The Role of Chromosome Deletions in Human Cancers

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Abstract

Chromosome deletions are a hallmark of human cancers. These chromosome abnormalities have been observed for over than a century and frequently associated with poor prognosis. However, their functions and potential underlying mechanisms remain elusive until recently. Recent technique breakthroughs, including cancer genomics, high throughput library screening and genome editing, opened a new era in the mechanistic studying of chromosome deletions in human cancer. In this chapter, we will focus on the latest studies on the functions of chromosome deletions in human cancers, especially hematopoietic malignancies and try to persuade the readers that these chromosome alterations could play significant roles in the genesis and drug responses of human cancers.

Keywords

Chromosome deletion · Human cancer · Knudson's two-hit hypothesis · Haploinsufficient tumor suppressor · Genome editing

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9.1 Introduction

Copy number variation (CNV) is one of the hallmarks of human cancers [1]. Deletions and amplifications of focal chromosome regions, chromosome arms or even whole chromosomes are frequent in both blood and solid cancers. Back to the end of the eighteenth century, German pathologist David Hansemann first observed asymmetric distribution of "chromatin loops" even though it was difficult to clearly see the chromosomes under the microscopes at his age [2, 3]. Following this seminal observation, another German pathologist Theodor Boveri proposed that "a particular, incorrect chromosome combination which is the cause of the abnormal growth characteristics passed on to daughter cells" [4].

Hansemann and Boveri's initial observations were further confirmed in the following more than 100 years. After the first chromosome abnormality in cancer, the Philadelphia chromosome, was discovered in 1960, sophisticated cytogenetics technologies have been developed to study the karyotyping of cancer, especially leukemia [5]. Given that CNVs are frequent and associated with poor prognosis, it is crucial to understand the functions of these chromosome alterations in tumorigenesis and drug response. It is generally believed that chromosome deletion regions contain tumor suppressors while chromosome amplification regions contain oncogenes [6, 7].

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However, so far, majority of chromosome deletions don't contain obvious confirmed tumor suppressors. It has been argued that most chromosome abnormalities, including chromosome deletions, are the consequences of genome instability of cancer. In other words, cancers first have loss-of-function mutations on genes critical for genome integrity, such as TP53 and BRCA1/2, and then, as a consequence, these mutant cells acquire largely randomly chromosome deletions, together with many other genome abnormalities [1]. Though this hypothesis has been widely accepted, there are emerging evidences suggesting that at least it might not be the entire story. First, there are multiple cases of human cancer have chromosome large deletions or chromosome losses, while no mutation on any known genome integrity regulator genes [8]. More importantly, there are emerging evidences demonstrating the functions of chromosome deletions, as a whole, on tumor initiation and progress [9–12]. More and more tumor suppressors have been identified in these frequently deleted chromosome regions [13, 14]. Interestingly, co-deficiencies of these tumor suppressors in the same region promoted faster tumorigenesis than knockdown of any single tumor suppressor, suggesting the synergy of these tumor suppressors [8, 10]. Therefore, we propose a "synergy of multiple tumor suppressors" theory that there are multiple collaborating tumor suppressors in the common deleted regions in cancer, which make the chromosome large deletions more detrimental than single tumor suppressor mutations.

In this chapter we will focus on the studies of the functions and mechanisms of chromosome deletions on cancer and explain our "synergy of multiple tumor suppressors" theory.

9.2 Chromosome Deletions Are Frequent in Human Cancers and Associated with Poor Prognosis

9.2.1 The Boveri's Hypothesis on the Origin of Cancer

David Paul Hansemann was the first person to report unbalanced anaphases and telophases in freshly isolated epithelial cancer cells in 1890 [2]. He described in details the mitotic chromosomes of 13 cultured epithelial cancer cells and noted the aberrant multipolar mitoses and anaphases with asymmetrical distribution of "chromatin loops". However, Hansemann considered that these features were not unique to cancers. He thought that these chromosome alterations in tumor cells were the same process as in normal embryonic development [2, 3].

Soon after Hansemann's initial observation, Boveri made his own similar observations of hypo- and hyperchromacy and proposed his famous tumorigenesis hypothesis that "a tumor originates from a single cell in which there is a defined but incorrect combination of chromosomes" [4]. His work on sea urchin let him conclude that individual chromosome transmitted different inheritance factors. Therefore, Boveri is credited to the chromosome theory, together with Sutton [15]. Boveri applied his concept of chromosome to explain tumorigenesis and made many bold and accurate predictions, including the existence of tumour-suppressor genes ("teilungshemmende chromosomen") and oncogenes ("teilungsfoerdernde chromosomen"). Majority of Boveri's hypothesis and concepts have been approved and widely accepted by subsequent scientists.

Though chromosome alterations were frequently observed in cancers, Boveri's hypothesis on tumorigenesis was not well appreciated until the historic discovery of the Philadelphia chromosome in 1960 [5]. Nowell described the Philadelphia chromosome as the first consistent chromosome alteration in human cancers. Now the Philadelphia chromosome has become the golden standard to diagnose chronic myeloid leukemia and the resulting fusion protein BCR-ABL is the target of the first target therapy drug Gleevec [16–18]. The discovery of the Philadelphia chromosome and the target therapy against it opened new era for cancer research and clinical practice.

9.2.2 Chromosome Deletions Are a Hallmark of Human Cancers

Cancer cytogenetics is a new field ushered by the description of the Philadelphia chromosome for

the diagnosis and prognosis of human cancer, especially hematopoietic malignancies [6]. Over the last half century, a series of technique advances have improved karyotyping with high resolution, accuracy and convenience. In the late 1960s, Torbjorn Caspersson developed Q-binding staining to reveal unique banding patterns of each chromosome [19]. This staining is generally applied to detect multiple types of chromosome abnormalities, including translocations, deletions and inversions. Later molecular cytogenetics was developed with fluorescent- or radioisotope-labeled molecular probes. Labeled sequence-specific probes were hybridized with chromosomes with the techniques as fluorescence in situ hybridization (FISH) [20, 21]. In 1990s, array comparative genomic hybridization (aCGH) was applied to analyze copy number variations of cancer cells compared to reference samples [22, 23]. Microarrays used for aCGH can contain limited customized probes or millions of probes for the whole genome. The increased number of probes will improve the resolution of CGH analysis and less than 100 kb focal copy number variations can be detected. With the introduction of high-throughput sequencing techniques, CNV-seq reaches the highest resolution to single nucleotides [24, 25]. With these advanced technologies, accumulating chromosome deletions in human cancers have been documented [26].

