[CONTRIBUTION FROM THE DEPARTMENT OF FOOD TECHNOLOGY, UNIVERSITY OF ILLINOIS]

The Synthesis of Long Chain Fatty Acid Derivatives of Pantothenic Acid[1]

BY TAKETAMI SAKURAGI AND FRED A. KUMMEROW

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The long chain fatty acid derivatives of pantothenic acid, ethyl dipalmitoxypantothenate and ethyl monopalmitoxypantothenate, were prepared. The dipalmitoxy compound was obtained as an amorphous fatty powder with a m.p. of 57.0–58.5°, while the monopalmitate was a somewhat waxy substance which had a m.p. of 36.0°. Both derivatives were readily soluble in fats, and showed full activity for rats as a source of pantothenic acid. Evidence was presented to show that the reaction product of pantolactone with ammonia in an aqueous medium was pantamide instead of the ammonium salt of pantoic acid. The synthesis of dipalmitoxypantothenyl palmitate, which melted at 49.0–53.0°, was also reported.

Synthesis of several fat-soluble derivatives of pantothenic acid has been well reviewed in the treatises on vitamins.[2-4] The long chain fatty acid derivatives of pantothenic acid, however, have as yet not been reported. In the present study, the palmitic acid esters of ethyl pantothenate and pantothenyl alcohol were prepared. They were readily soluble in fats, and showed full activity for rats as a source of pantothenic acid.[5]

Ethyl pantothenate could simply be acylated to give the dipalmitate. This synthesis was also attempted, without success, through the condensation of dipalmitoxypantoic acid and ethyl β-aminopropionate after treating the former with thionyl chloride. The latter procedure was the one applied to the synthesis of the diacetate of pantothenic acid.[6,7] For the conversion of pantolactone to pantamide, Reichstein, *et al.*,[8] first reported a procedure using alcoholic ammonia. The reaction was later confirmed by several workers,[9-12] and again by the present study. Harris, *et al.*,[6] however, pointed out that their attempts to prepare the amide from pantolactone by treating with either alcoholic ammonia or aqueous ammonia were unsuccessful, and the products obtained were mainly the ammonium salts of pantoic acid in both cases. In the present study, the reaction of pantolactone with aqueous ammonia gave pantamide instead of ammonium pantoate. The synthesis of ethyl monopalmitoxypantothenate was carried out by means of the condensation of monopalmitoxypantolactone with ethyl β-aminopropionate. No attention was paid to the resolution of the optical isomers; the compounds used and synthesized were *dl*-isomers, except dipalmitoxypantothenyl palmitate which was prepared from *d*-pantothenyl alcohol.

(1) This work was supported by research grant No. A-257 from the National Institute of Health, U. S. Public Health Service, Department of Health, Education and Welfare.

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Experimental

Ethyl N-[α,γ-Dipalmitoxy-β,β-dimethylbutyryl]-β-aminopropionate (Ethyl Dipalmitoxypantothenate).—One and a half grams (0.01 mole) of β-alanine ethyl ester hydrochloride[13] was dissolved in 10 ml. of absolute methyl alcohol, and this solution was added to a solution of sodium methoxide in methanol. The sodium methoxide solution was prepared by dissolving 0.23 g. (0.01 mole) of sodium metal in 10 ml. of absolute methanol. To this mixture, 1.3 g. (0.01 mole) of pantolactone was added and allowed to stand for 12 hours. The sodium chloride precipitate was removed by filtration, and the solvent was completely removed under diminished pressure. The viscous oily residue was kept at 100° for two hours under vacuum. Then the residual sirup was immediately taken up in a mixture of 10 ml. of dry chloroform and 10 ml. of dry pyridine. Approximately 5.6 g. (0.02 mole) of palmitoyl chloride dissolved in 10 ml. of dry chloroform was added dropwise at room temperature with stirring. The stirring was continued for two hours. The dipalmitate was extracted with ether, and washed with 0.5 *N* hydrochloric acid, 5% sodium carbonate solution and with water until neutral. After drying over anhydrous sodium sulfate, the solvent was removed. The residue was recrystallized from 95% ethyl alcohol; yield 2.2 g. (29.3%); m.p. 57.0–58.5°. *Anal.* Calcd. for $C_{43}H_{81}O_7N$: C, 71.31; H, 11.28; N, 1.93. Found: C, 71.53; H, 11.21; N, 1.96.

