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BIOMARKERS: THEORETICAL ASPECTS AND APPLICATIVE PECULIARITIES NOTE II. NUTRITIONAL BIOMARKERS

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Abstract

By analytical investigation of tissues and body fluids as well as of metabolites it is possible to evaluate certain substances that are considered "bioindicators". With a generic term these substances have been denominated "biomarkers" (biological markers). One category of biomarkers that is gaining an increased interest are the nutritional biomarkers. A nutritional biomarker can be any biological specimen that is an indicator of nutritional status with respect to intake or metabolism of dietary constituents. It can be a biochemical, functional or clinical index of status of an essential nutrient or other dietary constituent. Nutritional biomarkers are used for a variety of purposes in large-scale population surveys and epidemiologic studies as well as smaller clinical studies. The main reasons for using nutritional biomarkers are to provide measures of nutritional status that have less error than dietary data, nutrient status for nutrients with inadequate dietary data, to obtain a more proximal and integrated assessment of nutrient status that incorporates metabolism, to assess dietary change and compliance in intervention studies, and dietary intake for the validation of dietary questionnaires.
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1. Concept of Nutritional Biomarker

B*iological markers,* according to Hulk (1990), can be defined as "cellular, biochemical or molecular alterations that are measurable in biological media such as human tissues, cells or fluids". The term "*biomarker*" is used in a broad sense to include almost any measurement reflecting an interaction between a biological system and a chemical, physical or biological environmental.

A biomarker is a biologic specimen that may be a marker of exposure to some substance, of its metabolism, or of the integration of exposure and metabolism. Biomarkers may also reflect host characteristics.

One area of biomarker's research which gains increased attention is that of nutritional biomarkers. Traditionally, self-report measures of dietary intake have served as primary nutritional indicators, but biomarkers have several potential advantages. One is that they can capture dietary intake more objectively than can self-report measures.

Another advantage is that nutritional biomarkers can indicate nutritional status, or the integration of intake and metabolism. However, they must be used judiciously, as there are several limitations as well. Nutritional biomarkers do not always correlate well with dietary intake assessments, are not yet available for some dietary constituents, and variability in laboratory methods can affect comparisons across studies.

2. Typological Characterization of Nutritional Biomarkers

Generally, biomarkers can be included in three classes having in view the exposure, susceptibility, and effect. Biomarkers of exposure - exogenous substances, their metabolites or the product of interaction between a xenobiotic agent and the target molecule or cell that is measured in a compartment within an organism. Biomarkers of susceptibility - indicators of an inherent or acquired ability of an organism to respond to the challenge of exposure to a specific xenobiotic substance. Biomarkers of effect - measurable biochemical, physiological, behavioral or other alterations within an organism that, depending upon the magnitude, can be recognized as associated with an established or possible health impairment or disease (see Note I).

A *nutritional biomarke*r can be any biological specimen that is an indicator of nutritional status with respect to intake or metabolism of dietary constituents. It can be a biochemical, functional or clinical index of status of an essential nutrient or other dietary constituent.

Nutritional biomarkers may be interpreted more broadly as a biologic consequence of dietary intake or dietary patterns.

2.1. Classification of biomarkers

Biomarkers have been classified according to Perera (2000) based on the sequence of events from exposure to disease. The use of biomarkers improves the sensitivity and specificity of the measurement of exposure to risk factors.

Regarding the nutritional biomarkers they can be designated in three different categories:

Biomarkers of dietary validation – means of validation of dietary instruments.

Biomarkers of nutritional intake – surrogate indicators of dietary intake.

Biomarkers of nutritional status – integrated measures of nutritional status for a nutrient.

For a biomarker to be used for validation of a dietary instrument, it should have a strong direct relationship with dietary intakes and be an independent assessment of the dietary intake of the nutrient of interest. An important example of such a biomarker is the use of urinary nitrogen as a marker of dietary protein (Bingham, 2003).

A second category of biomarker would be indicators of dietary intake for situations where the direct measurement of dietary intake using traditional methods is difficult or impossible. Nutrients and food components can vary considerably for the same food depending on where or how the food was grown or how it was processed. In these cases, a biomarker may be a better indicator of dietary intake. In some cases too, complementary use of a biomarker and dietary assessment provides a better or different estimate of the nutritional exposure (Potischman, 2003).

The third category of biomarkers are those that are integrated measures of nutritional status for a nutrient, which reflect not only intake but also metabolism of the nutrient and possibly effects from disease processes. It is important to note that a single biomarker may not reflect the status of a single nutrient, but may be reflective of several nutrients, their interactions and metabolism.

2.2. Functional characteristics

Approaching the problem of functional characteristics of biomarkers, interesting aspects are revealed. These aspects will be successively discussed.

