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DNA Repair: Front and Center and Not Going Away!

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Abstract

This introduction to the book: DNA repair protocols: third edition, edited by Bjergbaek, discusses the history and more recent developments in the field of DNA repair. This research field started in the 1950 and developed from a small group of researchers interested in the damage caused to DNA by ultraviolet irradiation from the sun to become a large field of research today. DNA damage and its repair are now thought to play an important role in the etiologies of cancer, aging, and neurodegeneration and there is a great deal of interest in this venture. Thus, understanding of DNA processing is now a central field in molecular and cellular biology, and the field is still growing.

Keywords

DNA damage; DNA repair; Aging; Cancer; Neurodegeneration

1. Historical Perspective

DNA repair first emerged as a specialized area of genetic research in the mid 1950s, when scientists began investigating the ability of cells to recover from exposure to ultraviolet (UV) light. It took approximately 10 years of intense study to analyze and characterize this process, now known to involve at least two subpathways: photoreactivation and nucleotide excision repair. Detailed understanding of several additional prototypical DNA repair pathways followed relatively quickly. Since the mid-1970s, research on DNA repair has steadily gained more and more visibility, rapidly becoming recognized as a major research area. In the “early days,” only a handful of people worldwide were even interested in DNA repair: in contrast, the public health and clinical relevance of DNA repair are now widely recognized, many researchers focus on understanding one or two DNA repair pathways or subpathways, multiple international DNA repair conferences are held annually, and DNA repair research is funded by many major funding agencies, including those focused uniquely on cancer biology.

2. DNA Repair Pathways

DNA is continuously exposed to endogenous and exogenous agents that generate lesions or structural perturbations into DNA. Endogenous DNA damage is primarily caused by reactive oxygen species, formed as a by-product of mitochondrial oxidative phosphorylation. Exogenous DNA damage is caused by a large number of compounds and agents, including naturally occurring and manmade chemicals, as well as ambient and induced forms of radiation. It has been estimated that endogenous and exogenous sources combined generate approximately 100,000 lesions in the genome of a single human cell in one 24 h day (1).

All prokaryotic and eukaryotic cells express multiple DNA repair pathways, which collectively provide slightly redundant capacity to repair all types of DNA damage. However, the efficiency of repair of each DNA lesion is a function of many variables, including the nature of the lesion, the context/site of the lesion in the genome, as well as the type of cell and the specific cellular genotype, to mention a few.

Oxidative DNA damage and DNA strand breaks are repaired by *base excision repair (BER)* and *single-strand break repair (SSBR)*. Bulky DNA lesions are repaired by *nucleotide excision repair (NER)*, which includes both *global genome (GG-NER)* and *transcription-coupled (TC-NER)* subpathways. DNA double-strand breaks and interstrand cross-links are repaired by *DNA double-strand break repair (DSBR)*, a pathway under intense investigation, with many novel pathway components and subpathways being explored and more likely to be discovered. *DNA mismatch repair (MMR)* recognizes and repairs insertion/deletion loops, hairpins, and base–base mismatches introduced throughout the genome during semiconservative DNA replication and in heteroduplex DNA segments generated during homologous recombination and other DNA repair reactions. UV photoproducts can also be repaired by *direct reversal*, for example, by a *DNA photolyase*.

While all of these pathways are being studied as separate entities, it is also becoming more and more evident that there are significant overlaps and that some proteins participate in more than one pathway. It is therefore necessary to keep a rather open mind about the relative pathway entities and their integrities.

As mentioned above, the cellular capacity to repair a specific DNA lesion at a specific site under specific conditions varies significantly. Initial studies of the site or region specificity of DNA repair revealed that certain genome regions are repaired more efficiently than others. Another observation was that active genes are preferentially repaired, while relatively quiescent transcriptionally inactive regions of the genome are repaired more slowly. This phenomenon, most commonly referring to repair of UV-induced photolesions, is now known as *transcription-coupled repair*, or *TC-NER*. At this time, there is no convincing evidence for transcription-coupled repair of oxidative DNA lesions, base–base mismatches, or double-strand breaks in actively transcribed regions of the genome. *TC-NER* was first characterized using assays that detect repair of specific DNA lesions in specific genes and similar assays have more recently been used to detect preferential repair in specific gene promoter regions. In this book there are chapters on gene-specific repair and regional repair; some are based on quantitative southern analysis and others on PCR approaches. As these approaches are quite technically advanced, they require specific equipment and special skills.

3. Repair of the Mitochondrial Genome

While studies of the repair and replication of the nuclear genome advanced rapidly during the years of the “molecular biology” explosion (approximately 1960–1990), the repair and replication of the mitochondrial genome is now of increasing interest to many geneticists as well to as molecular and cell biologists. In fact, a strong consensus is emerging that mitochondria and the mitochondrial genome play essential roles in many aspects of cellular biology, including nuclear genomic stability and integrity. Early studies suggested that

aging-related DNA damage accumulates more rapidly in the mitochondrial than in the nuclear genome and that the mitochondrial genome is repaired very slowly, if at all. While these ideas have been strongly disputed in recent years, the technical challenges associated with studying mitochondria were at least in part responsible for slow progress in understanding mitochondrial genomics. That said, it is now clear that oxidative DNA lesions in mtDNA are repaired efficiently, and that both short and long patch BER subpathways are robustly expressed in mitochondria (Table 1). Mitochondria are also proficient in MMR, but appear to lack capacity to repair bulky DNA lesions by NER (or an NER-like process). Interestingly, many nuclear DNA repair proteins have been detected in mitochondria, despite early reports to the contrary, and despite accurate reports that these enzymes lack canonical mitochondrial targeting sequences.