Right now, up to millions of human cancers have been analyzed for their chromosome alterations (Table 9.1). It has been estimated that averagely about 30% of the genome is affected by chromosome arm-level or focal deletions in a typical human cancer [24, 25]. It seems that chromosome deletions in human cancers involve all regions of the genome. It is interesting that there are significant "peaks" of deletions and amplifications, while these peaks vary among different types of cancer. Some of these chromosome deletions are common among human cancers or a specific type of cancer. For example, one third of human cancers contain chromosome 17 loss or 17p deletions [10]. Chromosome 1p and 16q loss are common in solid cancer [27]. Acute myeloid leukemia (AML) frequently have chromosome 5 loss or 5q deletions (-5/del(5q)) and chromosome 7 loss or 7q deletions (about 10% in de novo AML and 50% in relapsed or treatment-related AML, respectively) [28, 29], while chromosome 3p

Table 9.1 The common chromosome deletions and their frequencies in selected types of human cancers

Disease category	Chromosome abnormality	Frequency of occurrence	References
Acute Myeloid Leukemia (AML)	-7/del(7q)	~10%	Greenberg et al. [31]
	-5/del(5q)	~10%	Nimer et al. [32]
	Del(20q)	~5%	Haase et al. [33]
	Del(17p)	~3%–4%	Valerie Soenen et al. [34] and Yvon Sterkers et al. [35]
Therapy-related AML	-7/del(7q)/-5/del(5q)	~75%	Smith et al. [36]
Non-Hodgkin lymphomas	Del(17p)	~19%	Levine et al. [37]
B-chronic lymphocyte leukemia	Del(13q)	~30%	Caporaso et al. [38]
Multiple myeloma	-13/del(13q)	~40%	Chng et al. [39]
Lung cancer	Del(13q)	~32%	Jun Yokota et al. [40]
	Del(17p)	~25%	Jun Yokota et al. [40]
Ovarian cancer	Del(17q)	~39%	Hiroko Saito et al. [41]
	Del(8p)	~33%	Mitsuru Emi et al. [42]
Breast cancer	Del(17q)	~41%	Hiroko Saito et al. [41]
	Del(10q23)	~40–48%	Garcia et al. [43]
	Del(8p)	~9%	Mitsuru Emi et al. [42]
Hepatocellular carcinoma	Del(8p)	~47%	Mitsuru Emi et al. [42]
Colorectal cancer	Del(8p)	~46%	Mitsuru Emi et al. [42]

deletions are detected in almost all small cell lung cancers and 90% of non-small cell lung cancers [30]. The high frequent common chromosome deletions suggest that these phenomena might be important to these diseases and of clinical value.

9.2.3 Chromosome Deletions Are Associated with Poor Prognosis in Some Cancers

Chromosome deletions, and other chromosome abnormalities have been widely applied for cancer diagnosis, prognosis and guiding clinical treatments. Back to 100 years ago, Boveri has proposed to detect malignant cells with chromosome abnormalities [4]. The Philadelphia chromosome is the golden marker for chronic myeloid leukemia [5].

Following Chromosome 5q deletion syndrome (5q- syndrome) is a hematopoietic disorder called myelodysplastic syndrome characterized with acquired interstitial chromosome 5q33.1 deletion and macrocytic anemia. In1974, Van den Berghe et al. reported the first 5q- syndrome [44]. Though most of these patients have only moderate thrombocytosis, erythroblastopenia, and megakaryocyte hyperplasia with a good prognosis, 10% of them would transform to AML [45, 46]. Generally these patients have less than 5% blast count in their peripheral blood and lenalidomide is the standard therapy. Interestingly, -5/del(5q) are one of the most frequent chromosome abnormalities in AML, especially relapsed or treatment-related AML. -5/del(5q) is associated with very poor prognosis, with less than 10% 5-year survival rate [47]. Of note, the chromosome regions involved in 5q- syndrome (5q33.1) and -5/del(5q) AML (5q31) are close but exclusive [48]. Thus characterizing chromosome deletions in detail is critic for clinic diagnosis and prognosis.

-7/del(7q) is the most frequent chromosome abnormality in AML, found in more than 50% secondary and 10% de novo myeloid disorders [49, 50]. Two minimal deleted regions, 7q22 and 7q35–36, have been mapped in -7/del(7q) AML [51, 52]. Both of them are associated with poor prognosis. While -7/del(7q) can happen independently, they also frequently co-occur with many other chromosome alterations, especially -5/del(5q) and -17/del(17p). When these multiple chromosome abnormalities happen together, these AML are called complex karyotype AML and have the worst prognosis with a 5-year survival of less than 5% [47].

Chromosome 17p deletions, generally involving the whole short arm of chromosome 17 and containing the well-known tumor suppressor TP53, are frequent in almost all human cancers, including AML, CLL and non-Hodgkin's lymphoma [53, 54]. In all of these cases, del(17P) are associated with poor prognosis [49, 55].

9.3 Identifying Tumor Suppressors in Chromosome Deletions

9.3.1 Knudson Theory

Given the frequency and prognosis value of chromosome deletions in human cancer, it is critical to understand the mechanisms of these chromosome abnormalities in cancer initiation, progress, metastasis and drug response. According to Boveri's theory, chromosome deletions would be rich of tumor suppressors [4]. Great efforts have done to uncover these functionally important genes over the last 30 years.

Traditionally there were two major criteria to identify tumor suppressors in chromosome loss or deleted regions. First the candidate tumor suppressors should located in the commonly deleted regions among multiple patients, echoing Koch's postulates. Chromosome loss or deletions generally involve large chromosome regions of several hundreds of genes, or the whole arms and sometimes the entire chromosomes of up to thousands of genes. In these cases, identifying critical tumor suppressors in these chromosome loss and deletions would be challenging [6, 10, 24]. To narrow down the candidate genes involved in specific types of cancer, a lot of work has been done to identify minimal deleted regions or commonly deleted regions among these patients, taking the

advantage of the variance of chromosome deletions and focal deletions in rare patients [56]. Recently, GISTIC (Genomic Identification of Significant Targets in Cancer), a powerful algorithm, is developed to identify tumor suppressors in chromosome deletion regions in cancer (and also oncogenic drivers in amplified regions) [57].

