

Analysis of glycosylated forms of vitamin B₆ in plant-derived foods. Velimatti Ollilainen,* Anja Pakkanen, Liisa Vahteristo & Pertti Varo

Department of Applied Chemistry and Microbiology, FIN-00014 University of Helsinki, Finland.

Free and phosphorylated B₆ vitamers usually compose the predominant part of food vitamin B₆. However, in plant foods glycosylated pyridoxine is the major vitamin, dominating especially in vegetables and fruits. Glycosylated vitamers can be utilized as sources of vitamin B₆ to some extent, but their bioavailability is limited (Gregory *et al.*, 1991; Gibert & Gregory, 1991).

In this study glycosylated and non-conjugated vitamers were extracted with perchloric acid and separated using reversed-phase liquid chromatography. Post-column derivatization with bisulphite reagent was used to enhance the fluorescence of B₆ vitamers, especially PLP. The amount of pyridoxine was measured before and after β -glucosidase treatment of the sample extract, and the increment of pyridoxine was assumed to be derived from the enzymatic hydrolysis of conjugated vitamers. In certain samples pyridoxine glucoside was also characterized using proton NMR spectroscopy.

In the cereals and vegetables studied, a significant part of vitamin B₆ constituted conjugated pyridoxine. In cereals, around 20–50% of the total amount of vitamin B₆ was derived from glycosylated pyridoxine. The amount in carrot, broccoli and tomato was 70%, 40% and 30%, respectively. Thus more information on the presence and chemical structures of glycosylated B₆ vitamers in foods is needed in order to estimate the significance of glucosylated vitamers as dietary sources of vitamin B₆ in plant-derived foods.

Gregory III, J. F., Trumbo, P. R., Bailey, L. B., Toth, J. P., Baumgartner, T. G. & Cerda, J. J. (1991) *J. Nutr.*, **121**, 177–86.

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*To whom correspondence should be addressed.

Analytical problems in the study of the dietary significance of natural toxicants in plant foods. Michael J. C. Rhodes* & Keith R. Price.

Food Molecular Biochemistry Department, Institute of Food Research, Norwich Research Park, Norwich NR4 7UA, UK.

Many secondary metabolites present in plant food are considered as natural toxicants. However there is increasing evidence which links some of these same compounds (including isothiocyanates in Brassicas and alkenyl polysulphides in Alliums) with the protective effects against cancers and degenerative diseases associated with diets rich in fruit and vegetables. We are interested in one such group of compounds, the flavonoids, which are thought to have an overall beneficial effect due to their activity as direct antioxidants, chelating agents and inducers of phase II anticancer enzymes.

In plant foods, flavonoids and related phenolic compounds exist in a multiplicity of complex conjugates with sugars and organic acids. Important questions remain on the fate of such conjugates during processing and digestion, on the nature of the nutritionally important form of flavonoids and on the extent of their uptake and metabolism in the gut. An important prerequisite for tackling such problems is the development of convenient high resolution separation systems to resolve and quantify flavonoid aglycones and conjugates. We will describe the use of the combination of diode-array based HPLC and capillary electrophoretic methods to study flavonoids and their conjugates applied to important dietary sources of flavonoids.

*To whom correspondence should be addressed.

Different biological forms of mercury. Marjatta Kantola.

University of Kuopio, Dept. of Chemistry, P.O. Box 1627, 70211 Kuopio, Finland.

In biological systems mercury can exist both in organic and inorganic forms. By inorganic mercury it is meant elemental Hg (Hg⁰), mercurous (Hg₂²⁺) and mercuric salts (Hg²⁺) as well as mercury in such complexes in which Hg²⁺ is reversibly bound to different ligands. The general formula for organic Hg compounds is R-Hg-R' or R-Hg⁺X⁻. R and R' are organic groups in which a carbon atom is covalently bound to Hg. Dialkyl and diaryl mercury compounds are nonpolar and volatile with variable toxicity. Their existence in nature is small in comparison with the existence of methylmercury compounds. Methylmercury compounds include all compounds derived from a methylmercury cation (CH₃Hg⁺). The mercury released to the environment as a metal (e.g. by losses from industry or from compounds like fungicides) or as inorganic forms like HgS (from sulphide ores) is converted to CH₃Hg⁺ by biological methylation. Because methylmercury compounds are lipophilic, they accumulate much more efficiently in mammals than inorganic mercury and therefore have a great toxic potential.

No metabolic functions in the human body are known for which mercury is required. The neurological symptoms and liver and kidney damage caused by acute mercury poisoning are well known. The relief of Hg from amalgam tooth fillings and the possible connection between increased Hg concentration in hair and excess risk of coronary heart diseases have raised a question about the health effects of longterm exposure to sufficiently low mercury and methylmercury concentrations. It has been supposed that mercury can promote lipid peroxidation through a free radical mechanism. The ability of mercury to form very stable compounds with S and Se can lower the protective effect of selenium.

