10500 P

Carbohydrates of Certain Vegetables and Fruits.

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A method is presented for the direct determination of the carbohydrate fraction of several classes of foods. The principal features of the method are: Materials very low in carbohydrate are concentrated by air-drying below 60°C until the residue can be pulverized. The soluble sugars are separated from the starch by solution in 60% ethyl alcohol. After inversion of the sucrose the sugars are determined before and after fermentation. The starch is separated from the unavailable carbohydrate residue by pancreatin digestion and further acid hydrolysis of the filtrate to convert the starch into glucose which is determined as such. The residue is then dissolved in 21.4 N sulfuric acid and further hydrolyzed to convert the cellulose into glucose, the hemicellulose into non-fermentable sugars. The lignin which is insoluble in dilute acid is determined gravimetrically.

Data obtained by this method and certain conventional procedures in a composite analysis account for more than 98% of the carbohydrate in a variety of common foods. Our results compare favorably in regard to available carbohydrates with those obtained by reliable direct determinations in other laboratories. We know of no data with which to compare our results on the unavailable carbohydrate fraction. In a previous publication[1] we emphasized the errors of the old crude fiber determinations.

10501 P

Aminoethyl Phosphoric Ester in the Small Intestine of Rabbits and Pigs.

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Outhouse reported the isolation of this ester from bovine malignant tumors, but found no trace of it in benign tumors or in the normal tissues investigated which included muscle, placenta, pancreas, liver

[1] Williams, R. D., and Olmsted, W. H., *J. Biol. Chem.*, 1935, **108**, 653.

and embryo.[1, 2] Consequently, he expressed the belief that this ester is peculiar to malignant tissue.

In a study of the acid-soluble phosphoric esters of the intestine in which a method of fractionation similar to that of Outhouse was used, small amounts of a crystalline quinine salt were isolated from both rabbit and pig intestine. This quinine salt dissociated when boiled in ethyl alcohol, leaving a crystalline residue which after several recrystallizations from 80 to 90% ethyl alcohol showed a N and P content close to the theory for aminoethyl phosphoric ester. A sample of synthetic ester, prepared in this laboratory by Dr. Welch according to the method of Plimmer and Burch,[3] was in all respects identical with the ester isolated from the intestine.

A much better yield of this substance was obtained by means of a new method involving fractionation of the phosphoric esters as the uranium salts. This method is based on the fact that uranium acetate precipitates aminoethyl phosphoric ester, nucleotides and other esters quantitatively from a neutralized trichloracetic acid extract of intestine, but forms soluble salts with compounds which contain no phosphorus. All other salts of aminoethyl phosphoric ester, including the basic lead salt, were found to be water-soluble. By shaking the uranium salts with a solution of $Ba(OH)_2$ the aminoethyl phosphoric ester is extracted as the Ba salt, leaving most of the other esters in the precipitate. After a second precipitation with uranium, extraction with $Ba(OH)_2$, concentration of the barium-free solution, and removal of a small precipitate formed on addition of mercuric acetate, the ester can be crystallized directly from 90% alcohol. After several recrystallizations and drying of the substance at 110°, a sample showed 10.2% N and 21.8% P; calc. for $C_2H_8O_4NP$; N 9.9%, P 22.0%.

In a preparation in which 137 g of rabbit intestine were used the yield of the ester per 100 g wet weight was 40 mg, which is about the same as the amount found in malignant tumors by Outhouse. It is estimated that the actual content in the small intestine is about twice as high, so that this ester would account for about ¼ of the acid-soluble organic P.

The barium hydroxide insoluble uranium fraction contains large amounts of a nucleotide which has been isolated in crystalline form and identified as adenylic acid; it corresponds roughly to ¼ of the acid-soluble organic P. The adenylic acid may not occur as such in the

[1] Outhouse, E. L., *Biochem. J.*, 1936, **30**, 197.

[2] Outhouse, E. L., *Biochem. J.*, 1937, **31**, 1459.

[3] Plimmer, R. H. A., and Burch, W. J. N., *Biochem. J.*, 1937, **31**, 398.

intestine, but may be formed during the course of the preparation from some more complex nucleotide. A third fraction corresponding to 12 to 16 mg P per 100 g intestine is not precipitated by uranium. The nature of the phosphate esters in this fraction has not been established.

Summary. A fractionation of the acid-soluble organic phosphoric esters of the small intestine as the uranium salts has been described. Aminoethyl phosphoric ester and adenylic acid have been isolated in crystalline form, each comprising about ¼ of the acid-soluble organic P of the rabbit small intestine. The occurrence of the former ester in pig intestine has also been demonstrated.

10502

Effects of Caffeine on Human Sugar-Tolerance Curves.*

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There is considerable evidence that caffeine affects the blood sugar of animals. In dogs, a rise follows caffeine injection[1] amounting in some cases to 40-60% above normal[2] in both arterial and venous blood;[3] a double dose is necessary to produce hyperglycemia during avitaminosis.[4] During work,[5] the sugar tolerance curve rises more rapidly with caffeine than without and stays up longer.

However, caffeine produces no hyperglycemia in castrated female rabbits,[6] nor does it affect the glycogen contents of the heart or liver in rats and guinea pigs.[7]

No report could be found regarding the effect of caffeine on the blood sugar or sugar tolerance curve for man.

Eight sugar tolerance curves were determined on the same subject (M.D.) using the sugar method of Folin and Malmros.[8] The nutri-

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1 Karger, K., *Klin. Wochschr.*, 1927, **6**, 1994.

2 Labbe, H., and Theorodesco, B., *Compt. rend.*, 1924, **178**, 886.

3 Bömer, M., *Arch. exp. Path. Pharmakol.*, 1930, **149**, 247.

4 Collazo, J. A., and Gohse, S. N., *Biochem. Z.*, 1923, **139**, 285.

5 Atzler, E., *Arbeitsphysiol.*, 1938, **10**, 30.

6 Laufberger, W., *Z. ges. exp. Med.*, 1924, **39**, 487.

7 Lasch, F., and Triger, K., *Z. ges. exp. Med.*, 1933, **88**, 588.

8 Folin, O., and Malmros, H., *J. Biol. Chem.*, 1929, **83**, 115.