

thiamine deficiency, while Group V was normal. The fat contents were $2.1 \pm 0.18^*$ % for Group IV; 4.3 ± 0.93 % for Group V and 3.7 ± 0.44 % for Group VI. These figures demonstrate that both Groups V and VI had synthesized fat from carbohydrate (P values for differences with Group IV < 0.05 and < 0.01 respectively).

Their mutual difference has no statistical significance whatever ($P > 0.5$).

Hence neither of these experiments has given the slightest indication that thiamine deficiency causes an impairment of the synthesis of fat from carbohydrate.

This work forms part of investigations on the metabolism and physiological function of thiamine carried out by H. G. K. WESTENBRINK and collaborators. Full details and discussion will appear shortly⁷.

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* Standard deviation of the mean.

A RAPID DETERMINATION OF GLYCOGEN IN TISSUES

by

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A quick method for the determination of glycogen in tissues has been elaborated by combining a rapid procedure of extraction, viz. grinding the tissue sample with sand and trichloroacetic acid, with the very simple spectrophotometric method of glucose determination according to MENDEL AND HOOGLAND¹. The latter makes use of a pink colour which develops when concentrated sulphuric acid is added to a glucose solution and the mixture is placed in a boiling waterbath for a few minutes. The glycogen need not be hydrolyzed before performing the glucose determination as 7 minutes' heating with the only slightly diluted sulphuric acid is sufficient to bring about hydrolysis of the glycogen as well as development of the colour.

The procedure is described below for the determination in rat liver and muscle tissue. The practically negligible amounts of free glucose present are included in the determination.

1 g of liver is ground in a mortar with 1 g quartz sand (purified by boiling with potassium bichromate and sulphuric acid) and 10 ml of a solution of 5% trichloroacetic acid and 0.1% silver sulphate, until a very fine suspension is obtained (this point is reached in about 2 minutes). The suspension is transferred to a volumetric flask of 50 ml, the mortar is rinsed with 10 ml of the trichloroacetic acid-silver sulphate solution and the volume is made up with water to 50 ml. Quartz sand and precipitate are removed by centrifuging.

According to MENDEL AND HOOGLAND the glucose determination is carried out by adding 1 ml glucose solution to 3 ml concentrated sulphuric acid, placing the mixture in a boiling waterbath for 7 minutes and measuring the extinction at 515 m μ . As the most suitable concentrations for a determination are situated between 100 and 200 γ glucose/ml, the glycogen determination can be carried out in 1 ml of the trichloroacetic acid-silver sulphate extract, when the glycogen concentration of the tissue does not exceed 1%. Extracts with higher contents must be diluted.

For muscle the following modifications are introduced in the above-mentioned procedure: 2 g of quartz sand should be used instead of 1 g; in order to obtain a sufficiently fine suspension it is

necessary to grind twice for about 2 minutes. It is advisable to interrupt the grinding during 1 minute after the first 2 minutes.

The efficiency of extraction and hydrolysis were checked by PFLÜGER's method: liberation and isolation of glycogen by treatment of the tissue with 60% KOH, precipitation and washing of the glycogen with ethanol, followed by hydrolysis with *N* sulphuric acid in a boiling waterbath for 2 hours.

The results are assembled in Table I. It is clear from the good agreement observed between the results obtained by both methods, that the lengthy procedure of PFLÜGER may be replaced by this rapid determination, which offers great advantages for routine analyses.

TABLE I
GLYCOGEN IN RAT LIVER AND MUSCLES

Rat No.	Condition of rat	Tissue	% glycogen	
			Extraction with trichl. acet. acid; glucose determ. without previous hydrolysis (duplicates)	Isolation of glycogen according to PFLÜGER; glucose determ. after hydrolysis (duplicates)
1	normally fed	liver	2.74; 2.67	2.40; 2.51
2	normally fed	liver	4.55; 4.47	4.25; 4.39
3	24 hrs fast	liver	1.09; 1.12	1.08; 1.12
4	24 hrs fast	liver	0.59; 0.59	0.53; 0.57
5	24 hrs fast	liver	0.97; 0.99	0.96; 0.97
6	normally fed	abdominal muscle	0.95; 0.90	0.85; 0.85
7	normally fed	abdominal muscle	0.51; 0.48	0.52; 0.58
8	normally fed	abdominal muscle	0.67; 0.65	0.60; 0.63
9	24 hrs fast	abdominal muscle	0.19; 0.21	0.18; 0.18
10	normally fed	leg muscle	0.24; 0.25	0.28; 0.23
11	normally fed	leg muscle	0.35; 0.34	0.32; 0.35
12	24 hrs fast	leg muscle	0.09; 0.09	0.08; 0.08

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¹ B. MENDEL AND P. L. HOOGLAND, *Lancet*, 1950 II, p. 16.

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FRACTIONATION OF THYROID CELLS

by

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From the earlier cytomorphological work about thyroid certain deductions have been made, concerning the rôle of cell organoids in the intracellular elaboration of the hormone^{1,2}. A possibility to study the properties of isolated organoids on analytical way was recently demonstrated by fractionations of liver cells^{3,4,5}. Thus, in order to investigate the chemical topography of the cell of thyroid gland, experiments have been performed in which the fractions of thyroid organoids were isolated and subjected to analytical procedures.