TIMELINE

DNA repair, genome stability and cancer: a historical perspective

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Abstract | The multistep process of cancer progresses over many years. The prevention of mutations by DNA repair pathways led to an early appreciation of a role for repair in cancer avoidance. However, the broader role of the DNA damage response (DDR) emerged more slowly. In this Timeline article, we reflect on how our understanding of the steps leading to cancer developed, focusing on the role of the DDR. We also consider how our current knowledge can be exploited for cancer therapy.

Carcinogenesis has long been recognized as a state of uncontrolled growth of our own cells. The earliest models proposed the notion of a mutational event, even before James Watson and Francis Crick's seminal discovery of the structure of DNA. By the 1980s, the initiation step of carcinogenesis was understood to necessitate the generation of mutations, with the concept of environmental mutagens and failed DNA repair being central to many models. By contrast, an understanding of the basis underlying tumour progression or outgrowth unfolded relatively slowly, and an appreciation of the critical role of the DNA damage response (DDR) took even longer to solidify (see BOX 1 for useful definitions). Indeed, even as the twenty-first century began, DNA repair remained a relatively minor component of the broad field of cancer research.

It is now appreciated that tumour progression necessitates the downregulation of damage surveillance mechanisms and an increase in genetic and epigenetic instability to achieve uncontrolled proliferation and the adaptability associated with aggressive tumours. In this Timeline article, we describe early concepts of mutation and cancer that predate knowledge of the structure of DNA, and we summarize how the links between mutagenesis and carcinogenesis were established. We go on to discuss the early studies of viral oncogenes and the insights that they provided into carcinogenesis, leading to the much more recent but critical understanding that oncogene-induced replicative stress promotes genomic instability. We provide a perspective on how the notions of tumour initiation and progression emerged, describing the key concept that tumour progression is inexorably linked to disruption of the DDR (FIG. 1). Finally, we consider the ironic conundrum that, although targeting the DDR can provide treatment strategies, it is the misregulation of the DDR that is often the route by which tumour cells evade therapy.

Mutagenesis underlies carcinogenesis

In the early twentieth century, long before the structure of DNA was defined, Theodor Boveri proposed that a cancer cell was a changed normal cell and advanced the theory that tiny microscopic bodies called chromosomes were abnormally distributed in cancer[1]. As the notion of hereditary units or 'determinants', and later 'genes', emerged alongside their relationship to chromosomes, the idea that permanent changes to these 'genes' (defined as mutations) could underlie heritable biological phenotypes became conceptualized. From there, it did not require a great leap to appreciate that such mutations might underlie the origin of the biological variation observed in cancer (reviewed in REF. 2). In the 1930s, it was recognized that, whereas a normal human cell has 46 chromosomes, cancer cells have chromosomes that are abnormally distributed and frequently present in excess of 46 (REF. 1). Paradoxically, non-tumorous cells and plants with an asymmetrical or unbalanced chromosome distribution grew less vigorously than normal cells, whereas cancer cells were characterized by an enhanced growth capacity[1]. Work in *Drosophila melanogaster* had revealed that chromosome aberrations correlated with the formation of genetic variants and, in the late 1920s, Hermann Muller demonstrated that exposure of *D. melanogaster* to X-rays induced "transmutation" of a gene, causing both visible chromosome aberrations and heritable phenotypic variations[3,4].

Intriguingly, in 1775, Percival Pott made the seminal observation that there was a high incidence of scrotal cancer in boys who assisted chimney sweeps, and he linked this finding to exposure to soot[5–7]. This represented the first evidence for a work-related and environmental cause of cancer. By 1955, shortly after the discovery of the structure of DNA, it was well appreciated that exposure to chemical mutagens could increase DNA mutation rates, with a correlation between mutagenesis and carcinogenesis being hypothesized although certainly not consolidated[2]. Remarkably, the suggestion that there could be cancer susceptibility genes was also proposed[2].

Linking mutagens, carcinogens and DNA

With the definition of the structure of DNA in 1953 (REFS 8,9) and the understanding that genetic mutation represented a change in the chemical structure of the DNA molecule, the first clear connections between the processes of mutation and carcinogenesis were made by Phil Lawley, who worked at the Chester Beatty Research Institute (United Kingdom; now known as the Institute of Cancer Research). Working with Peter Brooks, he showed that many classic alkylating agents worked by reacting directly with DNA to form stable chemical adducts that could disrupt the template function of the DNA molecule[10,11]. This led directly to their seminal observation that the carcinogenicity of polycyclic aromatic hydrocarbons — the likely causative agents of scrotal cancer in Pott's chimney sweeps and

also the classic carcinogenic components of tobacco smoke — was directly correlated with their ability to form DNA adducts, providing an unambiguous link between the initiation of cancer and chemical changes in DNA[12]. The importance of these early findings and the functional link between mutagenesis and carcinogenesis are demonstrated by the development and later adoption of tests classifying carcinogens on the basis of this relationship[13].

