1. Introduction

Soterios Kyrtopoulos
National Hellenic Research Foundation, Athens, Greece

Biomarkers constitute a potentially powerful tool for the study of environmental carcinogenesis. In particular, the use of biomarkers of carcinogen exposure and early effects can facilitate the search for the environmental etiology of cancer by helping to break down the gap between exposure to environmental carcinogens and clinical disease into a series of intermediate stages which can be recognised and quantified through corresponding measurable endpoints. This is particularly important in view of the fact that contact with environmental carcinogens normally involves prolonged, low-level exposure to multiple carcinogenic agents and that a long latent period, often lasting decades, intervenes between exposure and the appearance of clinical cancer. Thus, by providing objective measures of exposure to specific agents and early, biologically relevant effects, *at the level of the individual*, biomarkers can provide a way of investigating associations between exposure and disease.

During the past few decades, great efforts have been invested in the experimental identification of biomarkers of carcinogen exposure and early effects, and the development of analytical methods for their detection and quantification. As a result of these efforts, assays of exquisite sensitivity have been developed, enabling, for example, the measurement of the concentrations of metabolites, or adducts with macromolecules, of many environmentally relevant carcinogens at very low levels of exposure (e.g. polycyclic aromatic hydrocarbons, aromatic amines, benzene, 1,3-butadiene, aflatoxin B1, nitrosamines, etc.), or the detection and quantitation of early genetic effects at the level of the chromosomes or specific genes [1].

Such biomarkers provide quantitative information about the amounts of specific chemicals entering the human body or reaching critical cellular targets, or about effects on biological targets which may be on the causal pathway to cancer and therefore may serve as predictors of disease risk. However, for their potential to be realised, it is essential that biomarkers undergo the critical process of validation. Biomarker validation encompasses two different levels: one relates to the analytical and operational methodology (assay validity, reliability, intra- and inter-laboratory variability, influence of sampling and storage, etc.). The other, equally important, aspect of validation aims to evaluate the inherent ability of biomarkers to reflect what they are meant to reflect: ideally the chemical nature, level and duration of an individual's exposure (for biomarkers of exposure) and the degree of disease risk (for biomarkers of early effect). The current Report focuses on this second aspect of validation.

The studies necessary for the validation of biomarkers of exposure initially involve animal experiments to illuminate the dose–response and exposure–time relationships

governing biomarker levels and to provide information on their relative values in surrogate tissues employed in human studies (usually blood or urine) and the target tissues of highest interest in terms of disease risk. In the case of biomarkers of early biological effects, their ability to act as independent predictors of disease risk can be established in animal studies by examining their association with disease under the influence of different modulating factors.

While the availability of adequate experimental animal data of this kind constitutes an important step in the process of biomarker validation, full validation ideally requires additional, analogous observations in humans. Obviously this is much more difficult to achieve, since it requires that exposure to the agents of interest occurs at a range of levels which can be accurately estimated by independent means. Even if this is achievable, such data can normally provide information only about the degree to which different levels of exposure are reflected in the levels of a biomarker measured in a surrogate tissue, e.g. blood cells, while information about the relationship of the latter with the exposure of critical target tissues is much more difficult to obtain for humans. Additional information, important for validation, which can be obtained only via human studies, concerns the background levels of biomarkers, their origin and inter-individual variation.

Despite the substantial progress which has been achieved in the development of analytical methodologies, few biomarkers can be said to have completed the process described above so that they can be considered as adequately validated and mature for use in risk assessment. The present Report summarises the state of validation of the most important biomarkers of food-related carcinogen exposure and early effects and identifies the specific gaps which still remain. In this way it can serve as an important tool for the planning of future validation studies that may eventually enable the realisation of the full potential of biomarkers in the context of epidemiological studies to investigate the environmental etiology of cancer.

References

1. Farmer PB, Emeny JM, editors. Biomarkers of carcinogen exposure and early effects. Łódź: ECNIS Publications, Nofer Institute of Occupational Medicine; 2006.