

Fig. 2. Separation of calcium and strontium from 1 ml. of milk. Eluting solution: 1 per cent tetra sodium salt of ethylene diamine tetraacetic acid. pH 8. Flow-rate, 0.17 ml./cm.²/min.; column-length, 16.5 cm.; temperature, 22° C.

column operation was experienced from the milk-fat component, and the resin was readily regenerated for further use. These techniques may therefore be applied satisfactorily to the separation of calcium-free strontium in high yield directly from liquid milk.

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¹ Davis, P. S., *Nature*, **183**, 674 (1959).

² Mayneord, W. V., Anderson, W., Bentley, R. E., Burton, L. K., Crookall, J. O., and Trott, N. G., *Nature*, **182**, 1473 (1958).

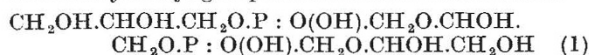
³ Hart, "Practical Physiological Chemistry", Thirteenth Edition, by Hawk, Oser and Summerson, 239 (1954).

⁴ Milton, G. W., and Grummit, W. E., *Canad. J. Chem.*, **35**, 541 (1957).

BIOCHEMISTRY

Position of the Fatty Acids in Cardiolipin

EVIDENCE was obtained previously¹⁻³ that cardiolipin is composed of two phosphoric acid, three glycerol and four fatty acid residues. According to this structure the polyglycerophosphate skeleton (1) has five hydroxyl groups available for esterification.



The position of the fatty acids has now been ascertained by identification of the fragments obtained on hydrolysis of cardiolipin in conditions in which splitting of the phosphoric diester bonds take place preferentially with little rupture of the fatty acid ester bonds.

Cardiolipin (620 μ moles phosphorus), prepared from ox heart muscle by the method of Faure and Morelec-Coulon⁴, was heated 90 min. in 50 ml. 90 per cent (v/v) acetic acid at 100° C., and after removal of the acetic acid *in vacuo* the water-soluble and ether-soluble fractions were separated. The ether-soluble fraction contained 68 per cent of the total glycerol, 19 per cent of the phosphorus and 98 per cent of the total fatty acids, only 5 per cent of which were free. By chromatography on silicic acid according to Borgström⁵ 76 per cent of the fatty acids were recovered in the diglyceride fraction (485 μ moles; molar ratio fatty acid-fatty acid ester-glycerol-phosphorus, 1.92 : 1.96 : 1.0 : 0 ; 1.7 double bonds/mole fatty acid); by periodic acid oxidation, not more than 2 per cent of this fraction was present as monoglyceride. The remaining fatty acids and the ether-soluble phosphorus were recovered in phosphatide fractions in partly degraded forms (phosphorus-glycerol-fatty acid, 1 : 1.1 : 2.2).

The water-soluble fraction of the hydrolysate contained 80 per cent (500 μ moles) of the phosphorus and 32 per cent (295 μ moles) of the glycerol, combined in a form hydrolysable by phosphomonoesterase; not more than 10 per cent of the phosphorus, however, was present as glycerolmonophosphate, as estimated by Burmaster's method⁶, and no free glycerol was detected. On fractionation 80 per cent of the water-soluble phosphorus was recovered as a barium salt sparingly soluble in water, corresponding in composition to barium glyceroldiphosphate (phosphorus-glycerol-barium, 2.0 : 1.06 : 2.2).

The main products of hydrolysis were therefore glyceroldiphosphate and diglycerides in molar proportion 1 : 2. These results are incompatible with the structure proposed for cardiolipin by Pangborn⁷ (a chain of four glycerol and three phosphoric acid residues, in which all six available hydroxyl groups are acylated), which entails the formation on hydrolysis of free fatty acid in proportion to the water-soluble phosphoric ester formed. The results are consistent with a structure for cardiolipin as a derivative of 1, in which the terminal glycerol residues are esterified as diglycerides, leaving a free hydroxyl group on the middle glycerol residue.

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¹ Macfarlane, M. G., and Gray, G. M., *Biochem. J.*, **67**, 25, P (1957).

² Gray, G. M., and Macfarlane, M. G., *Biochem. J.*, **70**, 409 (1958).

³ Macfarlane, M. G., *Nature*, **182**, 946 (1958).

⁴ Faure, M., and Morelec-Coulon, M. J., *Ann. Inst. Pasteur*, **91**, 537 (1956).

⁵ Borgström, B., *Acta Physiol. Scand.*, **30**, 231 (1953).

⁶ Burmaster, C. F., *J. Biol. Chem.*, **164**, 233 (1946).

⁷ Pangborn, M. C., *J. Biol. Chem.*, **168**, 351 (1947).

Differential Response of Human Serum Cholinesterase Types to an Inhibitor in Potato

DIFFERENCES in the degree of inhibition of the activity of human serum cholinesterase (pseudocholinesterase) produced *in vitro* by the local anaesthetic dibucaine (nupercaine) make it possible to classify individuals into three distinct types according to the character of the cholinesterase present in their serum¹. These types are inherited, and are thought to be determined by a pair of allelic genes each of which is responsible for the synthesis of a distinct form of