Lawley and Brooks were also among the first to document biological repair processes for chemical and environmental damage to DNA[10], a theme very actively adopted by many laboratories[14–19] (see REF. 20 for a review). Research over the next 30 years progressively revealed a plethora of repair pathways for chemical lesions of DNA, primarily in bacteria. Subsequently, human homologues for many of these repair enzymes and pathways were identified. For the most part, these systems — or possible defects in them — were not associated with the initiation or the progression of cancer in any significant way. There was, however, an emerging recognition of the role that DNA repair mechanisms might have in mediating resistance to alkylating agents used for cancer chemotherapy[21].

Insights from DNA repair disorders

A notable exception to this picture was a seminal observation made in 1969 by Jim Cleaver[22], who was studying the rare autosomal-recessive cancer predisposition syndrome xeroderma pigmentosum (XP), which affects 1 in every 250,000 individuals. Patients with XP have a 1,000-fold increased chance of developing skin cancer but display almost normal levels of tumour presentation in other tissues[23]. Cleaver found that cells from patients with XP were defective in the ability to repair DNA damage caused by ultraviolet (UV) light[22]. The DNA repair defects in most (but not all) XP cells were subsequently shown to result from mutations in components of the human nucleotide excision repair system[22,23]. Crucially, this process is responsible for the removal of helix-distorting UV-induced photodimers from DNA, which explains the highly specific skin cancer predisposition of patients with XP.

A second clear example of a defective DNA repair pathway being responsible for cancer initiation emerged in the early 1990s: patients with Lynch syndrome (also known as hereditary non-polyposis colorectal cancer) — a familial pattern of colorectal cancer characterized by microsatellite repeat instability — were found to carry mutations in the human homologues of the bacterial mismatch repair proteins MutS and MutL[24–27]. Both of these inherited diseases reinforced a model of cancer initiation in which random unrepaired point mutations eventually result in an alteration of the coding sequence of a key oncogene or tumour suppressor, thus initiating the first step in cell proliferation and enabling a subsequent cascade of mutagenic events in these precancerous cells.

A prediction that emerged from these studies of patients with hereditary predisposition to cancer was that mutations in DNA repair genes might frequently arise in cancer cells. As we discuss below, this has certainly proved to be the case. However, the early studies were carried out when there was not a comprehensive understanding of DNA repair pathways or the DDR of humans and when sequencing technology was considerably less sophisticated than it is now. Thus, the attempts to address this prediction were not very revealing. In those early studies, polymorphisms and tumour-associated mutations in DNA base excision repair enzymes — for example, 8-oxoguanine DNA glycosylase (OGG1) and apurinic–apyrimidinic endonuclease 1 (APE1; also known as APEX1) — and in components of the downstream XRCC1-based part of base excision repair were identified in some tumour cells[28,29]. However, the penetrance of such polymorphisms is weak, and the clinical relevance of these to the overall cancer burden in the population was unclear[30]. Subsequently, several complex conditions in which cancer predisposition is a feature — such as Bloom syndrome, Werner syndrome and Fanconi anaemia — have been shown to arise from genetic defects in DNA repair systems, as have subsets of familial breast, ovarian, prostate and pancreatic cancers[31–36].

Studies of radiation exposure

That X-ray exposure confers an increased risk of malignant disease, including leukaemia and skin cancers, became

Box 1 | **DNA damage response processes of relevance for cancer**

DNA repair
There are multiple DNA repair pathways, with subpathways providing lesion specificity. Nucleotide excision repair removes bulky DNA lesions; DNA non-homologous end joining and homologous recombination repair DNA double-strand breaks (DSBs); mismatch repair corrects mismatched base pairs; and base excision repair repairs damaged bases and links to single-strand break (SSB) repair. Mutations in these pathways increase cancer susceptibility.

DNA damage response signalling
There are two DNA damage response (DDR) signalling pathways: ataxia telangiectasia mutated (ATM)-dependent signalling is activated by DSBs; and ataxia telangiectasia and RAD3-related (ATR)-dependent signalling is activated by single-stranded regions of DNA. DDR signalling can activate apoptosis and checkpoint arrest, and can influence DNA repair. Mutations in ATM signalling components confer cancer susceptibility. However, ATR-deficient mice show reduced capacity for tumour formation[93].

Cell cycle checkpoints
DNA integrity is constantly monitored, with DNA damage triggering a checkpoint response that prevents cell cycle progression; arrest can be permanent or transient. Checkpoints prevent progression from G1 to S phase and from G2 to M phase, and an intra-S phase checkpoint regulates fork progression or origin firing. Many tumours have inactivated checkpoint responses.

Apoptosis
Apoptosis represents a programmed cell death pathway that functions in some tissues during normal development but also prevents proliferation of damaged cells. Apoptosis can be p53 dependent or independent. p53 is commonly mutated in cancer.

Fidelity of replication
Multiple processes function to maintain the accuracy of replication and enhance recovery from replication fork stalling or collapse. Homologous recombination has a key role, and genes involved in this process are commonly mutated in cancers.

DNA re-replication
Re-replication can cause aneuploidy and subsequently genomic instability. Several mechanisms prevent DNA re-replication. For example, increased cyclin-dependent kinase (CDK) activity is required for origin firing but is inhibitory for origin licensing.

