METHODS IN MOLECULAR BIOLOGY

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Molecular Toxicology Protocols

Second Edition

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Preface

Molecular Toxicology Protocols, Second Edition addresses a scientific field primed to explode upon the clinical and popular horizons. Toxicology, a subdiscipline of pharmacology, is actually the interface of chemistry and biology. This field also extends into nonchemical "agents" with deleterious biological effects, especially radiation, the purview of the radiobiologist and health physicist. With the huge increase in computational power now available over the last two decades, it has become possible to model and predict the potential toxicity of yet untested, and even unmade, chemicals. Perhaps, the greatest change in the recent practice of toxicology has been in applying the "tools of the trade" directly to the human population, in what are known "translational" studies, entering the realm of epidemiology. These studies expand the traditional public health aspect of toxicology from simple screening of agents for toxicological potential prior to their introduction into the environment to now include attempts to define "normal" or "background" exposures, elucidating the mechanistic basis of human disease and designing methods for preclinical intervention ("chemoprevention").

Thus, for our purposes, we define "molecular" toxicology as either any study of toxicological mechanism, or any translation or application of such studies into the human population. Today, such "molecular" toxicology is mostly genetic toxicology, where the genetic material, DNA, is the target molecule. Of course DNA is found throughout the human body, such that all of the traditional modulators of toxicological effect, such as uptake, distribution, and metabolism, must be taken into account. Although genetic damage can have many outcomes, the one most clearly linking exposure and disease has been cancer.

During the past several years, important progress has been made in the understanding of the molecular biology of the cell, the cellular responses to genotoxic agents, and the molecular biology of human cancer. This progress has been rapidly achieved thanks to the development of new state-of-the-art techniques and continuous improvement of existing methods. Such advances permit not only the study changes of in cellular morphology but also the detection of changes occurring in the cellular genetic material (DNA), the cellular transcript (RNA), and the translated product (proteins). These molecular methods have now offered many potential areas of clinical applications. Therefore, following a successful publication of the first edition of *Molecular Toxicology Protocols* in 2005, this second volume contains several new chapters. Subjects of these new chapters range from preparation of fluid specimens for analysis of cellular inflammatory responses to genotoxic insults to sensitive methods for proteomic analysis and aberrant DNA methylation patterns.

Several books are currently available on the applications of molecular methods to various types of biotechnology. To our knowledge, however, there is no book emphasizing the application of molecular methods to genetic toxicology.

Therefore, the aim of *Molecular Toxicology Protocols* is to bring together a series of articles, each describing validated methods to elucidate specific molecular aspects of toxicology. With such content, this book addresses the needs of not only molecular biologists and toxicologists, but also all individuals interested in applying molecular methods to

clinical applications, including geneticists, pathologists, biochemists, and epidemiologists. The volume is divided into ten parts, roughly corresponding to the spectrum of biomarkers intermediate between exposure and disease outcomes as proposed in molecular epidemiology models.

Thus, Part I contains chapters describing methods to analyze global changes in protein expression and identify low-abundance proteins in cells and clinical samples, while the chapters in Part II describe methods for detecting cellular secretions in response to toxicant-induced inflammation. Part III describes methods for the analysis of an essential epigenetic modification, DNA methylation, which modulates gene expression and is frequently altered in toxicant-treated cells and clinical samples. Part IV addresses the application of the new array technologies to genetic toxicology, including methods for the analysis of individual variations in biotransformation and the effects of genetic exposure on gene expression. Part V includes chapters describing the sensitive and specific detection of pro-mutagenic lesions in the genetic material, while Part VI includes chapters assessing gross or macroscopic genetic damage. Parts VII and VIII focus on the detection and characterization of viable mutations in surrogate markers and cancer-related genes, respectively. The chapters of Part IX describe methods for the analyses of various pathways of DNA repair, an important modulator of genotoxicology. Finally, Part X describes methods for the analysis of cytotoxicity caused by the induction of apoptosis since cell death can either protect the organism from a transforming cell or cause distinct health effects itself.

As time goes by we believe that "molecular" approaches will play an increasingly important role in all types of toxicology, not just genetic toxicology. Moreover, genetic damage and dysfunction will undoubtedly be found to play a role in many more diseases of aging than just cancer and is probably a fundamental mechanism of aging itself. Therefore, the focus of this second edition, genetic toxicology, and more specifically, the genetic toxicology of cancer, represents just the "tip of the iceberg" as far as the field of molecular toxicology will eventually be understood.

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