Biomarkers of nutritional exposure reflect the distribution of the nutrient, or in some cases of the xenobiotic, or its metabolite throughout the organism. Theoretically, this distribution can be tracked through various biological levels (e.g., tissue, cell, etc.) to the final target. A part will be distributed to internal macromolecules, and a smaller amount will reach the critical site on the macromolecule, with only a fraction of the latter amount acting as the biologically effective dose (Margetts, 1987).

Finding and defining the appropriate exposure of interest is not so easy. Although dietary databases exist or are being developed for the known food components that are of interest to health, it is unclear whether biomarkers or dietary estimates are more informative for some of these. In addition, it is often unclear if the important exposure from a food source is the parent compound, a metabolite or some unknown covarying constituent in the food source. Any factor that is under physiologic control at intakes beyond minimum requirements is in the third category. For example, serum calcium and vitamin A are under homeostatic control; therefore, for healthy individuals, they do not provide indications of dietary intake.

In the situation where currently available biologic markers provide less information than estimates of dietary intake, the use of biomarkers may not be possible. Some phytoestrogens may fall into this category (Lampe, 2003).

The problem of exposure evaluation by biomarkers can present interest also for investigations regarding the action of chemical xenobiotics of food interest of organic origin (polycyclic aromatic hydrocarbons, mycotoxins, steroid hormones etc.) and of inorganic origin (nitrates, nitrites, potentially toxic elements).

Knowledge of the kinetics of formation and removal from the body of these types of biomarkers provides a link between exposure and internal dose. Specific measures of internal dose are the active chemical species (either parent compound or metabolite) delivered to target tissues or cells, the reactive chemical species delivered to target organelles or macromolecules, or the reactive chemical species that participates in biochemical reactions.

3. Uses and limitations of nutritional biomarkers

The nutritional biomarkers can be used as: 1) means of validation of dietary instruments; 2) surrogate indicators of dietary intake; 3) integrated measures of nutritional status for a nutrient. Many biomarkers can fall into more than one of these categories, and for some food components, the biomarkers are not adequate and dietary intake methods may provide better information.

There are three main reasons for using nutritional markers. First, biochemical markers of nutrient status can have less error than dietary assessment of nutrient status (Kaaks, 1997). In addition to issues related to measurement errors in dietary assessment (i.e. errors in completion of the instrument and food-composition tables), there are additional issues such as the combination of foods eaten together and the extent of cooking of foods that influence nutrient content and absorption. These issues add error to the estimation of nutritional status that is obtained from dietary instruments.

Second, for some nutrients, dietary data are inadequate because of limitations in food-composition data, whereas biomarkers of nutritional status related to these nutrients are available. For example vitamin E who is particularly difficult to quantify from food-composition data (Beecher, 1990). The main source of vitamin E is fats and oils, and the content of vitamin E varies depending on processing procedures, type of oil, shelf life and addition of antioxidants to restore oxidized vitamin E. None of these issues can be addressed with dietary assessment instruments.

Third, biomarkers provide a more proximal measure of nutrient status than dietary intake data for disease outcomes and population nutritional status measures. The biomarker serves as an integrated measure of metabolism of the nutrient of interest.

The nutritional biomarker can be used as a measure of internal dose, which is an indication of the amount of nutrient available to the tissues after absorption and metabolism. The marker can also be used as a measure of dietary change (Le Marchand, 1994).

Biomarkers provide accurate measures that can be correlated to dietary intake and can have less error than dietary intake estimates. The errors associated with biologic variables are independent of those associated with questionnaire data, which are important for statistical analyses involving measurement-error correction (Kaaks, 1997). Combining nutrient estimates from questionnaire data with serologic measures of the same nutrient can provide a powerful tool for estimating the exposure of interest and assessing risk (Potischman, 1991).

Like every tool of analysis, biomarkers have their limitations. The limitations of the selected markers must be known for them to be used for validation purposes. For some purposes, dietary data can be more appropriate than biomarker data.

For some nutrients, there are a variety of reasons why the relation may not be simple and why other factors need to be considered when one is trying to relate biomarkers to dietary intake. Most of the problems are raised by factors regarding physiology and absorption. There is variation between individuals in physiology and nutrient metabolism. To a large extent, much of the interindividual variability is not well understood. Nonetheless it must be appreciated that these differences between individuals exist.

It is very easy in the case of biomarkers to believe that a single entity is having an effect because the easily measured, well-characterized constituent may have less error associated with it than with its dietary counterparts. Sometimes the effect may truly be related to that one dietary constituent [e.g., lycopene and prostate cancer (Campbell, 1994)], but more likely it is representative of a food or a food group. For example, genistein may be a good indicator of soy intake, but it may not be the sole player in any effect observed in an epidemiologic study. Clinical intervention studies provide important roles for differentiating single constituents from foods themselves as being influential in a disease process.