Telomere length
Shortened telomeres lead to senescence, and so cancer cells need to maintain telomere length to survive. Activation of telomerase or an alternative pathway to maintain telomere length is common in cancers.

accepted within a few decades after the discovery of X-rays in 1895. However, radiation studies were disappointing when it came to gaining mechanistic insight into the aetiology of cancer. Nonetheless, reports by the International Commission on Radiological Protection (ICRP) and the United Nations Scientific Committee on the Effects of Atomic Radiation (UNSCEAR) provide an invaluable resource for rationalizing the emerging concepts (for example, see REF. 37). In particular, the analysis of atomic bomb survivors provided a wealth of epidemiological data and revealed, for example, that there can be a long induction period before the onset of cancer, and in the UNSCEAR 1958 report it was concluded that radiation-induced mutations are biologically relevant for carcinogenesis[37]. However, the relationship between chromosome aberrations or rearrangements (which X-rays avidly induce) and point mutational changes (which X-rays inefficiently induce) remained puzzling. A further important concept emerged from these early radiation studies: it was demonstrated that the frequency of cell transformation — which was used as a surrogate for carcinogenicity — did not correlate with the cell-lethal effects when cells were exposed to different types of radiation[37,38] (note that ionizing radiation of different types produces DNA damage of different complexities). Thus, the carcinogenic impact of radiation cannot simply be attributed to its capacity to cause cell death.

These early studies that evaluated the response to radiation damage raised an additional dilemma: although patients with ataxia telangiectasia are cancer prone and profoundly radiosensitive, DNA was not hypermutated in patient cells that were exposed to X-rays and, despite their marked radiosensitivity, cells from these patients did not display an obvious defect in the repair of X-ray-induced DNA damage[39,40]. The cause of this apparent paradox only became clear after the discovery that the gene defective in patients with ataxia telangiectasia, *ATM*, encodes a protein kinase that triggers a signalling cascade that regulates cell cycle arrest and cell death responses rather than a DNA repair enzyme[41] (nonetheless, ATM signalling can indirectly influence DNA repair processes). This important distinction between signalling responses and direct DNA repair has proved crucial in the context of cancer avoidance. Indeed, the wider response to DNA damage (known as the DDR) is now usefully subdivided into damage response signalling and direct DNA repair[42] (BOX 1). Notably, DDR signalling frequently has a greater impact on genomic stability in response to DNA damage compared with DNA repair pathways, which more dramatically influence cell survival.

> “What emerged more slowly, however, was an appreciation that DDR mechanisms … are essential for cancer avoidance”

Competing models and research fields

The concept that cancer involves at least one genetic mutation was well accepted by the 1970s. However, the notion that oncogenesis is a multistage process was proposed by Isaac Berenblum and Phillipe Shubik as early as 1948, based on studies showing that tumour cells induced by carcinogen treatment could remain in a latent stage until outgrowth was promoted by subsequent treatment with croton oil, which contains phorbol diester and activates growth signals[43]. From 1970 to the turn of the twenty-first century, a range of studies — including epidemiological analysis of atomic bomb survivors, studies in mice and cell culture models of transformation — provided strong evidence that cancer is a multistage process[44]. It was understood that cancer incidence increased dramatically with age and that exposure to ionizing radiation brought forward the age of onset of most cancers, but that it still involved a marked lag period. Two contrasting models arose to explain these observations: one model stated that cancer involved a mutagenic initiation step, followed by a long period of outgrowth[45], and the other stated that cancer was a multistage process, involving multiple mutational hits[46]. Slowly, the concept of a multistage process became the predominant model, supported in part by the observation that the transformation of cultured cells occurred more rapidly, and at a higher frequency, if cells were transfected with two oncogenes rather than one[47] (see, for example, a review written in 1993 (REF. 48)).

The marked number of mutational and chromosome changes present in cancer cells, which were evident from the early studies, and the fact that the median number of rearranged chromosome arms correlated with cancer prognosis[49] made an important contribution to the shaping of models and thoughts. The multistep nature of carcinogenesis coupled with the evident chromosome changes led to several models that, at their core, involved clonal evolution: that is, the progressive selection of rare mutated cells[50]. Two extreme models were proposed. In the first, carcinogenesis required the activation of multiple oncogenes and/or the inactivation of tumour suppressor genes and each rearrangement contained an amplification or loss of a specific gene. This was supported by the identification of p53 as a tumour suppressor, the loss of which enabled the evolution of rare mutated cells[51]. Such hypotheses also suggested that elevated genome instability per se (a mutator phenotype; see below) would not necessarily be required, as each acquired mutation could theoretically increase growth potential independently. The alternative extreme model proposed that a mutator phenotype characterized by an intrinsically high level of mutations occurred in the founding clone. This suggested that the vast majority of rearrangements had no phenotypic consequence but rather represented ‘passenger mutations’. Although controversial at the time, the mutator phenotype model remains actively discussed as part of working models today[52–54]. Advances in single-cell sequencing procedures are demonstrating the enormous sequence changes between cells within a single tumour and have shown that the level of plasticity within a tumour correlates with aggressiveness[55]. However, these findings do not entirely allow the distinction to be drawn as to whether multiple mutations and a mutator phenotype cause malignancy or are instead a consequence of it. It will thus be crucial to deduce the stage at which genomic instability arises.