The α-Palmitoxy-β,β-dimethyl-γ-hydroxybutyrolactone (Monopalmitoxypantolactone).—Five grams (excess) of pantolactone was dissolved in a mixture of 15 ml. of pyridine and 10 ml. of chloroform. Ten grams of palmitoyl chloride dissolved in 20 ml. of chloroform was then added dropwise at room temperature with stirring and the reaction mixture was set aside for 12 hours. The solvent was then removed under reduced pressure and the residue extracted thoroughly with ethyl ether. The solvent was removed, and the remaining solid residue was taken up in acetone. Petroleum ether (b.p. 39–41°) was added to the acetone solution until the mixture became slightly turbid. The mixture was then placed in the refrigerator. The precipitate was recrystallized from 95% ethyl alcohol; yield 7.5 g. (55.8%); m.p. 56.0–56.5°, fine needles. *Anal.* Calcd. for $C_{22}H_{40}O_4$: C, 71.49; H, 10.91. Found: C, 72.00; H, 10.81.

Ethyl N-[α-Palmitoxy-β,β-dimethyl-γ-hydroxybutyryl]-β-aminopropionate (Ethyl Monopalmitoxypantothenate).—One and a half grams (0.01 mole) of ethyl β-aminopropionate hydrochloride in 10 ml. of absolute methanol was added to 10 ml. of methanol containing an amount of sodium methoxide equivalent to 0.23 g. (0.01 mole) of sodium metal. To this solution, 3.7 g. (0.01 mole) of pantolactone monopalmitate was added, and dissolved completely by slight warming. The mixture was then allowed to stand for 48 hours. The solvent was removed under vacuum and the oily residue heated at 100° for two hours under vacuum. After cooling, the product was extracted with ethyl ether, and the ether layer washed with water, 5% sodium carbonate solution, and with water to neutrality. The ether extract was dried over anhydrous sodium sulfate. Upon removal of the solvent, a viscous oily substance was obtained, which was recrystallized from petroleum ether (b.p. 34–38°) in the refrigerator[14]; yield 2.0 g. (41.6%); m.p. 36.0°.

(13) The hydrochloride of β-alanine ethyl ester was prepared following the procedures reported by W. J. Hale and E. M. Honan, THIS JOURNAL, **41**, 770 (1919).

(14) When the ethyl monopalmitoxypantothenate was first obtained, it was a viscous oily substance at room temperature. Upon standing in a vacuum desiccator for a period of a week, it crystallized.

Anal. Calcd. for $C_{27}H_{51}O_6N$: C, 66.76; H, 10.58; N, 2.88. Found: C, 67.13; H, 10.68; N, 2.78.

α,γ-Dihydroxy-β,β-dimethylbutyramide (Pantamide).—The synthesis of the pantamide was carried out by two different methods.

(A).—Ten grams of pantolactone was dissolved in 50 ml. of absolute methyl alcohol. This solution was saturated with dry ammonia, and was set aside for 48 hours. At the end of this period, the solvent was removed under vacuum, and the residue recrystallized from ethyl acetate; yield 8.5 g. (74.5%); m.p. 129.0–130.0°. *Anal.* Calcd. for $C_6H_{13}O_3N$: N, 9.51. Found: N, 9.30.

(B).—Ten grams of pantolactone was dissolved in 10 ml. of water and was added to 10 ml. of concentrated ammonium hydroxide solution. Such mixtures were allowed to stand for two hours, for 24 hours or were refluxed for 8 hours. The water was then removed under diminished pressure. The residue was recrystallized from 95% ethyl alcohol, ethyl acetate or a methyl alcohol–ethyl acetate mixture; yield (example) 7.3 g. (64.5%); m.p. 129.0–130.0°. No depression in m.p. was observed when mixed with the pantamide prepared by the preceding procedures.

Anal. Calcd. for $C_6H_{15}O_4N$: C, 43.62; H, 9.15; N, 8.48. Calcd. for $C_6H_{13}O_3N$: C, 48.97; H, 8.90; N, 9.51. Found: C, 49.17; H, 8.73; N, 9.38.

Approximately 166 mg. (one millimole as pantoic acid ammonium salt) of each preparation was accurately weighed and dissolved in 15 ml. of neutral 95% ethyl alcohol. This solution was further diluted with 20-ml. portion of absolute ethyl ether, and titrated with 0.1 *N* alcoholic potassium hydroxide solution with phenolphthalein. One hundred sixty six mg. was the amount of ammonium pantoate theoretically equivalent to 10 ml. of 0.1 *N* alkali solution. Both of these compounds prepared under (A) and (B), however, did not require a single drop of the alkali solution before it showed an alkalinity to phenolphthalein. Under the same conditions, one millimole of pure ammonium palmitate consumed the theoretical amount of the standard alkali solution. Thus the compounds must have been pantamide.