The discovery of new biomarkers of nutritional intake and status, and the critical evaluation of them and the known biomarkers are essential to the continuing progress in nutritional epidemiology.

Conclusions

The use of biomarkers for the evaluation of the intake and nutritional status is a rapidly changing field, due especially to the fact that the nutritional biomarkers can provide measures of nutritional status that have less error than dietary data. The nutritional biomarkers can be used as: means of validation of dietary instruments; surrogate indicators of dietary intake; integrated measures of nutritional status for a nutrient.

Finding and evaluating new nutritional biomarkers is crucial for the continuing progress of science, but in order to do that we must understand that biological markers also have their limitations. Knowing the limitations of the selected biomarkers is crucial in establishing if in a given case is better to use a biomarker evaluation method or a dietary data based method of investigations. Also we must take in consideration that in some cases combining nutrient estimates from questionnaire data with serologic measures of the same nutrient can provide a powerful tool for estimating the exposure of interest and assessing risk.

Remarks. This paper entitled "Biomarkers: Theoretical Aspects and Applicative Peculiarities", presented in two parts: Note I and Note II were elaborated as a result of our collaboration in the framework of the UE Project "Biotechnology Thematic Network" (initiated in 1999 by the Letter of Agreement nr. 4750 / 3012.1999) and coordinated by University of Perugia – Dept. of Biochemistry and Molecular Biotechnology (Prof. Dr. M.V. Magni). Also, in the framework of this Project the book «Molecular Biology » 5th edition (2005) was published.

References

Beecher, G.R., Matthews, R.H. (1990). *Nutrient composition of foods*. In: Present Knowledge in Nutrition, 6th ed. (Brown, M. L., ed.), pp. 430–443. International Life Sciences Institute, Washington, D.C.

Bingham, S.A. (2003). Urine nitrogen as a biomarker for the validation of dietary protein intake. *J. Nutr*. 133, 921S–924S.

Campbell, D.R., Gross, M.D., Martini, M.C., Grandits, G.A., Slavin, J.L., Potter, J.D. (1994). Plasma carotenoids as biomarkers of vegetable and fruit intake. *Cancer Epidemiol. Biomarkers Prev*. 3, 493–500.

Gârban, Z. (2004). *Xenobiotice chimice de interes alimentar,* Editura Eurobit Timişoara

Gârban, Z. (2005). *Biologie moleculară*, ediția 5-a, Editura Eurobit Timişoara

Gârban, Z. et al. (2005). Biomarkers: Theoretical aspects and applicative peculiarities. *Scientifical Researches. Agroalimentary Processes and Technologies,* XI(1), 139-146.

Hulka, B.S., Griffith, J.D., Wilcosky, T.C. (eds.). (1990). *Biological markers in epidemiology*, New York: Oxford University Press.

Lampe, J.W. (2003). Isoflavonoid and lignan phytoestrogens as dietary biomarkers. *J. Nutr.* 133, 956S–964S.

Le Marchand, L., Hankin, J.H., Carter, F.S., Essling, C., Luffey, D., Franke, A.A., Wilkens, L.R., Cooney, R.V., Kolonel, L.N. (1994). A pilot study on the use of plasma carotenoids and ascorbic acid as markers of compliance to a high fruit and vegetable dietary intervention. *Cancer Epidemiol. Biomarkers Prev*. 3, 245–251

Kaaks, R., Riboli, E., Sinha, R. (1997). *Biochemical markers of dietary intake*. In: Application of Biomarkers in Cancer Epidemiology (Toniolo, P., Boffetta, P, Shuker, D. E. G., Rothman, N., Hulka, B. & Pearce, N., eds.), pp. 103–126. IARC Scientific Publications No. 142, International Agency for Research on Cancer, Lyons, France.

Margetts, B.M., Nelson, M. (eds.). (1987). *Design concepts in nutritional epidemiology*, New York, Oxford University Press

Mayeux, R. (2004). Biomarkers: Potential use and limitations, *J. Am. Soc. Experim. Neuro Therap*., 1 (2), 182-188.

Perera, F.P., Weinstein I.B. (2000). Molecular epidemiology: recent advances and future directions, *Carcinogenesis*, 21, 517-524.

Potischman, N., Herrero, R., Brinton, L.A., Reeves, W.C., Stacewicz-Sapuntzakis, M., Jones, C.J., Brenes, M.M., Tenorio, F., de Britton, R.C., Gaitan, E. (1991). A case-control study of nutrient status and invasive cervical cancer. II. Serologic indicators. *Am. J. Epidemiol.* 34, 1347–1355.

Potischman, N. (2003). Biologic and methodologic issues for nutritional biomarkers. *J. Nutr*. 133, 875S–880S.