In parallel with the emerging concepts that carcinogenesis necessitated DNA sequence changes, thinking also focused on the fact that cancer is predominantly a disorder of deregulated growth that is likely to involve changed patterns of differentiation or dedifferentiation. By this time, it was generally accepted that differentiation during development was epigenetic[56]. This led to experiments in which tumour cell nuclei with a normal modal chromosome number were transplanted into anucleated eggs to generate adult animals[57–59]. Notably, such injections allowed for the development of normal animals, potentially demonstrating a developmental totipotency that suggested a non-mutational basis for transformation to malignancy. In the

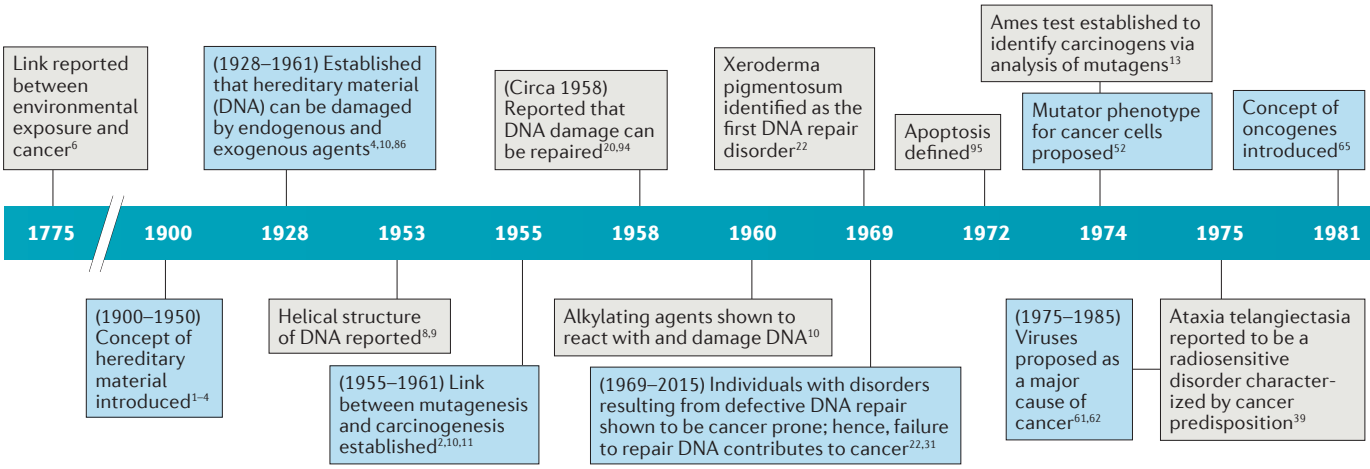


Figure 1 | **Timeline showing the key concepts and findings relating to the role of the DNA damage response in the development of cancer.** Key concepts are shown in blue and key findings in grey. DDR, DNA damage response; MMR, mismatch repair; *MSH2*, MutS homologue 2; ROS, reactive oxygen species.

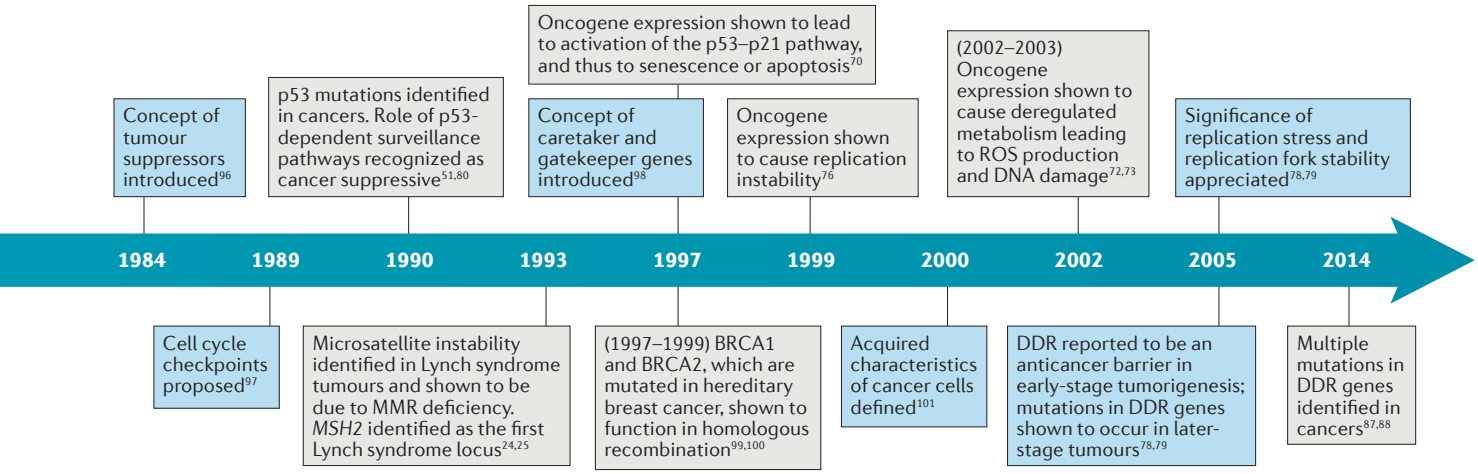
context of current knowledge, such experiments were likely to be flawed, or at least exceptional. However, developments in the DNA methylation field provoked research into methylation changes associated with cancer, leading to proposals that methylation changes drive expression changes and thus cancer development[60]. Indeed, we now know that epigenetic changes are commonly found in cancer cells and, like mutagenesis, provide a route to changed gene expression and thus function.