The second criterion is Knudson theory or the two-hit hypothesis. It was assumed that most of the mutations on tumor suppressors were loss-of-function mutations and recessive. Thus, both of the alleles of a putative tumor suppressor must be mutated. It is proposed that there is a first hit in a tumor suppressor, classically assumed to be a point mutation, and followed by a second hit, which is commonly thought as a chromosome deletion. This loss-of-heterozygosity hypothesis is called as the two-hit hypothesis, proposed by Alfred Knudson in 1971 [58]. Knudson theory has been the basis for identifying tumor suppressors during the last four decades [59].

The first example of Knudson theory is the retinoblastoma gene RB1 on chromosome 13q14. Knudson observed that retinoblastoma patients with bilateral retinoblastoma were first diagnosed at significantly earlier age than those patients with unilateral disease and sufferers of bilateral Rb1 were six times more likely to develop other cancer than those of a unilateral Rb1 [58]. Knudson explained that in the case of a bilateral Rb1 (familial form), one allele is already mutated in all somatic cells and only a second hit is needed to mutate the second working allele, a process of loss of Heterozygosity [60]. Thus, Knudson proposed his two-hit hypothesis through his studies on RB1.

Many negative regulators of cell cycle display similar mutation pattern as RB1. For example, cyclin-dependent kinase inhibitor 2A (CDKN2A) is a regulator of RB1 through inhibiting cyclin-dependent kinase 4 and 6, which in turn inhibiting RB1 [61]. Therefore CDKN2A blocks cells in from G1 phase to S phase. CDKN2A resides on chromosome 9p21, which is one of the most commonly deleted regions in human cancers, especially in melanoma, small cell lung cancer and lymphoma [62]. Similar to those with familial retinoblastoma, familiar melanoma patients are more likely to carry inherited mutations in one allele of CDKN2A gene, and the second allele of this loci is deleted through the loss-of-heterozygosity process.

TP53 is the most frequently mutated tumor suppressor in human cancers, which is also recognized as an example for Kudson theory. Interestingly, TP53 was first found to be overexpressed in many human cancers, which is in contrast to classic tumor suppressors. Therefore it was assumed to be an oncogene at the beginning instead of tumor suppressor. Later, it turned out that the overexpressed "TP53" is a gain-of-function mutant and TP53 fits to the classic two-hit tumor suppressor [63]. TP53 is located on chromosome 17p13. Generally one allele of TP53 carries missense or frameshift mutations, with hotspots on R175, R248 and R273, which have been confirmed as gain-of-function mutations, and the second allele is generally deleted together with the whole short arm of chromosome 17 [64]. Familial TP53 mutations count for about half of Li-Fraumeni syndrome, almost all of these patients would develop multiple types of cancers, including sarcoma, leukemia, breast cancer and brain cancers as results of loss of heterozygosity of TP53 [65].

Following these examples, great efforts have been applied to reveal putative tumor suppressors in chromosome deletions through mapping minimal deleted regions to narrow down the candidate genes and searching the point mutations or epigenetic silencing on the second allele as an evidence of loss of heterozygosity [59]. A long list of tumor suppressors, including PTEN on chromosome 10q23 [66], APC on chromosome 5q22 [54], NF1 on chromosome 17q11 [67], BRCA1 on chromosome 17q21 [68] and VHL on chromosome 3p25 [69], have been identified.

9.3.2 Haploinsufficient Tumor Suppressors

Despite the large success of Knudson theory, there are two obvious puzzles about chromosome deletions in human cancers. First there are no verified classic tumor suppressors in many chromosome deletions even after great efforts of

searching. And second, chromosome deletions generally contain several hundreds genes while only one or very few of them have been validated as tumor suppressors [24, 25]. These contradictions suggest that classic tumor suppressors consistent with the two-hit hypothesis might not be the whole stories. Around 2000, a novel type of tumor suppressors, haploinsufficient tumor suppressor, has been proposed. Heterozygous loss of function of these genes, such as mutations or deletions on only one allele (and the second allele is still functioning), would contribute tumor genesis and progression [70, 71]. The new concept of haploinsufficiency dramatically expands the candidate genes for tumor suppressors, especially in chromosome deletion regions.

One of the first identified haploinsufficient tumor suppressor is the cyclin-dependent kinase inhibitor p27Kip1 [72]. p27kip1, a regulator of RB1-E2F pathway, is in chromosome 12p12, a region frequently deleted in pediatric acute lymphoblastic leukemia. All deletions involved chromosome 12p12 are heterozygous while neither missense nor truncated mutations were detected in the retained allele [73, 74]. And expression of p27Kip1 was detected in the nuclei of these effected cancer cells by immunostaining though at a reduced levels [72], suggesting a non-Knudson mechanism. With a genetically engineered mouse model, Fero et al. clearly demonstrated that p27Kip1 heterozygous loss resulted in spontaneous multiple organ tumors at a penetrance of 32% in mice. When exposed to X-ray irradiation, these mice developed dramatically more tumors than wildtype control mice, though fewer than those of p27Kip1 homozygous loss. More importantly, all of the tumors from p27Kip1+/− mice retained the wildtype allele and the expression of p27Kip1 were revealed by west blotting [72]. Thus p27Kip1 is a haploinsufficient tumor suppressor.

Haploinsufficent tumor suppressors may also residue in chromosome 7q, the most frequently deleted region in AML. Since its mapping by cytogenetics, great efforts of decades to identify classic tumor suppressors in this region have been in vain. By analyzing the big data of cancer genomics and in vivo function tests, we showed that the mixed lineage leukemia 3 gene, MLL3, was a haploinsufficient tumor suppressor in chromosome 7q36 [13]. MLL3 is a member of the MLL protein family with a SET domain capable of methylating lysine 4 on histone H3 and a core component of the COMPASS-like complex regulating transcription elongation [75]. MLL3 is one of the most frequently mutated chromatin modifiers in solid cancers. But all of these mutations are heterozygous [76, 77]. 7q is the most commonly deleted region in AML but so far no loss-of-function mutation of MLL3 (nor other genes) was found in 7q loss patients [49]. shRNAs knocking down Mll3, together with p53 and Nf1 loss, promoted full blown AML genesis, indicating Mlll3 as a tumor suppressor. Though these shRNAs could potently reduce the expression level of Mll3 in NIH3T3 cells at 90%, the inhibition of Mll3 expression by the same shRNAs in the resulting AML cells were only about 50%. Further CRISPR/Cas9-mediated genome editing of Mll3 leukemia cells also remained one intact wildtype allele. All of these evidences demonstrated that Mll3 is a haploinsufficient tumor suppressor in AML [13]. These results are striking given that MLL3 is an epigenetic regulator, which affects the expressions of many downstream genes but at a moderate level. The remaining questions would be how the moderate dosage change of an epigenetic gene would transform hematopoietic stem cells and whether restoring the expression of MLL3 (two-fold increase) in leukemia would be able to restrain the progression of the disease.