4. DNA Repair Defects: The Clinical Consequences

In 1968, James E. Cleaver published the first direct evidence that the skin cancer susceptibility disorder *xeroderma pigmentosum* is an inherited disease syndrome, in which skin pathology might be causally linked to mutations that inactivate or reduce the efficiency of NER (2, 3). Consistent with this, reduced exposure to UV light dramatically reduces skin cancer incidence in patients with *xeroderma pigmentosum*. This, as it turns out, was the “tip of the iceberg,” and at least partly because of Paul Modrich’s discovery that defects in human MMR genes are linked to inherited susceptibility to human colon cancer (HNPCC), the DNA repair enzymes got “front page news” status as *Science* magazine’s *Molecule of the Year* in 1994 (4–8). Since 1994, many additional links between DNA repair defects and human disease have been discovered. Moreover, DNA repair defects have not only been linked to cancer susceptibility, but also appear to play a significant role in human neurodegenerative diseases as well as normal and premature human aging.

5. DNA Repair Research: The Devil Is in the Details (i.e., The Assay)

The idea and purpose behind this book is to gather and present in one volume technical information on DNA repair assays, with the hope that the book will facilitate research progress. In their day-to-day activities, researchers who study DNA repair must choose between a very large number of well-established DNA repair assays, novel less well-characterized assays, and the option of embarking themselves on new assay development. These are important and difficult choices, and there is usually no “correct” answer, or single good approach ... in fact, the best approach is to ask the same question using two or more different methods or approaches, each of which is carefully selected based on the research goal (i.e., what is the biologically important question being asked) and the strengths and limitations of each method. Because these questions are so critical and difficult, the goal of this volume is to make it a bit easier to answer them and develop a well-designed experimental approach to address any given question about DNA repair.

6. The Future of DNA Repair Research: Critical Goals

Although DNA repair defects cause several human diseases including skin and colon cancer, with minor exceptions, DNA repair assays cannot yet be used for routine clinical diagnosis

or to evaluate disease progression in patients receiving chemotherapy. This is because most available DNA repair assays are either not sufficiently sensitive, or are too cumbersome (i.e., low throughput) or too expensive for clinical use. Thus, an important focus for the future is to develop DNA repair assays that can be used effectively in the clinic.

A second important future research goal is to develop highly specific inhibitors of individual DNA repair enzymes, pathways, or subpathways. Once available, it is widely believed that it will be possible to develop combination therapies using one or more DNA repair inhibitors and one or more known cancer drugs, dramatically enhancing therapeutic efficacy and potentially preventing cancer progression or even inducing cancer regression.

Clearly, well-characterized assays for specific DNA repair enzymes and pathways will be critical tools for achieving these research goals. It is hoped that this volume will be useful to researchers working on these critical research areas or asking other interesting questions about DNA repair.

References

1. Lindahl T. Instability and decay of the primary structure of DNA. *Nature*. 1993; 362:709–715. [PubMed: 8469282]
2. Cleaver JE. Defective repair replication of DNA in xeroderma pigmentosum. *Nature*. 1968; 218:652–656. [PubMed: 5655953]
3. Setlow RB, Regan JD, German J, Carrier WL. Evidence that xeroderma pigmentosum cells do not perform the first step in the repair of ultraviolet damage to their DNA. *Proc Natl Acad Sci U S A*. 1969; 64:1035–1041. [PubMed: 5264135]
4. Koshland DE. Molecule of the year: the DNA repair enzyme. *Science*. 1994; 266:1925. [PubMed: 7801114]
5. Culotta E, Koshland DE Jr. DNA repair makes its way to the top. *Science*. 1994; 266:1926–1929. [PubMed: 7801115]
6. Modrich P. Mismatch repair, genetic stability, and cancer. *Science*. 1994; 266:1959–1960. [PubMed: 7801122]
7. Parsons R, Myeroff LL, Liu B, Willson JK, Markowitz SD, Kinzler KW, Vogelstein B, et al. Microsatellite instability and mutations of the transforming growth factor beta type II receptor gene in colorectal cancer. *Cancer Res*. 1995; 55:5548–5550. [PubMed: 7585632]
8. Marx J. DNA repair comes into its own. *Science*. 1994; 266:728–730. [PubMed: 7973626]

Table 1

Nuclear and mitochondrial DNA repair pathways

Pathways	Subpathways	Nuclear	Mito
Base excision	Long patch	Y	Y
	Short patch	Y	Y
Nucleotide excision	General genome	Y	N
	Gene	Y	N
	Transcription associated	Y	N
Mismatch		Y	Y
Recombination	HR	Y	?
	NHEJ	Y	?