During the latter part of the twentieth century, the various models tended to be considered as exclusive, generating unfortunate conflicts that also influenced the battle that emerged regarding a viral aetiology for cancer. The identification of numerous oncogenes from work on viruses, along with revelations that many viruses encode proteins related to human growth factors and that expression of these could promote deregulated growth, led to a widespread belief that the majority of cancers were of viral origin[61,62]. The viral community, in part because of its huge contributions to oncogene discovery (see below), gained considerable influence. In hindsight, the strength of the arguments made by the viral community discouraged full consideration of a genomic instability model for cancer development. With our current knowledge, a model that proposed genomic instability as the origin of cancers would predict that viral infections could be carcinogenic, given the ability of many viruses to subvert components of the DDR (for example, see REFS 63,64). Any remaining controversy concerns the magnitude of the viral aetiological contribution, which cannot easily explain, for example, diet- and smoking-related tumorigenesis.

Oncogenes and oncogene-induced stress

Oncogenes were first identified by studying retroviruses: the prototype oncogene, *src*, is a chicken Rous sarcoma virus gene that was hijacked from the chicken genome[65]. It soon became clear that a defined, but significant, number of oncogenes were involved in cancer initiation and that oncogene expression caused neoplastic transformation[66]. In the early 1990s, it was reported that genome instability occurred rapidly after *HRAS* oncogene expression, and subsequent reports demonstrated that this was not an isolated phenomenon, as it was observed after expression of other oncogenes[67–69]. By the late 1990s, it had been shown that the product of an alternative reading frame (ARF) within the cyclin-dependent kinase inhibitor 2A (*CDKN2A*) locus, $p14^{ARF}$, which binds to MDM2 and upregulates p53, responded to RAS and MYC expression by activating the p53–p21 pathway to drive senescence or apoptosis[70,71] (BOX 1). As it was known that DNA damage activated the p53–p21 pathway to drive senescence or apoptosis, this led to the suggestion that oncogene expression directly caused DNA damage[72].

It was initially proposed that deregulated metabolism due to MYC-dependent transcription increased the production of reactive oxygen species (ROS) and thus DNA damage, an idea consistent with models postulating that a mutator phenotype underpinned cancer development[72]. Although the interpretation of these findings at the time was controversial, the emerging consensus was that it was replication stress that, either directly or indirectly, caused the observed DNA damage. The link between oncogene expression and DNA damage and/or its repair generated considerable interest: for example, MYC expression in non-cancerous cells was shown to reduce DNA repair efficiency and induce p53 and its ATM–ATR-dependent (but $p14^{ARF}$-independent) phosphorylation[73,74]. Concurrently, it was shown that p53 and p21 prevented cell proliferation if cyclin E or cyclin-dependent kinase 2 (CDK2) was overexpressed and that this operated through an ATM–ATR-dependent and $p14^{ARF}$-independent mechanism[75]. Cyclin E expression had been previously shown to cause chromosome instability[76], and it was later demonstrated to interfere with replication[77]. The scene was thus set: oncogene-induced proliferation of otherwise normal cells perturbed DNA replication mechanisms, producing measurable DNA damage and genome rearrangements and activating the p53–p21 pathway via ATM–ATR-dependent mechanisms. In 2005, two key papers[78,79] clearly demonstrated both the activation of DDR signalling, including proteins required for cell cycle checkpoint arrest (BOX 1), and increased expression of DNA damage markers in precancerous tissue. These papers proposed that this reflected oncogene-induced DNA damage arising



from replication stress, synthesizing cell culture data and demonstrating a direct relevance to clinically derived cancer tissue.

DDR downregulation in tumours

As discussed above, the notions that endogenous and exogenous DNA damage cause mutations that lead to carcinogenesis, and that cancer avoidance necessitates active DNA repair mechanisms, emerged as key concepts early in work on the aetiology of cancer. What emerged more slowly, however, was an appreciation that DDR mechanisms in general, as distinct from specific repair pathways per se, are essential for cancer avoidance. Initially, based on studies of apoptosis, an important concept for our understanding of cancer onset emerged: it was not necessarily the DNA damage itself that killed (or prevented the growth of) a cell, but rather the signalling pathways activated by such damage. Cell cycle checkpoint pathways, initially defined as systems that monitor genome integrity, are now understood to be pivotal in precluding the continued proliferation of damaged cells[80]. p53 has a key role in this, and the frequent loss of p53 function in tumours contributed to the emerging notion that DDR pathways must be downregulated to allow uncontrolled proliferation[81].