Interestingly, many of the putative classic tumor suppressors also show haploinsufficiency in preventing tumorigenesis. One example is PTEN, residing in chromosome 10q23 and encoding a lipid phosphatase that negatively regulates PI3K-AKT pathway [78]. It was estimated that up to 70% prostate cancer patients carried a heterozygous loss of PTEN, generally covered by a large deletion of one copy of chromosome 10 similar to MLL3 in chromosome 7q, while only less than 10% of the patients had homozygous deletions or mutations at diagnosis [79]. Consistent with the human clinic genetics, Pten Heterozygosity dramatically increased the rate of prostate cancer progression in TRAMP mice

[80]. Later, with a Pten hypermorphic mouse model whose expression level of Pten was 80% of that in wildtype control mice Alimonti et al. reported that even such subtle reduction of Pten dosage would promote the development of a wide spectrum of cancers [81]. Thus haploinsufficiency is a general principle for tumorigenesis.

Arguably all potential tumor suppressors in chromosome deletions without loss-of-function mutations on the second allele may be haploinsufficient tumor suppressors, which would strikingly deep our understanding of the molecular mechanisms of chromosome deletions in human cancers. It is also interesting to test whether these haploinsufficient tumor suppressors might be valuable therapeutic targets for the cancers with the corresponding defects.

9.4 The Role of Chromosome Deletions as a Whole in Carcinogenesis

9.4.1 Modeling Chromosome Deletions with Genetic Engineered Mouse Models

Identifying tumor suppressors in chromosome deletions is very important to study the functions of chromosome deletions in tumorigenesis. However, given the broad effects of chromosome deletions with generally several hundreds genes and structural abnormalities, none of any single tumor suppressor could recapitulate all of the phenomena of a chromosome deletion in cancer. Thus the full functions of chromosome deletions must be studied as a whole. Investigating the biological roles of chromosome deletions as a whole has been significantly delayed due to lack of available techniques to precisely model these chromosome configurators and confused by the results of spontaneous aneuploidies. At odds to being a hallmark of cancer, aneuploidy, including chromosome loss and large chromosome deletions, has been shown to be detrimental to normal cells, specifically yeast cells and mouse embryonic fibroblast cells, in some context [82]. It is argued that both gene-specific and general non-gene-specific effects of aneuploidy could interfere cell proliferation through "aneuploidy associated stresses". These experimental observations seem at odds with the frequent chromosome alterations associated with human cancers and Boveri's chromosome theory of carcinogenesis [83]. Therefore it is critical to provide direct evidences that chromosome deletions are able to drive tumorigenesis.

Recent technique advances including sophisticated genetically engineered mouse modeling, genome editing and high throughput library screening, made it possible to reveal the biological consequences of chromosome deletions in cancer [14, 84–87]. The first example is chromosome 17p deletion [10]. 17p loss is one of the most, if not the most, frequently genetical abnormalities found in various cancers and associates with tumor aggressiveness and poor prognosis [88]. Given the well-studied tumor suppressor TP53 on chromosome 17p13, it was generally assumed that chromosome 17p loss is to loss of Heterozygosity of the second allele of TP53, following the classic Knudson theory [63]. However, by analyzing the CNV and mutation data of more than 4000 human cancers, we found that one third of cases with TP53 alterations had heterozygous chromosome 17p loss but didn't have any detectable mutation of TP53 on the other allele [10]. Therefore it is very important to investigate whether chromosome 17p has more tumor suppression capacity beyond TP53 only. Taking the advantage of the high synteny between mouse chromosome 11B3 and human chromosome 17p13, which share the exact same over than 100 coding genes and noncoding microRNA genes even at the same order, we genetically engineered a conditional 11B3 knockout mouse model. Compared to p53 deleted tumors, heterozygous deletion of chromosome 11B3 can promote either Myc-driven lymphomagenesis or Nf1; Mll3-defective leukemogenesis with shorter tumor latency and overall survival. Moreover, the resulting 11B3-deleted tumor cells are more resistant to chemodrug like cyclophosphamide, vincristine and methotrexate. Interestingly, many lymphomas generated from heterozygous deletion of 11B3 carry spontaneously missense or frame-

shift mutations on the wildtype p53 allele, likely resulting from the procession of *Trp53* loss-of-heterozygosity. Other 11B3-deleted lymphomas keep wildtype Trp53 allele. Together, 11B3 tumors represent the chromosome 17p deletion configurations in human cancers [10]. These findings would not only shed light on understanding the molecular mechanisms under which chromosome 17p deletions impact on cancer biology, but also provide a platform to develop new therapeutic methods.

Chromosome 7q22 is another frequently deleted region in AML and so far no classic tumor suppressor has been validated in the context of AML [89]. To shed light on the sealed function of 7q22 deletions to Myelodysplastic Syndrome (MDS) pathogenesis, Wong et al. generated mice with a heterozygous germ line deletion of a 2 Mb interval of the murine chromosome band 5A3, which removing 13 genes correspondent to a commonly deleted segment of human 7q22 [12]. The resulting 5A3+/del mice exhibited typical characterizations of MDS. The 5A3+/del mouse model provided a novel platform for the studies of human 7q22 deletion MDS or AML.

These genetically engineered mouse models provide clear and direct evidences that chromosome deletions as a whole can be drivers of tumorigenesis and experimentally prove the 100-year-old Boveri's cancer theory. However, the big limitation of this strategy is that, though 99% of human and mouse genes are identical, the synteny between human and mouse chromosomes are poor [90, 91]. Therefore it is difficult to model chromosome large deletions of human cancers in mouse models.

