

hope to further establish the utility of this enzyme in TG structure investigations.

R. G. JENSEN

J. SAMPUGNA

J. G. QUINN

Department of Animal Industries, University of Connecticut, Storrs, Connecticut

ACKNOWLEDGMENT

Supported in part by Public Health Service Research Grant AM-02605-08 from the Institute of Arthritis and Metabolic Diseases.

REFERENCES

1. Alford, J. A., and D. A. Pierce, *J. Food Sci.* **26**, 518 (1961).
2. Alford, J. A., D. A. Pierce and F. G. Suggs, *J. Lipid Res.* **5**, 390 (1964).
3. Brockerhoff, H., *J. Lipid Res.* **6**, 10 (1965).
4. Brockerhoff, H., *Lipids* **1**, 162 (1966).
5. de Haas, G. H., L. Sarda and J. Roger, *Biochim. Biophys. Acta* **106**, 638 (1965).
6. Jensen, R. G., J. Sampugna, J. G. Quinn, Dorothy L. Carpenter, T. A. Marks and J. A. Alford, *JAOCS* **42**, 1029 (1965).
7. van Deenen, L. L. M., in "Metabolism and Physiological Significance of Lipids," edited by R. M. C. Dawson and D. N. Rhodes, John Wiley & Sons Ltd. New York, 1964 p. 155.

[Received May 13, 1966]

A Solved Problem of Triglyceride Analysis

Sir: A complete stereospecific analysis of triglyceride mixtures requires a considerable amount of data. However a recent letter in your March issue incorrectly states that "with our present methods this is still an impossible task." We wish to affirm that the difficulties can be overcome with perseverance and the methods now available.

The triglycerides that contain only one type of acid cause no difficulty. Fortunately the triglycerides that contain two different types of acid can often be resolved to a greater extent than the total number of alkene groups in the molecule would indicate (e.g., 18:0-18:0-18:2 is separable from 18:0-18:1-18:1 and 18:0-18:0-18:3 from 18:0-18:1-18:2 on AgNO₃-silicic acid plates). The triglyceride species with only two acids have only 3 possible isomers and are easily analyzed.

The triglycerides that contain three different acids require the most consideration in structure determination. They can be readily classed into three isomeric pairs which must then be further resolved.

Position	Triglycerides		
1	AB	AC	BC
2	BA	CA	CB
3	CC	BB	AA

The relative amounts of these triglycerides most probably reflects the availability of the different diglyceride precursors. With no steric selectivity, the esterification of C at position 3 would be expected to produce ABC and BAC in the same relative abundance as the pairs ABB and BAB or ABA and BAA are produced. The latter sets of triglycerides are isomeric pairs that are easily analyzed and could be used to suggest the ABC/BAC ratio.

A fully rigorous structure proof of the three-acid triglycerides requires only the following steps:

- (a) Cleavage of the six-isomer mixture to 1,2- and 2,3-diglycerides by triglyceride lipase.
- (b) Isolation and analysis of the 1,2-diacylglycerol-3-phosphates produced by diglyceride kinase action as described in Federation Proceedings **25**, 521 (1966).
- (c) Separation of the three isomeric pairs of phosphatidate derivatives according to their acid content.

AB	AC	BC
BA	CA	CB
PP	PP	PP
x	y	z

The relative amounts of x, y and z are indicated by the content at the 3 position of C, B and A respectively.

- (d) Phospholipase A cleavage of each pair to determine the AB/BA, AC/CA and BC/CB ratios.

If the results of the unequivocal analysis show that the AB/BA ratio is constant in all the triglyceride species, future analyses could then use the values obtained from the more easily measured diacid triglycerides. We hope these comments will adequately indicate that the difficulties of triglyceride analysis can be overcome.

WILLIAM E. M. LANDS

SISTER P. M. SLAKEY

Department of Biological Chemistry University of Michigan, Ann Arbor, Michigan

[Received April 28, 1966]