α,γ-Dipalmitoxy-β,β-dimethylbutyramide (Dipalmitoxypantamide).—One and a half grams (0.01 mole) of pantamide was acylated with 5.5 g. (0.02 mole) of palmitoyl chloride in a mixture of pyridine–chloroform. The reaction product was extracted with chloroform, and washed with 0.5 *N* hydrochloric acid, water, 5% sodium carbonate solution and with water until neutral. After the extract had been dried over anhydrous sodium sulfate, the solvent was removed under reduced pressure. The residue was recrystallized from petroleum ether (b.p. 34–38°) and from 95% ethyl alcohol; yield 4.8 g. (77.0%); m.p. 74.0–75.0°; very fluffy amorphous powder. *Anal.* Calcd. for $C_{38}H_{73}O_5N$: C, 73.02; H, 11.77; N, 2.24. Found: C, 72.92; H, 11.77; N, 2.24.

α,γ-Dipalmitoxy-β,β-dimethylbutyric Acid (Dipalmitoxypantoic Acid).—Two grams of dipalmitoxypantamide was dissolved in 20 ml. of glacial acetic acid, and treated with 10 ml. of freshly prepared isoamyl nitrite.[15] The mixture was then placed on a steam-bath, and refluxed gently for two hours. The solvent was removed almost completely under vacuum, and the residue recrystallized from petroleum ether (b.p. 34–38°) in the refrigerator; yield 1.1 g. (55.0%); m.p. 62.0–63.0°. Calcd. for $C_{38}H_{72}O_6$: neut. equiv., 625. Found: neut. equiv., 627.

N-[α,γ-Dipalmitoxy-β,β-dimethylbutyryl]-3-amino-*n*-propyl Palmitate (Dipalmitoxypantothenyl Palmitate).—Two grams of *d*-pantothenyl alcohol (Nutritional Biochemical Corporation), was added to 30 ml. of chloroform containing 9 g. of palmitoyl chloride. To this mixture, 10 ml. of dry pyridine was added slowly with vigorous shaking at room temperature. As the reaction proceeded, the insoluble pantothenol went into the solution gradually and finally a clear reaction mixture was obtained, which was then set aside for 12 hours. The reaction product was extracted with chloroform and washed with water, 0.5 *N* hydrochloric acid, 5% potassium carbonate solution, and again with water successively. After drying the extract with anhydrous sodium sulfate, the solvent was removed and the residue recrystallized from 95% ethyl alcohol; yield 5.6 g. (60.9%); m.p. 49.0–53.0°; $[\alpha]^{34}D$ +9.5 (*c* 4.22%, in chloroform). *Anal.* Calcd. for $C_{57}H_{109}O_7N$: C, 74.37; H, 11.94; N, 1.52. Found: C, 74.59; H, 11.78; N, 1.56.

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URBANA, ILLINOIS

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The Synthesis of Long Chain Fatty Acid Derivatives of the Vitamin B_6 Group[1,2]

BY TAKETAMI SAKURAGI AND FRED A. KUMMEROW

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The following long chain fatty acid derivatives of the vitamin B_6 group have been prepared: pyridoxine tripalmitate, pyridoxine trilinoleate, pyridoxine trioctanoate, pyridoxine tridecanoate, pyridoxal dipalmitate, pyridoxamine tripalmitate, pyridoxine 3,5-dipalmitate, pyridoxal 3-palmitate, pyridoxine 5-palmitate and 4-desoxypyridoxine dipalmitate. The ultraviolet spectra of these derivatives were essentially the same, having one maximum absorption peak in the neighborhood of 265–270 mμ or at 283 mμ (pyridoxine 5-palmitate) in diethyl ether as a solvent.

Since 1938, several workers have synthesized pyridoxine triacetate[3–8] and the tribenzoate.[3,6] Selective acetylation of pyridoxine also made it possible to prepare the diacetate of the vitamin.[5,9]

Three patent descriptions[6–8] deal with the preparation of the fat-soluble derivatives by acylating with short chain fatty acids, such as acetic and propionic acids. The esters of long chain fatty acids, however, have as yet not been prepared. The present paper describes the preparation of several long chain fatty acid derivatives of the vitamin B_6 group. These derivatives were biologically active as a source of vitamin B_6 for rats.[10]

The fully acylated compounds of the vitamin B_6 group, which are listed in Table I, have been prepared by treating the corresponding vitamin hydrochloride with the respective fatty acid chlorides in pyridine. Of these derivatives, pyridoxine trioctanoate (I) and pyridoxine trilinoleate (I) were liquid at room temperature; the latter compound solidified at −18 to −19°. Others were obtained as

(1) This work was supported by research grant No. A-257 from the National Institute of Health, U. S. Public Health Service, Department of Health, Education and Welfare.

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