described. Protoverine 3,6,16-triacetate[1] on treatment with a limited amount of (*l*)-2-methylbutyryl chloride[6] afforded protoverine 15-(*l*)-2-methyl butyrate 3,6,16-triacetate, m.p. 234–235° dec., $[\alpha]^{28}D$ −4° (*c* 0.98, py.), which was stable toward sodium periodate but consumed 1.0 mol. eq. of chromic acid. Acetylation of the latter compound gave a pentaester identical with the product of acetylation of the monoester methanolysis product (III) from protoveratrine A. Thus the diester methanolysis product is IV, and protoveratrine A is protoverine 3-(*d*)-2-hydroxy-2-methylbutyrate 6,7-diacetate 15-(*l*)-2-methylbutyrate (I).[7,8]

(6) F. L. Weisenborn, J. W. Bolger, D. B. Rosen, L. T. Mann, Jr., L. Johnson and H. L. Holmes, THIS JOURNAL, **76**, 1792 (1954).

(7) Satisfactory analytical and spectral data were obtained for all the new compounds reported herein.

(8) We thank Dr. Harold A. Nash of the Pitman–Moore Company for a generous gift of protoveratrine A, and the National Institutes of Health (H-2275(C3)) and the Wisconsin Alumni Research Foundation for generous grants in support of this work.

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RECEIVED DECEMBER 24, 1958

THE USE OF HIGH EFFICIENCY CAPILLARY COLUMNS FOR THE SEPARATION OF CERTAIN *cis-trans* ISOMERS OF LONG CHAIN FATTY ACID ESTERS BY GAS CHROMATOGRAPHY[1]

Sir:

A new concept in gas chromatography was introduced by Golay,[2] who suggested the use of columns made by coating the inner surface of narrow bore capillary tubing with a thin layer of stationary phase. Such columns possess a performance and operating efficiency far greater than is possible with conventional packed columns. The very small quantity of stationary phase lining the inner surface of the capillary tube requires, however, sample loads in the region of one microgram or less if the performance of the column is to be realized in full. This in turn makes severe demands on the detector used to sense the low vapor concentrations emerging from the column. An ionization detector with a sensitivity of 10^{-13} mole and a sensing volume of only a few microliters was described recently by Lovelock.[3] This detector, modified, formed part of the apparatus used in this investigation.

Table I shows the results of a gas chromatographic analysis of a known mixture of the methyl esters of saturated and unsaturated fatty acids extending from C-8 to C-20. A 200 foot stainless steel capillary column with an internal diameter of 0.010 inch which was coated with Apiezon "L" was used. The column was maintained at 240°. The inlet pressure of the argon carrier gas was 0.68 atm.; the outlet flow rate was 0.5 ml./min. The sample was introduced into the column by means of a T-shaped glass bypass device maintained at 300°. In this manner approximately 99.9% of the volatilized sample was vented to the atmosphere. The remainder, approximately one gamma, entered into the capillary column.

TABLE I

Methyl ester	Composition, %	Corrected retention time, min.	S_f[a]	Calcd. theor. plates
Octanoate	4.1	2.8	0.04	21,400
Nonanoate	6.8	4.4	.07	25,400
Decanoate	3.5	6.4	.10	30,600
Undecanoate	1.2	9.6	.15	40,000
Laurate	9.0	14.1	.21	55,400
Tridecanoate	2.9	20.6	.31	60,200
Myristate	10.7	30.6	.47	64,100
Pentadecanoate	0.2	44.4	.68	80,800
Palmitoleate	1.4	58.6	.89	101,800
Palmitate	14.2	65.6	1.00	36,800
Margarate	4.4	95.1	1.45	94,500
Linolenate	7.5	118	1.80	...
Linoleate	3.7			
Oleate	14.9	124	1.89	31,800
Elaidate	2.2	125	1.91	76,500
Stearate	8.4	139	2.12	59,200
Arachidonate	1.7	204	3.11	200,000
Arachidate	3.2	296	4.52	128,000

[a] Separation factor based on methyl palmitate equal to 1.00.

Under these experimental conditions an extremely efficient column was obtained making possible for the first time the separation of certain *cis–trans* isomers, *i.e.*, methyl elaidate from methyl oleate (Table I).

The highest calculated theoretical plate efficiency for any one component was 200,000 (methyl arachidonate) or 1,000 plates per foot. Despite the fact that the Apiezon coated capillaries provided excellent efficiencies, the separation of methyl linoleate from methyl linolenate was not achieved.

Preliminary experiments employing capillary columns containing certain polyesters as stationary liquids[4] provided the rapid resolution of most components including linoleate and linolenate with good separation factors but low theoretical plate efficiencies.

(1) This work was supported by the National Heart Institute of the National Institutes of Health, the National Dairy Association and the Nutrition Foundation.

(2) M. J. E. Golay, "Gas Chromatography," Academic Press, Inc., New York, N. Y., 1958.

(3) J. E. Lovelock, *Nature*, **182**, 1663 (1958).

(4) S. R. Lipsky, R. A. Landowne and M. R. Godet, *Biochim. Biophys. Acta*, **31**, 336 (1959).

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RECEIVED DECEMBER 18, 1958

SYNTHESIS OF 18-OXYGENATED PROGESTERONES

Sir:

Steroid metabolites, oxygenated at C-18 but lacking an oxygen at C-11, have been detected recently.[1,2] However, biological evaluation of this new type of compounds has been hampered because of the minute amounts available. We wish to report therefore a practical method for the conversion of the readily available alkaloid conessine (I) to C-18 oxygenated progesterones and other related steroids.

Conessine (3β-dimethylamino-con-5-enine,[3] I) on treatment with sodium borohydride and aluminum

(1) K. H. Loke, G. F. Marrian, W. S. Johnson, W. L. Meyer and D. D. Cameron, *Biochim. Biophys. Acta*, **28**, 214 (1958).

(2) R. Neher and A. Wettstein, *Helv. Chim. Acta*, **39**, 2062 (1956).

(3) R. D. Haworth and M. Michael, *J. Chem. Soc.* 4973 (1957).