In 1997, Manuel Serrano *et al.*[70] proposed that expression of the *HRAS* oncogene leads to activation of the p53–p21 pathway, which drives senescence or apoptosis. Thus, for proliferation to occur, the p53–p21 pathway must become downregulated in *HRAS*-expressing cells[70]. Similar findings were observed following expression of the *MYC* oncogene, although in this case proliferation necessitated downregulation of the p14^{ARF}–MDM2–p53 pathway[82]. Slightly distinct but related examples also followed, such as the demonstration that, although tumours in *Brca2*$^{+/-}$ mice undergo loss of heterozygosity, the proliferation of homozygous BRCA2-deficient tumour cells demands additional mutations in spindle checkpoint genes[83]. The full breadth of the relevant pathways that require downregulation, and subsequently their importance in contributing to tumour progression, was slowly revealed, as was the realization that downregulation of these pathways could create a mutator phenotype that causes genomic instability, as postulated many years earlier.

Lessons from history

Current models of cancer would argue that an initiating event (or several initiating events), often caused by a mutation, leads to oncogene activation and ensuing replication stress[84]. However, this must be coupled with subsequent downregulation of DDR mechanisms — possibly by genetic alterations as a consequence of replication stress — to allow continued proliferation and continued genome instability, a prerequisite for a cancer cell to rapidly adapt to its ever-changing microenvironment. Although the historical reflection in this article has considered the timeline of emerging seminal concepts, this does not reflect the order of events in the aetiology of cancer (FIG. 1). The initiating event (or events) causing oncogene activation most likely precedes a state of replication stress, but it remains unclear whether downregulation of the DDR is always directly caused by errors that arise from replication stress or if it could precede replication stress. Findings from the laboratories of Jiri Bartek[78] and Thanos Halazonetis[79], that upregulation of the DDR occurs in precancerous lesions and that p53 mutations occur subsequently in late-stage tumours, strongly support an order of events in which the onset of replication stress represents an early event, promoting the subsequent mutations that allow outgrowth.

However, a more recent study involving ultradeep sequencing of cancer genes in sun-exposed normal skin biopsies revealed a substantial accumulation of cancer driver mutations (with the characteristic signature of UV-induced mutations) that had undergone positive selection in the absence of evidence for cancer formation[85]. This suggests a different aetiology in which there is a strong initial selection pressure to upregulate growth-enhancing genes; cells with such changes then await further steps that lead to a genetically unstable state. The enhanced cancer predisposition caused by mutations in genes that affect both the early (for example, mutations enhancing an initiation event such as in XP) and perceived late (for example, damage surveillance mechanisms such as in Li–Fraumeni syndrome) steps of cancer would be consistent with there not being a defined order of events that lead to carcinogenesis.

Our historical reflection in this article considers the many models or factors that have been proposed to contribute to carcinogenesis, which include viruses, epigenetic changes, DNA-damaging agents, replication stressors and oxidative stress. On the basis of our current knowledge, all of these factors are indeed valid contributors, and they all merge into a model that ultimately leads to the generation of a genetically unstable state, which is in most cases essential for carcinogenesis (FIG. 2). Strikingly, this pinpoints the enormous significance of the DDR processes: their