Hair, serum, red cell, urine and faeces samples are used for monitoring Hg status in a human. Cold-vapour techniques with atomic absorption or atomic fluorescence detection with suitable digestion procedures are mostly used for the total mercury analyses in human

samples. Neutron activation is suitable for almost all media. X-ray methods are used especially for environmental samples. For the speciation of organic Hg compounds, selective organic extractions with different chromatographic methods are the most extensively used.

Determination of folates in foods — challenges and advances of the use of HPLC. Liisa Vahteristo,* Velimatti Ollilainen, Pekka Koivistoinen & Pertti Varo.

Department of Applied Chemistry and Microbiology, FIN-00014 University of Helsinki, Finland.

There is increased need for specific data on folates and their vitamers distribution in foods. Many procedures for folate determination do not meet these needs, but are chiefly meant for measuring total folates or added folic acid. Use of HPLC enables separation and quantitation of the most abundant folate vitamers in their monoglutamate forms (Gregory, 1984).

In this study reduced folate vitamers were determined in some foods, e.g. liver and liver products, after heat extraction and deconjugation of polyglutamate forms of folate moiety into folate monoglutamates using hog kidney conjugase. These folate forms were separated using reversed-phase high performance liquid chromatography and quantitated using fluorescence detection. The methods used provided data on the amounts and the distribution of the folate vitamers present. The amounts determined in e.g. pork and beef liver were much higher than expected from literature. Folate content of some other foods were much more in accordance with previous findings.

The data on folate content of foods should be carefully evaluated for its validity, and in many cases new analysis should be performed before such data can be used, for example for the estimation of the dietary intake of folate. Additional data on folate forms present in food, determined using HPLC, provide more in-depth information of the chemical nature of this vitamin. But careful validation of the method and specific identification procedures are essential for HPLC methods intended for folate analysis.

1. Gregory III, J. F. (1984) *J. Assoc. Off. Anal. Chem.*, 67(5), 1015–19.

*To whom correspondence should be addressed.

New components included in future food composition tables. Gary. R. Beecher.

Food Composition Laboratory, Beltsville Human Nutrition Research Center, ARS/USDA, Beltsville, MD 20705, USA.

Nutrients and food components for which data are essential are dependent on the food supply and health status of the population of each country or region. Several regions of the world have diseases resulting from inadequate intakes of several micronutrients including iodine, iron and vitamin A active components. Several

debilitating diseases, in 'well-nourished' regions of the world have strong dietary associations. With regard to cancer and cardiovascular disease (CVD), many nutrients and food components with antioxidant activity are being investigated as preventive agents. These components include carotenoids, flavonoids, phytate, tocopherols, selenium and vitamin C. Several food component, coumestrol, isoflavonoids and lignans, either have anti-estrogenic activity or are converted to compounds with this activity in the gastro-intestinal tract and are proposed to reduce the risk of hormone related cancers. The risk of CVD also is altered by dietary levels of total fat, individual fatty acids, trans fatty acids and cholesterol as well as dietary fiber and its fractions. Low folic acid intake has been implicated in the risk of several diseases. Dietary levels of calcium, boron and vitamins D and K are important determinants for osteoporosis. The intake of sodium is one of the primary risk factors to stroke. Example of priority setting and justification for the addition of nutrients to databases will be discussed.

The selenium content of human milk and infant formulae in Finland. Päivi Ekholm,* Maija Ylinen & Pertti Varo.

University of Helsinki, Department of Applied Chemistry and Microbiology, Post Box 27, Viikki-D, FIN-00014 University of Helsinki, Finland.

The aim of this study was to monitor the effects of selenium (Se) fertilization (started in 1985) on the Se content of human milk and infant formulae.

The samples of human milk were received from the Helsinki University Central Hospital. Each sample was a pool of milks from several donors (median 20). The infant formulae were sampled from food stores in the Helsinki area. Se was determined from freeze-dried samples by the electrothermal atomic absorption method.

The Se content of human milk was 0.08 mg/kg DM in 1994, more than twice as high as in the mid 1970s. The present level in Finland is nearly equal to that prevalent in many other countries. The mean Se content of infant formulae was 0.06 mg/kg DM in 1994, which is three times higher than in the mid 1970s. The Se content of infant formulae has remained lower than that of human milk.

The present Se content of human milk is an indication of the improved Se status of the Finnish population. The Se fertilization has increased the Se content of all Finnish foods effectively, including the infant formulae.

*To whom correspondence should be addressed.

Trends in the cadmium contents of bovine and porcine liver between 1982–1995. Merja Euroola* & Pertti Varo.

University of Helsinki, Department of Applied Chemistry and Microbiology, P.O. Box 27, Viikki, General Chemistry, SF-00014 University of Helsinki, Finland.

The aim of the study was to monitor retrospectively the development of the cadmium (Cd) contents of bovine and porcine livers from the beginning of 1980.