because the decreasing effects of chloride on k_5, respectively k_4' then predominate. Such effects would have been hard to understand before the theory of the system was developed.

Finally, we can now calculate the influence of any of the variables on the redox potential of a solution in equilibrium. For instance, if DPN, DPNH and ADH, all in the concentration 10^{-4} *M*, were mixed with ethanol, 2×10^{-3} *M*, the system would have a potential 34 mV higher than for the same system at low [ADH]. If now 0.15 *M* NaCl were added, the potential would drop not less than 9 mV. We think such anion effects may be of physiological importance in regulating the potentials in the living tissues, where chloride-free cells are surrounded by chloride containing intercellular fluid.

The effects of chloride seem to differ considerably in various enzyme-coenzyme systems. For instance, the redox potential of the yeast ADH system is not influenced by chloride. For the oxidized form of the old yellow ferment the "off" velocity constant is increased by chloride, whereas it is decreased for the DPN · ADH complex.

The complete data will be given in a forthcoming paper in this journal.

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Studies on Antimetabolites

IV. Synthesis of the α,α-Dimethyl Analogue of (+)-Panthothenic Acid *

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Following up our investigations on the possibility of designing antimetabolites by the introduction of a suitably situated *gem*dimethyl group into the molecule of an essential metabolite (outlined in Part I[1] of this series), we have now prepared the α,α-dimethyl analogue (I) of (+)-panthothenic acid (II).

The synthetical method used was essentially adopted from Kuhn and Wieland's synthesis [2] of panthothenic acid, the purification of the crude material being, however, carried out in a different way. (−)-Pantholactone was condensed with the benzylester (III) of α,α-dimethyl-β-alanine, the ester group was removed by hydrogenolysis, and the crude acid, which could not be induced to crystallise, was converted into its calcium salt, which was obtained as a microcrystalline powder. The intermediate benzylester (III) was prepared from α,α-dimethyl-β-alanine *via* the hydrochloride of α,α-dimethyl-β-alanyl-chloride, obtained from the amino acid and phosphorus pentachloride in acetyl-chloride solution. No attempt was made to improve the rather low yield of panthothenic acid analogue.

Preliminary tests with *Lactobacillus arabinosus* 17-5, using an agar cup method, showed that the panthothenic acid analogue inhibited the growth of this organism, and that this inhibition was competitively reversed by addition of (+)-panthothenic acid. The inhibition index was about 10 000. A detailed report will be published by Dr. A. Bolinder, Department of Food Chemistry, Royal Institute of Technology, Stockholm.

Experimental *. *Benzyl α,α-dimethyl-β-alaninate hydrochloride.* Ethyl β-formamino-α,α-dimethylpropionate [3] (60 g) and 20 % hydrochloric acid (150 ml) were refluxed overnight. The mixture was evaporated to dryness *in vacuo*, the dry salt extracted twice with ether, powdered, and added in small portions to acetyl chloride (400 ml) with shaking. When all had been added, finely ground phosphorus pentachloride (80 g) was added and the stoppered flask shaken overnight. Anhydrous ether (750 ml) was added and the precipitated solid was centrifuged and repeatedly washed with dry ether, mixed with benzyl alcohol (100 ml) and heated on the water bath until the evolution of hydrogen chloride ceased. The solution was filtered and diluted with much dry ether, and the precipitated ester hydrochloride filtered off and washed with ether. For purification the product was dissolved in the minimum amount of dry acetone, filtered, and reprecipitated by the addition of much ether. Lustrous plates, m.p. 94.5—96°. Yield 41 g. (Found:

* Part III. *Acta Chem. Scand.* **8** (1954) 1389.

* All melting points uncorrected.

$HOCH_2 \cdot C(CH_3)_2 \cdot CH(OH) \cdot CO \cdot NH \cdot CH_2 \cdot CR_2 \cdot COOH$

I R = CH_3 II R = H

$H_2N \cdot CH_2 \cdot C(CH_3)_2 \cdot COO \cdot CH_2 \cdot C_6H_5$

III

N 5.81; Cl 14.5. $C_{12}H_{18}ClNO_2$ requires: N 5.75; Cl 14.6).

The *picrate* was obtained by addition of a hot, saturated solution of picric acid to an aqueous solution of the ester hydrochloride. Yellow needles from aqueous ethanol, m.p. 147—148°. (Found: C 49.7; H 4.60; N 12.8. $C_{18}H_{20}N_4O_9$ requires: C 49.6; H 4.62; N 12.8).

Calcium (+)-α,α-dimethylpanthothenate. Benzyl α,α-dimethyl-β-alaninate hydrochloride (15 g) in water (25 ml) was made strongly alkaline with 6 *N* sodium hydroxide solution. The free amino acid ester was immediately extracted with ether, washed and dried (K_2CO_3). Evaporation of the ether yielded the benzyl ester as a colourless oil (12 g). (—)-Pantholactone (10 g, $[\alpha]_D^{20}$ −50.5°, water, *c* 2 %) was added, and the mixture was heated on the water bath for 5 hours. It was then diluted with water (50 ml), acidified with 2 *N* hydrochloric acid to pH 1—2 and repeatedly extracted with ether. The combined extracts were washed with two 20 ml portions of 2 *N* sodium hydroxide and then with water. After drying (Na_2SO_4) and removal of the ether, a colourless, sticky oil (13.5 g) was obtained. This was dissolved in glacial acetic acid (50 ml), palladium on charcoal (2 g, 10 %) added and the mixture shaken in an atmosphere of hydrogen for 5 hours. The catalyst was filtered off and the solution evaporated *in vacuo* on a water bath. A colourless syrup was obtained, which could not be induced to crystallise. It was dissolved in acetone (75 ml), filtered from a small amount of solid and evaporated to dryness *in vacuo*. The syrup was dissolved in water (300 ml), stirred for 2 hours with an excess of calcium carbonate, filtered, and continuously extracted with ether overnight. The aqueous solution was evaporated to dryness *in vacuo* at 40° and yielded a glass. This was dissolved in methanol (75 ml) and the solution was filtered and evaporated to dryness giving a white solid (5.0 g). This solid was purified by dissolution in methanol (50 ml), pouring into acetone (500 ml, reagent grade), storing overnight in a refrigerator, filtering off a small amount of precipitated material, and evaporation to dryness. The solid salt thus obtained was ground with little acetone (20 ml), filtered off, and washed with a little acetone and with ether. Yield 4.2 g, $[\alpha]_D^{20}$ + 27.2° (water, *c* 0.44 %). The product contained one mole of water of crystallization. (Found: C 48.0; H 7.81; Ca 7.37. $CaC_{22}H_{46}N_2O_{10} \cdot H_2O$ requires: C 48.0; H 7.69; Ca 7.28). The water could be removed by prolonged heating at 120° in high vacuum. (Found: H_2O (by loss of weight) 3.2 %. Calculated: 3.3 %).

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Studies on Antimetabolites

V. Improved Reduction of α-Oximino-β-phenylisovaleric Acids *

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The demand for relatively large amounts of "*neo*phenylalanine" (I) and "*neo*tyrosine" (II) for physiological tests prompted a reinvestigation of the reactions leading to these compounds in order to improve the yields. A detailed study of the reduction of the intermediate α-oximino acids showed that the use of anhydrous ethanolic hydrogen chloride instead of the lactic acid previously used [1] to control the pH in the reduction with sodium amalgam

* Part IV. *Acta Chem. Scand.* **8** (1954) 1491.