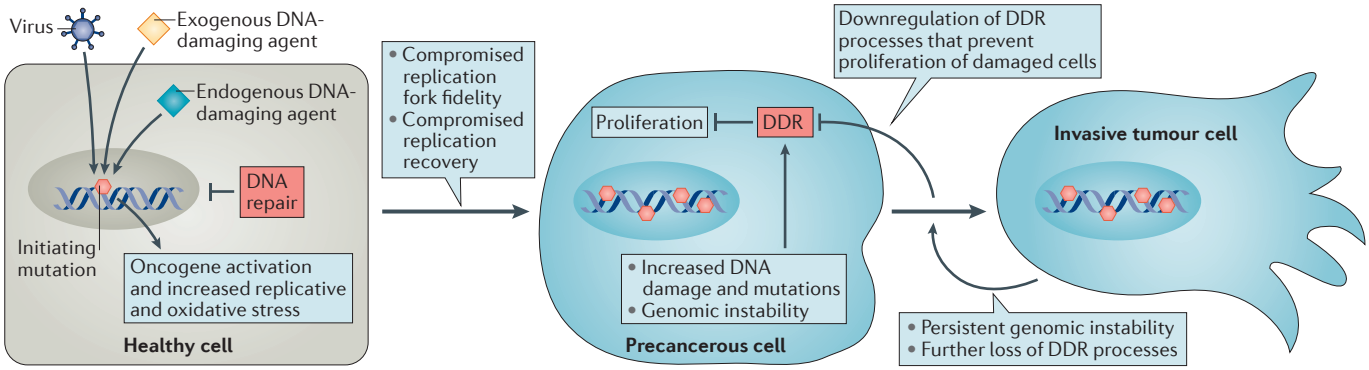


Figure 2 | **How the DNA damage response pathways influence steps leading to cancer.** The figure shows how changes in the DNA damage response (DDR) pathways promote critical steps in the aetiology of carcinogenesis. A healthy cell has a plethora of DDR processes to protect its DNA from exogenous and endogenously arising DNA-damaging agents, and respond to viral infections. Nonetheless, the processes are not perfect, and an early step in the aetiology of cancer is the generation of one or more mutational changes. This may directly or indirectly result in oncogene activation, which leads to replicative and/or oxidative stress. Genetic predisposition to cancer can arise when one of these DNA repair processes is compromised. However, although enhanced replication stress increases the level of DNA breakage, mutation or rearrangement, a range of responses — for example, the ability to accurately recover replication, the activation of checkpoint arrest or other p53-dependent responses — can prevent the proliferation of damaged cells. Progression from this precancerous state to ongoing proliferation requires the downregulation of these DDR processes, thereby facilitating persistent genomic instability. For clarity, these steps have been depicted to arise in a linear fashion, which may not be the case.

importance was evident in the earliest studies but has emerged to be far more substantial than originally predicted. Although early studies demonstrated that cells can recover from exposure to external DNA-damaging agents, revealing that they harbour DNA repair mechanisms[20], perceptive scientists also saw that the DNA structure revealed by Watson and Crick could accumulate endogenously arising DNA damage, predicting that DNA repair pathways might be essential even during normal growth and metabolism[86]. However, these early studies did not predict that the DDR mechanisms encompass not only DNA repair processes that seek to avoid the initiator mutations, but also DNA damage surveillance mechanisms that preclude the proliferation of genetically unstable cells. Furthermore, there is also a requirement for an efficient replication machinery that restricts replication errors and for processes that allow recovery from the inevitable difficulties encountered during replication in a manner that maintains genomic stability (BOX 1). We now know that cancer cells not only downregulate these pathways but often subvert them to fast-track evolution and gain adaptability, which is the ultimate driver of carcinogenesis and metastasis[87].

The future

This Timeline article highlights the significance of the role of DDR processes in cancer aetiology. However, the plethora of DNA integrity surveillance, repair and signalling pathways, alongside their profound interconnectedness, has only been appreciated more recently. Similarly, the advent of tumour genome profiling by deep sequencing has only recently demonstrated that DDR genes are frequently mutated in all cancer types, with many individual pathways or genes found to be mutated in more than 50% of cancers of a specific type (for example, more than 50% of ovarian cancers harbour mutations in genes involved in homologous recombination)[87,88] (FIG. 3).

The steps during carcinogenesis can be summarized as shown in FIG. 2. Although an understanding of these steps is of significant academic interest, it also provides a key tool for informed, targeted cancer therapy[87]. The enhanced sensitivity of many cancer cells to DNA-damaging agents, including radiation exposure, has been evident for many years and indeed has been exploited for therapeutic benefit. The rationale for such sensitivity was poorly understood and was often unsatisfactorily attributed to the more rapid growth of tumour cells. Consequently, the approach relied on serendipity, coupled with random trial and error. Our current knowledge of how the DDR pathways are changed in cancers is providing routes for more specific and rationally targeted therapy.

A significant concept in this regard is that of synthetic lethality, in which the goal is to identify a drug that will cause lethality to a cancer cell that has inherent DDR defects without harming a normal cell[87]. The foremost and very elegant example of exploiting a synthetically lethal genetic relationship is the treatment of breast cancers arising in BRCA1- or BRCA2-deficient patients[89]. The key insight came from the realization that BRCA1 and BRCA2 function during homologous recombination, a key process that promotes replication fork stability, and that drug-targeted inhibition of a specific enzyme (poly(ADP-ribose) polymerase 1 (PARP1)) confers a reliance on homologous recombination and hence drug sensitivity[89]. Although such an approach might be anticipated to uniquely benefit BRCA1- or BRCA2-deficient patients, current studies revealing that homologous recombination can be downregulated in approximately 50% of ovarian cancers dramatically expand the scope for such therapy[88]. A plethora of related studies are in progress, which include strategies to promote synthetic lethality based on changes such as the subversion of apoptosis, altered pathways of DNA double-strand break repair and loss of checkpoint arrest in cancer cells[87,89]. Conversely, a similar but distinct phenomenon called 'synthetic viability' can allow the rescue of a cell-growth defect that is imposed either by a preceding mutation in the cancer cell or by drug treatment (or by a combination of both). It is well established that cancer cells often gain mutational changes that confer synthetic viability in the context of drug therapy, including in response to PARP inhibitors[90]. It is likely that many cancer cells have equivalent synthetic viability mutations (for example, p53 mutations, as discussed above) that compensate for preceding genetic changes[91,92].

