred for 4 hr. and then hydrolysed by ether saturated with water. The inorganic solid was filtered off and extracted with ethanol. Evaporation of the ethanol gave a gum which on treatment with oxalic acid in ethanol gave (*2-amino-3-hydroxypropyl 3-hydroxypropyl sulphide*) *hydrogen oxalate*, m. p. 104—105° (from methanol–ethyl acetate) (Found: C, 37·5; H, 6·5; N, 5·3. $C_8H_{17}O_6NS$ requires C, 37·6; H, 6·7; N, 5·5%).

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