(60:20:0.4) on development gave a spot with the same R_f as methyl lithocholate.

Gas liquid chromatography (GLC) of this material showed several peaks, one of which had the same retention time as methyl lithocholate in two systems (3% QF-1 and 1.5% SE-52 on Gas Chrom Q); relative retention times of 2.06 (relative to cholesterol-5.2 min) and 1.10 (relative to cholesterol-11.4 min) respectively. GLC conditions were: for 3% QF-1, column temperature 240 C, for 1.5% SE-52, column temperature 220 C; for both columns, detector 270 C, and flash heater 270 C. The carrier gas was nitrogen at a flow rate of 40 ml/min. The columns were 2 m in length with 4 mm i.d. It has previously been shown that lithocholic acid methyl ester can be separated from the related isomers on QF-1 and on a phenyl-containing phase (5). Examination of the methylated fraction by GLC mass spectrometry revealed that the peak with a retention time equivalent to a methyl lithocholate standard (13.3 min) had a spectrum with major peaks at M/e 372 (M-18), 357 (M-[18 + 15]), 318, 257 (M- [18 + 115]) and 215. GLC-mass spectrometry conditions on LKB 9000 were: column temperature 223 C, flash heater, 268 C, molecular separator 258 C, and helium flow 30 ml/min. The column was a 1.5% SE-52 column 2 m in length with 4 mm i.d. The filament temperature was 270 C and the ionizing beam was at 70 ev. Spectra of the compound from EAE guinea pig brains (1), a lithocholic acid standard, and the human MS brain are shown in Figure 1. The spectra of the MS material is similar to that of the others shown and also to other published spectra (6).

The identification of lithocholic acid in the EAE brains of guinea pigs and human multiple sclerosis brain raises the question of its origin. Experiments are underway in our laboratory to determine whether normal or EAE brain tissue possesses the capacity to form this or similar bile acids from cholesterol. It is also possible that lithocholic acid and other related acids are present in normal brain tissue, but in concentrations much smaller than those found in diseased tissue. Further work will also be directed toward establishing the precise location of the acid within the tissue. It has not been possible to extimate the quantity of lithocholic acid present in the specimen examined.

S. H. MUJTABA NAQVI[1]
ROBERT B. RAMSEY
H. J. NICHOLAS
St. Louis University School of Medicine
St. Louis, Missouri 63104

[1]On leave from the Pakistan Atomic Energy Commission when this work was conducted.

ACKNOWLEDGMENTS

Supported by National Institute of Health Grant NB-06011. R. T. Aexel assisted in the GLC and mass-spectrometer analyses, which were supported by NIH Grant AM-09992. W. W. Tourtellotte, University of Michigan Medical Center, provided the formalin-fixed coronal sections.

REFERENCES

1. Nagvi, S. H. M., B. L. Herndon, M. T. Kelley, V. Bleisch, R. T. Aexel and H. J. Nicholas, J. Lipid Res. 10:115 (1969).
2. Willmon, T. L., Ann. N.Y. Acad. Sci. 122:1 (1965).
3. Roizin, L., and L. C. Kolb, "'Allergic' Encephalomyelitis," Edited by M. W. Kies and E. C. Alvord, Jr., Charles C. Thomas, Springfield, 1959.
4. Palmer, R. H., Report to the U.S. Atomic Energy Commission, ACHR-1000-42, Clearinghouse for Federal Scientific and Technical Information, 1967.
5. Elliott, W. H., L. B. Walsh, M. M. Mui, M. A. Thorne and C. M. Siegfried, J. Chromatog. 44:452 (1969).
6. Ryhage, R., and E. Stenhagen, J. Lipid Res. 1:361 (1960).

[Received February 26, 1970]

Pancreatic Lipolysis of Enantiomeric Triglycerides[1]

ABSTRACT

Pancreatic lipase hydrolyzed fatty acids in equimolar quantities from the *sn*-1- and 3-positions of three synthetic enantiomeric triglycerides, two of which could make a racemic pair. The monoglycerides from digestions of five enantiomeric triglycerides were at least 99% representative of the 2-position. The data confirm that pancreatic lipase did not distinguish between the *sn*-1- and 3-positions and that with these triglycerides pancreatic lipolysis can be used to help establish structure.

