Research in DNA damage detection is a fast-paced field of study. The continuous requirement in this field for ever faster DNA damage assays is driven by the rapidly increasing numbers of samples to be analyzed and by the intensified competition between various scientific groups.

The faster assays make possible rapid high-volume and machine-based assessments. Such capability is in demand in drug design, high-throughput industry, environmental studies, and in molecular and clinical pathology research. It enables a pathology laboratory to perform rapid evaluations of DNA damage in either fixed archived tissues or unfixed clinical samples. The ubiquitous nature of DNA damage and its outcomes to a cell, such as apoptosis or necrosis, makes such assays applicable to studies across a wide range of organs and tissues. As time- and cost-saving techniques, the faster assays are useful for basic research in academia and in clinical studies of pathologies where DNA damage and apoptotic cell death play an essential role: cancer, ischemic disorders, and degenerative diseases.

To satisfy this requirement many new approaches are being developed. In addition quicker variations of the established techniques are constantly introduced. These methods use various tactics to cut down the detection time including the homogenous “closed-tube” formats with FRET reporting, new and faster labeling enzymes, quicker DNA probes, advances in the design of analytical instruments, and new ways to probe the samples, such as ultrasound scans.

Although such rapid techniques are in demand in the “research trenches,” they are not covered well in the literature. This volume is the first such compendium of the time-saving techniques for detection of DNA damage and its direct physiological outcomes including apoptosis, necrosis, and phagocytic clearance.

The term “fast detection” in the book title is an umbrella designation indicating three types of time-saving assays. The assays which take around 1 h to perform comprise a group of true express techniques. These are followed by the rapid assays which take 3 h or less to complete. Next are the accelerated techniques. Although being lengthier than the previous two groups of methods, these represent significantly shortened, speeded up versions of routinely used techniques.

In line with this the book is divided into three parts. Part one—Express Detection—includes the fastest protocols which require less than 60 min to complete. The strategies which these express methods use to minimize the detection time include near instantaneous FRET reporting, agarose trapped cells, ultrafast labeling enzymes, as well as advantageous instrumental approaches, such as flow cytometry, and ultrasound. Most protocols in this section require 30–40 min, although several such as FRET or ultrasound assays need only 3–5 min.

Part two of the volume—Rapid Detection—describes the techniques which can be finished in 3 h or less. This part presents the isothermal “zebra tail” and RT PCR amplification assays to label DNA breaks, in addition to flow and image cytometry, and immuno-cytochemical detection of γ-H2AX and DNA damage. This section also contains the original Fast Micromethod procedure for genotoxicity assessments in cell suspensions and homogenized tissues.
The third part describes the accelerated detection of DNA damage and apoptosis. It contains time-saving modifications of the popular techniques. These speeded up assays mostly take 4–6 h to complete. The presented techniques include a high-throughput version of the comet assay for quicker examination of DNA damage and repair, immunofluorescence analysis of γ-H2AX foci, RAPD-PCR for the evaluation of genotoxin-induced DNA damage, and the fast-tracked ex vivo detection of γ-H2Av foci in Drosophila imaginal discs.

Overall the book presents a comprehensive collection of quick assays for the detection of nuclear and mitochondrial DNA damage and its effects in live and fixed cells and tissues, and in bacterial genomes. In addition to mammalian cells the protocols describe the use of cells from invertebrate species, such as sea mussels and Drosophila, which are convenient models for the environmental tests and toxicity studies. The volume demonstrates all levels of detection, starting from the molecular level up to the level of the entire live organism.

Such a broad methodological resource will be equally useful to both beginners in the DNA damage field and experienced researchers. Its particular usefulness is for those who perform day-to-day investigations of DNA damage and its effects, such as postdoctoral fellows, research group leaders, and academic faculty.

Researchers in molecular and cell biology, biochemistry, oncology, radiobiology, experimental and clinical pathology, toxicology, embryology, experimental pharmacology, drug design, and environmental sciences will find the book beneficial.

This book describes easily reproducible techniques requiring only a few steps to perform, and therefore, it will be useful for those who would like to gain quick access to the technical arsenal needed to study DNA damage. As such it will provide help to senior students and clinical scientists who would like to familiarize themselves with research possibilities in DNA damage detection and to explore this unique field.

I thank Candace Minchew for her highly useful and expert technical assistance. I am also grateful to Professor John Walker for his advice and encouragement throughout the review process.

Houston, TX, USA

Vladimir V. Didenko
Fast Detection of DNA Damage
Methods and Protocols
Didenko, V.V. (Ed.)
2017, X, 216 p. 42 illus., 24 illus. in color., Hardcover
A product of Humana Press