9.4.2 Modeling Chromosome Deletions in Human Cell Models

Obviously human cells can be the best model to study chromosome alterations in human cancers. The efficiency of genome editing made it feasible [92]. It is widely known that chromosome 8p loss recurrently occurs in human breast cancer patients and it is tightly associated with poor patient survival. In order to elucidate the role of 8p loss in tumorigenic transformation, Cai et al. made a good use of TALEN-directed genomic engineering technology to generate human cellular models based on an non-malignant MCF10A mammary epithelial cell line, which mimicking 8p loss of heterozygosity and avoiding introducing other genomic abnormalities [9]. Though the entire loss of 8p chromosome showed limited tumor transformation capacity alone or cooperating with other driver genes like MYC, ERBB2 or loss of TP53, these cells displayed abnormal fatty acid and ceramide metabolism. The shift of fatty acid metabolism led to actin filament reorganization and further contributed to cell invasiveness. Besides, alterations in ceramide metabolism rendered cells increased autophagy capacity and better growth ability under hypoxia context. Primary human breast cancers with 8p loss deriving from clinical patients bear these metabolic changes as well. These discoveries suggest that models of chromosomal large deletions could be used to predict the responsiveness of cancer patients to anticancer therapies and could help to improve our understandings of human cancer [93].

Taking advantage of induce pluripotent stem cells (iPSCs), Papapetrou's laboratory investigated the biological consequences of chromosome 7q loss, the most frequent chromosome abnormalities in AML [11, 94]. First they generated iPS cells from chromosome 7q loss and intact cells from the same patients and showed that iPS cells with chromosome 7q deletions had defects to differentiate into hematopoietic cells and had increased apoptosis, similar to those observed in MDS patients with chromosome 7q deletions. Then using AAV-delivered CRISPR/Cas9, they generated chromosome 7q deletions in normal human iPS cells. These genome edited iPS cells also displayed reduced capacity to differentiate into CD45+ hematopoietic cells while increased percentage of CD34+ (a marker of hematopoietic stem and progenitor cells) population. These phenotypes are consistent with those in chromosome 7q deleted MDS patients [11]. It is of interest that spontaneous correction of chromosome 7q by a chromosome 7 trisomy largely rescued most of these abnormalities associated

with the disease [94]. These studies indicate that chromosome 7q deletions as a whole are responsible for the pathology of MDS with chromosome 7q loss. The combination of iPS cells and genome editing opens a new era to study chromosome alterations in human cancers. In principle, this strategy could model all kinds of chromosome deletions in various types of human cancers [95]. A shortcoming is that in patients somatic chromosome deletions assumably occur in tissue-specific stem or progenitor cells while genome edited iPS cells are not physiologically related. Thus direct genome editing of cell-of-origin of human cancers might be more accurate to investigate the biological functions of chromosome abnormalities in the right context.

A new era in preclinical cancer research is emerging, in which human-based models are taking center stage and patient-derived cells are increasingly being used as primary discovery platforms. In this modern era of basic cancer research and precision oncology, iPSCs derived from patients with cancer can substantially expand the experimental repertoire applicable to human cells in ways that were hitherto restricted to model organisms. We envision that models for at least some cancers can be developed using iPSC technologies, and that these will occupy a unique place in this new era, bridging primary cells with immortalized cell lines by combining the physiological relevance of the former with the amenability to experimentation of the latter. Interdisciplinary collaborations between stem cell researchers, cancer researchers, physicians, translational scientists, bioengineers and drug developers will be paramount to harness the full potential of iPSCs as a new tool in this modern era of cancer research.

9.4.3 The Collaborative Effect of Multiple Tumor Suppressors in Chromosome Deletions

There are accumulating evidences indicating that chromosome deletions are powerful drivers for carcinogenesis and distinguishable to deficiency of single tumor suppressors. A plausible explanation is that there are multiple tumor suppressors in a chromosome deletion region and these tumor suppressors collaborate to inhibit tumor genesis and progress. To dissect these cooperative tumor suppressors, shRNA, CRISPR/Cas9 and ORF library screening have been successfully performed on several commonly deleted chromosome regions. Since chromosome 17p has tumor suppression capacity beyond TP53, it was proposed that there were other tumor suppressors besides TP53 in this region. To identify potential new tumor suppressors in chromosome 17p, Liu et al. generated a shRNA library against all of the coding genes except p53 and performed a high throughput in vivo screening. Multiple candidate tumor suppressors, including a cluster of Alox genes, were scored. After validating Eif5a and Alox15b as tumor suppressors in lymphoma, they further showed that simultaneously knocking down Eif5a and p53, or Alox15b and p53 led to shorter tumor-free survival of recipient mice compared to knocking down any single of these genes, indicating the collaboration between Eif5a and p53, and Alox15b and p53, respectively [10]. Kotini et al. applied ORF screening to identify key players in chromosome 7q with an iPS cell-blood cell differentiation assay. Multiple candidate tumor suppressors were hit and further work is needed to validate them in the context of AML genesis (Fig. 9.1) [11].

More high throughput library screenings have been performed in multiple cancer types. Zender et al. did in vivo shRNA library screening for genes recurrently deleted in human HCC cells in a mouse HCC model and identified 12 novel tumor suppressors [14]. Further they showed that these tumor suppressors from chromosome 8p could synergistically restrained HCC growth at least in mice [96]. A survey of genes in 82 recurrently focal deletions from 3131 tumors, Solimini demonstrated that these regions are rich of so called STOP genes than GO genes, which negatively and positively regulated cell growth and proliferation. They proposed that though majority of these STOP genes were hemizygously deleted and each of them had moderate effects on tumorigenesis, the cumulative haploinsufficien-

