

TABLE I
A Comparison of the GLC-Method of
Analysis for myo-Inositol with Chemical
and Microbiological Methods.

Sample	μg Myo-Inositol		
	GLC	Microbiological	Chemical
Kidney PIC (3)	198	166	197
Brain TPI (5)	480	520	493
	Literature values		
		Dawson & Hauser (9)	Freinkel (10)
Brain ^a (7)	672 \pm 125	730 \pm 104	—
Testes ^a (7)	256 \pm 76	—	280 \pm 70
Kidney ^a (8)	546 \pm 141	879 \pm 223	610 \pm 260

^a Data expressed as μg myo-inositol per gram wet weight of tissue.

PIC — phosphoinositide complex.

TPI — triphosphoinositide.

Numbers in parentheses represent numbers of experiments.

Chromatographic conditions: Instrument, Barber-Colman (radium cell - 0.056 μC); column, $\frac{1}{4}$ in. \times 6 ft. glass column, U-tube; 3% SE - 30 on Chromosorb W; column temperature, 145°C; flash heater (inlet), 210°C; cell temperature (detector), 190°C; inlet pressure, 18 psi (argon); detector sensitivity, 10/2; chart speed, 7.5 in./hr; reaction time, 12 min.

phosphoinositide (8) of beef brain and on tissue extracts. It can be seen that there was satisfactory agreement between the values obtained when using the GLC method as contrasted with the older methods. The agreement between these methods and the reproducibility of our analytical data obtained over a period of time using GLC for myo-inositol analysis indicates the validity of the procedure.

The use of DMSO, thus, increases the utilization of GLC for analysis of myo-inositol by decreasing the time necessary for the formation of TMS derivatives.

This procedure has also been found by Hamilton and his colleagues (6) to be very useful in the analysis of other polyhydroxy compounds.

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A Sensitive Method for Phosphorus in Lipids¹

THERE ARE MANY METHODS for the wet ashing of lipid material, and there are many variations of the molybdenum blue colorimetric determination of ortho-phosphate (1). Perchloric acid wet ash methods are the most rapid, and stannous chloride as a reducing agent gives great sensitivity in the colorimetric determination. But one cannot use stannous chloride as the reducing agent in the colorimetric determination in the presence of excess perchloric acid. The method reported here enjoys both speed of digestion with perchloric acid and high sensitivity with stannous chloride. The excess perchloric acid is evaporated before the color is developed.

A solution of lipid material that contained at least 1 μg of phosphorus was put in a 30 ml Kjeldahl flask. A few glass beads (5 mm in diameter) were put in the flask, and the solvent was evaporated on a hot plate. Then, 1 ml of concentrated HNO_3 was added, and the flask was heated until the initial oxidation was completed. This was indicated by the formation of a homogenous solution and the subsidence of vigorous boiling and foaming. The flask was cooled, and 1 ml of 70% HClO_4 /0.2 g of lipid was added. The flask was heated with a microburner under a perchloric acid fume head, such as described by Diehl and Smith (2), until heavy white fumes were evolved and the digest was clear, indicating that the digestion was complete. The entire digestion procedure takes about 5 min for the average sample.

The excess perchloric acid was evaporated rapidly

by placing the flask in a second fume head which was pierced by a piece of glass tubing which extended into the neck of the Kjeldahl flask. Pyrex wool was packed in the space between the fume head and the neck of the flask, so that the head drew a current of air through the tube and into the Kjeldahl flask and quickly evaporated the acid as the flask was heated.

The digested material was transferred to a 25 ml glass stoppered volumetric flask, and the analysis was completed by Fontaine's (3) colorimetric phosphorus determination. For samples containing about 1 μg of phosphorus, greater accuracy was obtained by using a 10 ml volumetric flask and scaling down the amounts of reagents proportionately. One can detect 0.5 μg of phosphorus using this procedure. The use of buffers (1) to increase the sensitivity was investigated. The sensitivity was increased, but the results were not reproducible. A standard curve for phosphorus was prepared by using known quantities of anhydrous KH_2PO_4 . The calibration curve needs to be determined only once.

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