Eugenolglycolic Acid Thiosemicarbazide.—Eugenolglycolic acid chloride obtained from 2.22 g of eugenolglycolic acid was added dropwise to an ice-cold solution of 0.91 g of thiosemicarbazide in 4.0 ml of pyridine. The reaction mixture was allowed to stand at room temperature for 1 hr. It was then poured into H_2O and the product obtained was recrystallized from 50% EtOH to give white needles, mp 193°, yield 93.4%.

Anal. Calcd for $C_{13}H_{17}N_3O_3S$: C, 52.88; H, 5.81; N, 14.23. Found: C, 52.99; H, 5.92; N, 14.06.

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A Ready Synthesis of Pantothenoylcystine

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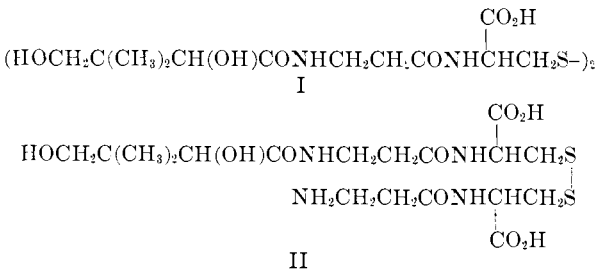
A new synthesis of the title compound I, which we feel will prove simpler in operation than that given by Baddiley and Mathias[2] for N-pantothenoyl-L-cysteine, is described.



I

II

Experimental Section[3]

N,N′-Bis(β-alanyl)-L-cystine.—Carbobenzyloxy-β-alanine[4] (22.3 g) was added portionwise with vigorous shaking during 20 min to a suspension of freshly ground PCl_5 (25 g) in dry ether (100 ml) at 0°. The mixture was filtered free of excess PCl_5 and concentrated *in vacuo* to an oil, dissolved in xylene (50 ml), and reconcentrated at low temperature and pressure (bath temp 40°). The resultant oil was dissolved in dry ether (50 ml) and added in six portions during 15 min with vigorous shaking to a solution of L-cystine (12 g) in 1 *N* NaOH (10 ml) at 0° in a stoppered flask. During the addition a further quantity of 1 *N* NaOH (130 ml) and water (100 ml) was added in portions. Shaking was continued for a further 15 min after addition was complete, and the mixture was acidified to congo red with 6 *N* HCl and extracted with ethyl acetate. The extracts were washed with water, dried overnight (Na_2SO_4), concentrated to 70 ml, and left at 0° when N,N′-bis(carbobenzyloxy-β-alanyl)-L-cystine crystallized; yield 24.3 g (75%), mp 118–125°.

The crude peptide (24.3 g) was dissolved in anhydrous liquid NH_3 (140 ml) and treated with stirring with small pieces of Na until a permanent blue color formed (required 5.8 g). The NH_3 was allowed to evaporate overnight and the residual solid dissolved in ice–water (60 ml) and was neutralized with HI (126 ml, 2 *N*, equivalent to 5.8 g of Na). The solution was cooled to 0° and treated slowly with aqueous H_2O_2 (20 vol, *ca.* 30 ml) until a test probe gave a negative nitroprusside reaction. After filtering, the solution was concentrated *in vacuo* then treated with ethanol when a viscous oil precipitated. This oil was washed free of NaI by decantation and dissolved in water (10 ml), and ethanol was added to turbidity when, on standing at 0° for several days, the peptide crystallized as feathery needles, yield 15.4 g, mp 201–202° dec. A further 3.0 g of the peptide was obtained from the ethanol washings; yield 71%, $[\alpha]^{20}D$ −126° (*c* 2.0, H_2O). This compound is reluctant to crystallize if it is very impure or if it is wet.

Anal. Calcd for $C_{12}H_{22}N_4O_8S_2$: C, 37.7; H, 6.2; N, 15.0; S, 16.8. Found: C, 37.7; H, 6.2; N, 15.0; S, 16.8.

N,N′-Bis(pantothenoyl)-L-cystine (I).—N,N′-Bis(β-alanyl)-L-cystine (9.55 g) in methanol (200 ml) was treated with 1 equiv of $NaOCH_3$ in methanol (37.0 ml, 1.37 *N*) and then with (−)-pantolactone (6.5 g). The solution was concentrated to an oil and held under N_2 at 100° for 3 hr. The resultant brittle foam is substantially pure disodium N,N′-bis(pantothenoyl)-L-cystine (I). Purification was effected by treating an aliquot (0.05) with the equivalent of 1 *N* HCl (2.5 ml) then partitioning the product between 1-butanol and water. The countercurrent distribution was effected by using 13 tap funnels each containing 1-butanol (20 ml), placing the material in the first funnel. The moving phase was water saturated with 1-butanol (10 ml) which passed through the system and was collected on issuing from the last funnel. This process was continued until 12 aqueous eluents had passed through the system and been collected. Examination of these eluents and of the contents of the tap funnels (homogenized by the addition of 5 ml of ethanol) by paper chromatography (1-butanol–acetic acid–water, 4:1:5) showed that the unreacted peptide was located in the first three aqueous eluents (R_f 0.11, detected by ninhydrin and by $NaCN-Na_2Fe(CN)_5NO$), while the monosubstituted peptide II was located in the first six eluents (R_f 0.20, detected by ninhydrin and by $NaCN-Na_2Fe(CN)_5NO$). Pure I was found to predominate in tap funnels 8–13 and in the last four eluents (R_f 0.3–0.5, elongated spot, detected only by $NaCN-Na_2Fe(CN)_5NO$). Concentration of these fractions gave the material as a resin, $[\alpha]^{20}D$ −77° (*c* 1.4, H_2O).

Anal. Calcd for $C_{24}H_{42}N_4O_{12}S_2$: C, 44.8; H, 6.6; N, 8.7; S, 10.0. Found: C, 44.7; H, 6.6; N, 8.6; S, 9.7.

(1) John Wyeth & Brother Ltd., Taplow, Maidenhead, Berkshire, England.
(2) J. Baddiley and A. P. Mathias, *J. Chem. Soc.*, 2803 (1954).
(3) Melting points were measured in open capillaries and are uncorrected. Microanalyses were performed by Drs. Weiler and Strauss, Oxford, England.
(4) R. H. Sifferd and V. du Vigneaud, *J. Biol. Chem.*, **108**, 753 (1935).