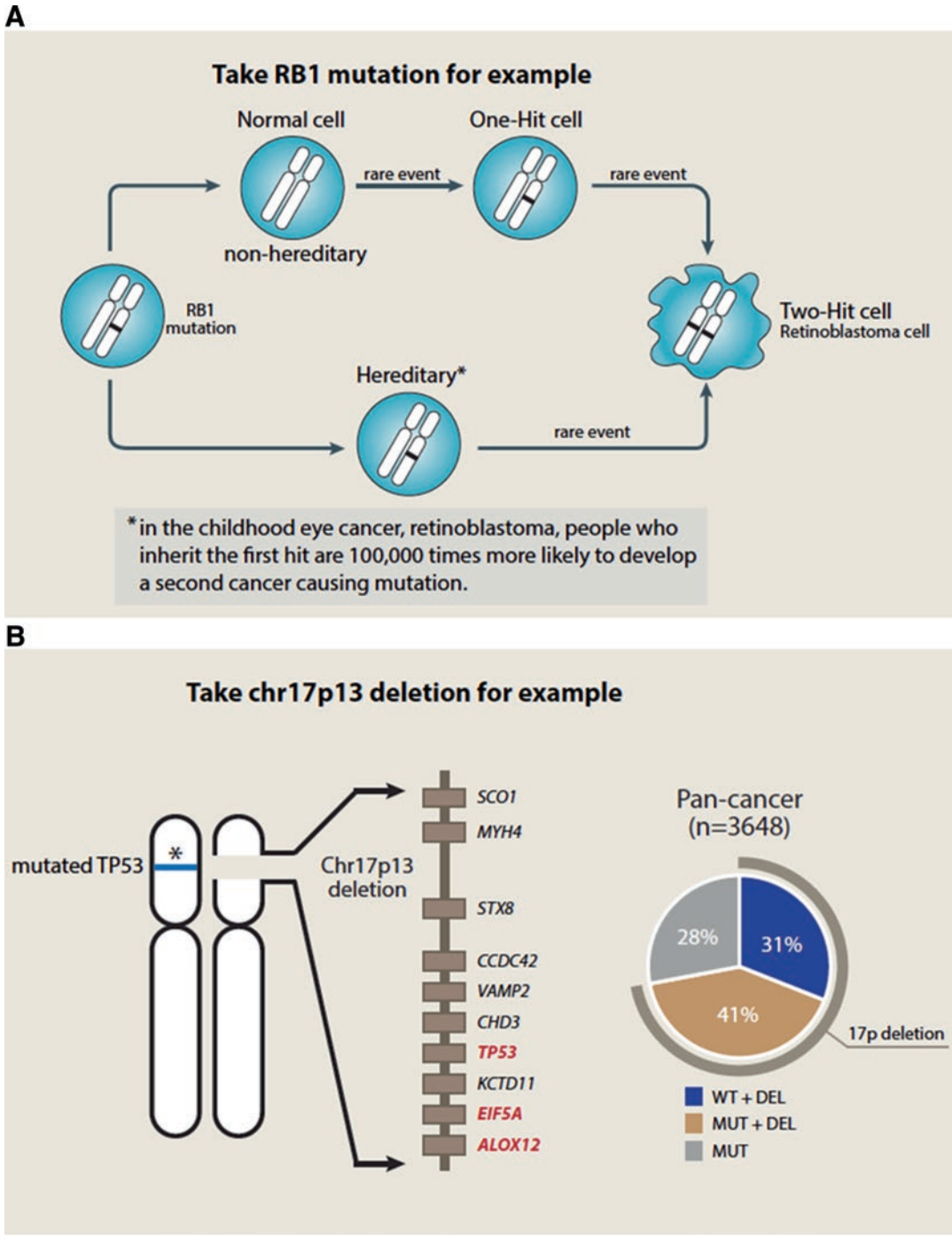

Fig. 9.1 (**a**) Knudson "Two-hit" theory of tumorigenesis. (**b**) "synergy of multiple tumor suppressors" theory on the role of chromosome large deletions in human cancers

cies led to tumorigenesis, which explained the driver role of chromosome deletions in human cancer [8, 97].

9.5 Perspective

It has been over 100 years since Hansemann's initial observations of chromosome abnormalities in cancer and Boveri's seminal hypothesis of chromosome alterations as drivers of cancer. Amounting data have documented them as a hallmark and association with pathology and prognosis of cancer. However, partially due to the technical challenges, we just start to understand the mechanisms of this critical phenomenon in cancer with both conceptual and technic breakthroughs. Solid evidences have provided that chromosome deletions are distinguishable and powerful drivers of cancers. These critical drivers display significant characteristics in terms of genetic configurations, biological consequences and more important, treatment vulnerabilities [98]. For example, passenger deletions of ENO1 in chromosome 1p36 give rise to sensitivity of the mutant GBM cells to ENO2 inhibition [99]. Chromosome deletions, together with other chromosome abnormalities, might also change the expressions of certain immune markers through unknown mechanisms, rendering the affected cancer cells resistance to immunotherapies [100]. Thus further efforts are in need to fully understand the biological functions, molecular mechanisms and vulnerabilities for the treatment of the diseases driven by these numerous and notorious chromosome abnormalities.

References

1. Hanahan D, Weinberg RA (2011) Hallmarks of cancer: the next generation. Cell 144:646–674. https://doi.org/10.1016/j.cell.2011.02.013
2. Hansemann D (1890) Asymmetrical cell division in epithelial cancers and its biological significance. Arch Pathol Anat etc Berl 119:299–326
3. Hansemann D (1891) Festschr. Rudolf Virchow, von seine Assistenten, Berl:1–12
4. Boveri T (1929) The origin of malignant tumors. Williams and Wilkins, Boston
5. Nowell PCH (1960) D.A. a minute chromosome in human chronic granulocytic leukemia. Science 132:1497
6. Shlien A, Malkin D (2009) Copy number variations and cancer. Genome Med 1:62. https://doi.org/10.1186/gm62
7. Sherr CJ (2004) Principles of tumor suppression. Cell 116:235–246
8. Solimini NL et al (2012) Recurrent hemizygous deletions in cancers may optimize proliferative potential. Science 337:104–109. https://doi.org/10.1126/science.1219580
9. Cai Y et al (2016) Loss of chromosome 8p governs tumor progression and drug response by altering lipid metabolism. Cancer Cell 29:751–766. https://doi.org/10.1016/j.ccell.2016.04.003
10. Liu Y et al (2016) Deletions linked to TP53 loss drive cancer through p53-independent mechanisms. Nature 531:471–475. https://doi.org/10.1038/nature17157
11. Kotini AG et al (2015) Functional analysis of a chromosomal deletion associated with myelodysplastic syndromes using isogenic human induced pluripotent stem cells. Nat Biotechnol 33:646–655. https://doi.org/10.1038/nbt.3178
12. Wong JC et al. (2015) Functional evidence implicating chromosome 7q22 haploinsufficiency in myelodysplastic syndrome pathogenesis. Elife 4. https://doi.org/10.7554/eLife.07839
13. Chen C et al (2014) MLL3 is a haploinsufficient 7q tumor suppressor in acute myeloid leukemia. Cancer Cell 25:652–665. https://doi.org/10.1016/j.ccr.2014.03.016
14. Zender L et al (2008) An oncogenomics-based in vivo RNAi screen identifies tumor suppressors in liver cancer. Cell 135:852–864. https://doi.org/10.1016/j.cell.2008.09.061
15. Mader SS (2007) Biology. 9th edn. McGraw Hill Higher Education, New York
16. Rowley JDL (1973) A new consistent chromosomal abnormality in chronic myelogenous leukaemia identified by quinacrine fluorescence and Giemsa staining. Nature 243:290–293
17. Druker BJ et al (2001) Activity of a specific inhibitor of the BCR-ABL tyrosine kinase in the blast crisis of chronic myeloid leukemia and acute lymphoblastic leukemia with the Philadelphia chromosome. N Engl J Med 344:1038–1042. https://doi.org/10.1056/NEJM200104053441402
18. Druker BJ et al (2001) Efficacy and safety of a specific inhibitor of the BCR-ABL tyrosine kinase in chronic myeloid leukemia. N Engl J Med 344:1031–1037. https://doi.org/10.1056/NEJM200104053441401
19. Caspersson T, Zech L, Johansson C, Modest EJ (1970) Identification of human chromosomes by DNA-binding fluorescent agents. Chromosoma 30:215–227
20. Speicher MR, Carter NP (2005) The new cytogenetics: blurring the boundaries with molecular biology.