[1]Scientific contribution No. 419 Agricultural Experiment Station, University of Connecticut, Storrs.

As part of the preparation of some cryptoactive triglycerides for investigating a method

TABLE I

Fatty Acid Composition of Intact Triglycerides and of Monoglycerides and Free Fatty Acids Derived From Pancreatic Lipolysis of Enantiomeric Triglycerides

	Intact triglyceride			Monoglyceride			Free fatty acids		
Triglyceride	16:0	18:0	18:1	16:0	18:0	18:1	16:0	18:0	18:1
	Mole %								
18:1-18:1-16:0[a]	33.0	---	67.0	Trace	---	99+	48.3	---	51.7
18:1-16:0-16:0	67.6	---	32.4	99+	---	Trace	48.0	---	52.0
16:0-16:0-18:1	68.0	---	32.0	99+	---	Trace	49.4	---	50.6
16:0-18:0-18:1	33.1	33.3	33.6	0.3	99.4	0.3	---	b	---
18:1-18:0-16:0	33.1	34.2	32.7	Trace	99.5	0.5	---	b	---

[a]Triglycerides are numbered 1,2 and 3 from left to right in *sn* nomenclature; 18:1-18:1-16:0 would be *sn*-glycerol-1,2-dioleate-3-palmitate.

[b]Not done.

of stereospecific analysis (1), positional integrity was determined by pancreatic lipolysis. Although pancreatic lipase is known not to be stereospecific (2,3), very few data are available on the pancreatic lipolysis of synthetic enantiomeric triglycerides.

The triglycerides used as substrates were *sn*-glycerol-1,2-dioleate-3-palmitate (18:1-18:1-16:0), *sn*-glycerol-1-oleate-2,3-dipalmitate (18:1-16:0-16:0), *sn*-glycerol-1,2-dipalmitate-3-oleate (16:0-16:0-18:1), *sn*-glycerol-1-palmitate-2-stearate-3-oleate (16:0-18:0-18:1) and *sn*-glycerol-1-oleate-2-stearate-3-palmitate (18:1-18:0-16:0). These were made by standard procedures (4) from *sn*-3-acetone glycerol (5). The purity of the intermediate 1,3-diglycerides, determined as described by Sampugna and Jensen (1) was close to 99%. The pancreatic lipolysis procedure and recovery and analysis of the lipolysis products have been described (1).

Compositional data for the monoglycerides and in three cases, the free fatty acids are given in Table I. The free fatty acids were hydrolyzed from positions *sn*-1- and 3- in close to equimolar quantities, therefore pancreatic lipase did not differentiate between the two positions and earlier findings are confirmed (2,3). Since this was the case with both enantiomers of a racemic pair (18:1-16:0-16:0 and 16:0-16:0-18:1) composition of the fatty acids was not involved. The monoglycerides were more than 99% representative of the 2-position. The presence of monoglycerides, is, in itself, proof that pancreatic lipase is not stereospecific because a stereospecific lipase would hydrolyze either the *sn*-1- or 3-position only, leaving a diglyceride and no monoglyceride.

R. G. JENSEN
R. E. PITAS
Department of Animal Industries
University of Connecticut
Storrs, Connecticut 06268
J. G. QUINN
Graduate School of Oceanography
University of Rhode Island
Kingston, Rhode Island
J. SAMPUGNA
Chemistry Department
University of Maryland
College Park, Maryland

ACKNOWLEDGMENT

Supported in part by Public Health Service Research Grants AM-02605-9-11 from the Institute of Arthritis and Metabolic Diseases and by U.S. Department of Agriculture Grant No. 12-14-100-7660(73).

REFERENCES

1. Sampugna, J., and R. G. Jensen, Lipids 3:519-529 (1968).
2. Karnovsky, M. L., and D. Wolff, "Biochemistry of Lipases," Vol. 5, Edited by G. Popjak, Pergamon Press, New York, 1960, p. 53-59.
3. Tattrie, N. H., R. A. Bailey and M. Kates, Arch. Biochem. Biophys. 78:319-327 (1958).
4. Quinn, J. G., J. Sampugna and R. G. Jensen, JAOCS 44:439-442 (1967).
5. Baer, E., "Biochemical Preparations," Vol. 2, Edited by E. G. Ball, John Wiley and Sons, Inc., 1952, p. 31-38.

[Received January 12, 1970]