In conclusion, to fully exploit the genome instability that is now recognized as an inherent property of most — if not all — cancers, it is critical that we enhance our understanding of the DDR pathways and also exploit imaginative ideas to progress cancer treatment. Ironically, however, our understanding of the stages of carcinogenesis has also provided an explanation for why our targeted therapies will frequently fail.

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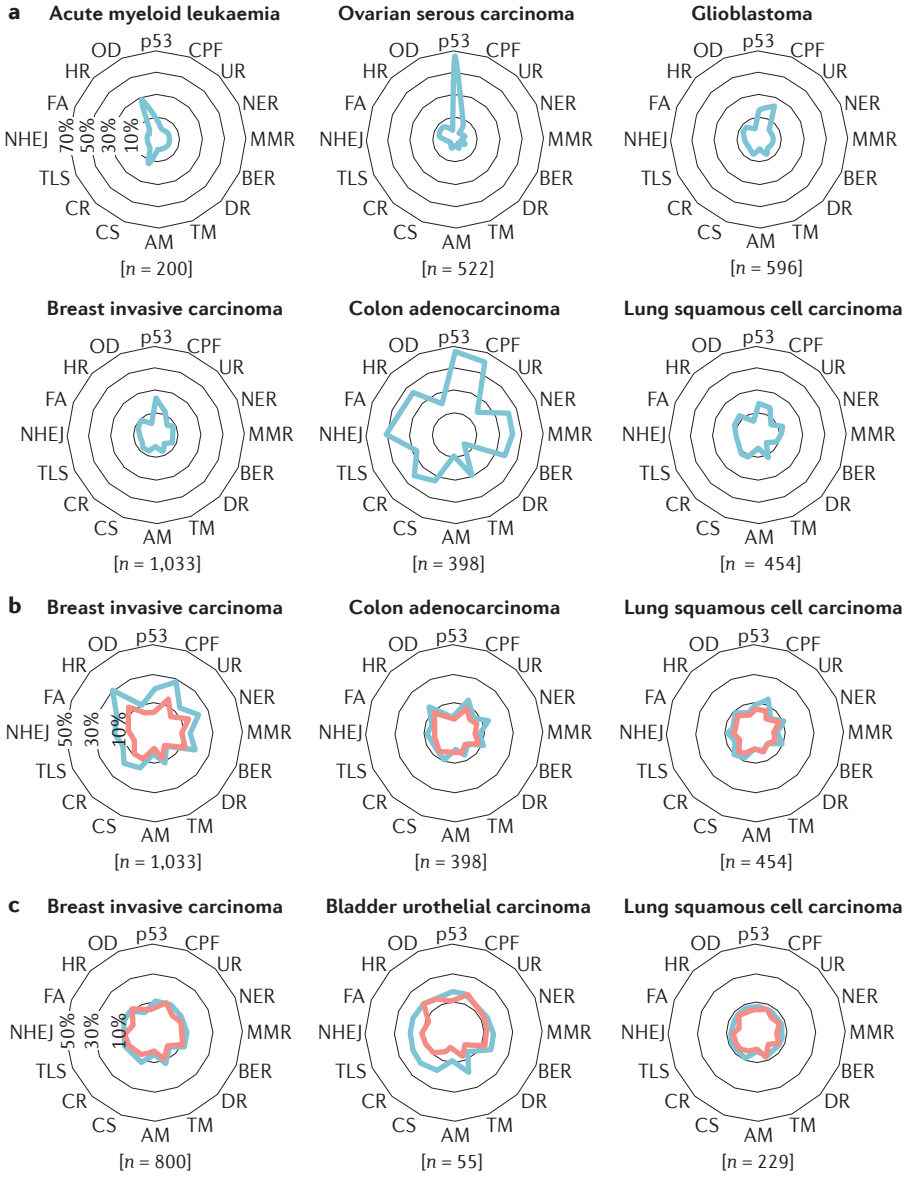

Figure 3 | **Many proteins involved in the DNA damage response are mutated in cancer.** Data from The Cancer Genome Atlas have revealed the extent to which mutations in DNA damage response (DDR) proteins are observed in cancers. These changes differ for different tumour sites. The concentric circles indicate the percentages of patients affected. Part **a** shows radial plots in which the radius length indicates the proportion of patients with protein-coding mutations that are predicted to disrupt particular DDR pathways in each cancer type. All mutations included are present in at least two distinct patient samples. Part **b** shows copy number variation in the different DDR pathways (red indicates loss of gene copies; blue indicates amplification of genes). Part **c** shows expression level variation in DDR pathways (red indicates decreased expression; blue indicates increased expression). AM, alternative mechanism for telomere maintenance; BER, base excision repair; CPF, checkpoint factor; CR, chromatin remodelling; CS, chromosome segregation; DR, direct repair; FA, Fanconi anaemia pathway; HR, homologous recombination; MMR, mismatch repair; NER, nucleotide excision repair; NHEJ, non-homologous end joining; OD, other double-strand break repair; TLS, translesion synthesis; TM, telomere maintenance; UR, ubiquitylation response. Figure reproduced from REF. 87, Nature Publishing Group.

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Competing interests statement

The authors declare no competing interests.

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