Nat Rev Genet 6:782–792. https://doi.org/10.1038/nrg1692
21. Langer-Safer PR, Levine M, Ward DC (1982) Immunological method for mapping genes on Drosophila polytene chromosomes. Proc Natl Acad Sci U S A 79:4381–4385
22. Kallioniemi A et al (1992) Comparative genomic hybridization for molecular cytogenetic analysis of solid tumors. Science 258:818–821
23. Pinkel D, Albertson DG (2005) Array comparative genomic hybridization and its applications in cancer. Nat Genet 37(Suppl):S11–S17. https://doi.org/10.1038/ng1569
24. Beroukhim R et al (2010) The landscape of somatic copy-number alteration across human cancers. Nature 463:899–905. https://doi.org/10.1038/nature08822
25. Bignell GR et al (2010) Signatures of mutation and selection in the cancer genome. Nature 463:893–898. https://doi.org/10.1038/nature08768
26. Meyerson M, Gabriel S, Getz G (2010) Advances in understanding cancer genomes through second-generation sequencing. Nat Rev Genet 11:685–696. https://doi.org/10.1038/nrg2841
27. Albertson DG, Collins C, McCormick F, Gray JW (2003) Chromosome aberrations in solid tumors. Nat Genet 34:369–376. https://doi.org/10.1038/ng1215
28. Kenneth Kaushansky ML, Prchal J, Levi MM, Press O, Burns L, Caligiuri M (2015) Williams hematology, 9th edn. McGraw-Hill Education, New York
29. Neuman WL et al (1992) Chromosomal loss and deletion are the most common mechanisms for loss of heterozygosity from chromosomes 5 and 7 in malignant myeloid disorders. Blood 79:1501–1510
30. Zabarovsky ER, Lerman MI, Minna JD (2002) Tumor suppressor genes on chromosome 3p involved in the pathogenesis of lung and other cancers. Oncogene 21:6915–6935. https://doi.org/10.1038/sj.onc.1205835
31. Greenberg P et al (1997) International scoring system for evaluating prognosis in myelodysplastic syndromes. Blood 89:2079–2088
32. Nimer SD (2006) Clinical management of myelodysplastic syndromes with interstitial deletion of chromosome 5q. J Clin Oncol 24:2576–2582
33. Haase D et al (2007) New insights into the prognostic impact of the karyotype in MDS and correlation with subtypes: evidence from a core dataset of 2124 patients. Blood 110:4385–4395
34. Soenen V et al (1998) 17p deletion in acute myeloid leukemia and myelodysplastic syndrome. Analysis of breakpoints and deleted segments by fluorescence in situ. Blood 91:1008–1015
35. Sterkers Y et al (1998) Acute myeloid leukemia and myelodysplastic syndromes following essential thrombocythemia treated with hydroxyurea: high proportion of cases with 17p deletion. Blood 91:616–622
36. Smith SM et al (2003) Clinical-cytogenetic associations in 306 patients with therapy-related myelodysplasia and myeloid leukemia: the University of Chicago series. Blood 102:43–52
37. Levine EG et al (1990) Sequential karyotypes in non-Hodgkin lymphoma: their nature and significance. Genes Chromosom Cancer 1:270–280
38. Caporaso N et al (2007) Chronic lymphocytic leukaemia genetics overview. Brit J Haematol 139:630–634
39. Chng W, Glebov O, Bergsagel P, Kuehl W (2007) Genetic events in the pathogenesis of multiple myeloma. Best Pract Res Clin Haematol 20:571–596
40. Yokota J, Wada M, Shimosato Y, Terada M, Sugimura T (1987) Loss of heterozygosity on chromosomes 3, 13, and 17 in small-cell carcinoma and on chromosome 3 in adenocarcinoma of the lung. Proc Natl Acad Sci 84:9252–9256
41. Saito H et al (1993) Detailed deletion mapping of chromosome 17q in ovarian and breast cancers: 2-cM region on 17q21. 3 often and commonly deleted in tumors. Cancer Res 53:3382–3385
42. Emi M et al (1992) Frequent loss of heterozygosity for loci on chromosome 8p in hepatocellular carcinoma, colorectal cancer, and lung cancer. Cancer Res 52:5368–5372
43. Garcia JM et al (1999) Allelic loss of the PTEN region (10q23) in breast carcinomas of poor pathophenotype. Breast Cancer Res Treat 57:237–243
44. Van den Berghe H et al (1974) Distinct haematological disorder with deletion of long arm of no. 5 chromosome. Nature 251:437–438
45. List A et al (2006) Lenalidomide in the myelodysplastic syndrome with chromosome 5q deletion. N Engl J Med 355:1456–1465. https://doi.org/10.1056/NEJMoa061292
46. Boultwood J, Pellagatti A, McKenzie AN, Wainscoat JS (2010) Advances in the 5q- syndrome. Blood 116:5803–5811. https://doi.org/10.1182/blood-2010-04-273771
47. Dastugue N et al (1995) Prognostic significance of karyotype in de novo adult acute myeloid leukemia. The BGMT group. Leukemia 9:1491–1498
48. Qian Z et al (2010) Cytogenetic and genetic pathways in therapy-related acute myeloid leukemia. Chem Biol Interact 184:50–57. https://doi.org/10.1016/j.cbi.2009.11.025
49. Johnson E, Cotter FE (1997) Monosomy 7 and 7q--associated with myeloid malignancy. Blood Rev 11:46–55
50. Jerez A et al (2012) Loss of heterozygosity in 7q myeloid disorders: clinical associations and genomic pathogenesis. Blood 119:6109–6117. https://doi.org/10.1182/blood-2011-12-397620
51. Le Beau MM et al (1996) Cytogenetic and molecular delineation of a region of chromosome 7 commonly deleted in malignant myeloid diseases. Blood 88:1930–1935
52. Liang H et al (1998) Molecular anatomy of chromosome 7q deletions in myeloid neoplasms: evidence for multiple critical loci. Proc Natl Acad Sci U S A 95:3781–3785

53. Jary L et al (1997) The 17p-syndrome: a distinct myelodysplastic syndrome entity? Leuk Lymphoma 25:163–168. https://doi.org/10.3109/10428199709042506
54. Sankar M et al (1998) Identification of a commonly deleted region at 17p13.3 in leukemia and lymphoma associated with 17p abnormality. Leukemia 12:510–516
55. Miller LD et al (2005) An expression signature for p53 status in human breast cancer predicts mutation status, transcriptional effects, and patient survival. Proc Natl Acad Sci U S A 102:13550–13555. https://doi.org/10.1073/pnas.0506230102
56. Rowley JD, Le Beau MM (1989) Cytogenetic and molecular analysis of therapy-related leukemia. Ann N Y Acad Sci 567:130–140
57. Mermel CH et al (2011) GISTIC2.0 facilitates sensitive and confident localization of the targets of focal somatic copy-number alteration in human cancers. Genome Biol 12:R41. https://doi.org/10.1186/gb-2011-12-4-r41
58. Knudson AG Jr (1971) Mutation and cancer: statistical study of retinoblastoma. Proc Natl Acad Sci U S A 68:820–823
59. Knudson AG (2001) Two genetic hits (more or less) to cancer. Nat Rev Cancer 1:157–162. https://doi.org/10.1038/35101031
60. Machiela MJ et al (2016) Mosaic 13q14 deletions in peripheral leukocytes of non-hematologic cancer cases and healthy controls. J Hum Genet 61:411–418. https://doi.org/10.1038/jhg.2015.166
61. Witkiewicz AK, Knudsen KE, Dicker AP, Knudsen ES (2011) The meaning of p16(ink4a) expression in tumors: functional significance, clinical associations and future developments. Cell Cycle 10:2497–2503. https://doi.org/10.4161/cc.10.15.16776
62. Sulong S et al (2009) A comprehensive analysis of the CDKN2A gene in childhood acute lymphoblastic leukemia reveals genomic deletion, copy number neutral loss of heterozygosity, and association with specific cytogenetic subgroups. Blood 113:100–107. https://doi.org/10.1182/blood-2008-07-166801
63. Levine AJ, Oren M (2009) The first 30 years of p53: growing ever more complex. Nat Rev Cancer 9:749–758. https://doi.org/10.1038/nrc2723
64. Olivier M, Hollstein M, Hainaut P (2010) TP53 mutations in human cancers: origins, consequences, and clinical use. Cold Spring Harb Perspect Biol 2:a001008. https://doi.org/10.1101/cshperspect.a001008
65. Varley J, Germline M (2003) TP53 mutations and Li-Fraumeni syndrome. Hum Mutat 21:313–320. https://doi.org/10.1002/humu.10185
66. Steck PA et al (1997) Identification of a candidate tumour suppressor gene, MMAC1, at chromosome 10q23.3 that is mutated in multiple advanced cancers. Nat Genet 15:356–362. https://doi.org/10.1038/ng0497-356
67. Costa RM, Silva AJ (2002) Molecular and cellular mechanisms underlying the cognitive deficits associated with neurofibromatosis 1. J Child Neurol 17:622–626; discussion 627–629, 646–651. https://doi.org/10.1177/088307380201700813
68. Albertsen H et al (1994) Genetic mapping of the BRCA1 region on chromosome 17q21. Am J Hum Genet 54:516–525
69. Nordstrom-O'Brien M et al (2010) Genetic analysis of von Hippel-Lindau disease. Hum Mutat 31:521–537. https://doi.org/10.1002/humu.21219
70. Santarosa M, Ashworth A (2004) Haploinsufficiency for tumour suppressor genes: when you don't need to go all the way. Biochim Biophys Acta 1654:105–122. https://doi.org/10.1016/j.bbcan.2004.01.001
71. Quon KC, Berns A (2001) Haplo-insufficiency? Let me count the ways. Genes Dev 15:2917–2921. https://doi.org/10.1101/gad.949001
72. Fero ML, Randel E, Gurley KE, Roberts JM, Kemp CJ (1998) The murine gene p27Kip1 is haplo-insufficient for tumour suppression. Nature 396:177–180. https://doi.org/10.1038/24179
73. Komuro H et al (1999) p27KIP1 deletions in childhood acute lymphoblastic leukemia. Neoplasia 1:253–261
74. Kawamata N et al (1995) Molecular analysis of the cyclin-dependent kinase inhibitor gene p27/Kip1 in human malignancies. Cancer Res 55:2266–2269
75. Shilatifard A (2012) The COMPASS family of histone H3K4 methylases: mechanisms of regulation in development and disease pathogenesis. Annu Rev Biochem 81:65–95. https://doi.org/10.1146/annurev-biochem-051710-134100
76. Zang ZJ et al (2012) Exome sequencing of gastric adenocarcinoma identifies recurrent somatic mutations in cell adhesion and chromatin remodeling genes. Nat Genet 44:570–574. https://doi.org/10.1038/ng.2246
77. Kandoth C et al (2013) Mutational landscape and significance across 12 major cancer types. Nature 502:333–339. https://doi.org/10.1038/nature12634
78. Song MS, Salmena L, Pandolfi PP (2012) The functions and regulation of the PTEN tumour suppressor. Nat Rev Mol Cell Biol 13:283–296. https://doi.org/10.1038/nrm3330
79. Chen Z et al (2005) Crucial role of p53-dependent cellular senescence in suppression of Pten-deficient tumorigenesis. Nature 436:725–730. https://doi.org/10.1038/nature03918
80. Kwabi-Addo B et al (2001) Haploinsufficiency of the Pten tumor suppressor gene promotes prostate cancer progression. Proc Natl Acad Sci U S A 98:11563–11568. https://doi.org/10.1073/pnas.201167798
81. Alimonti A et al (2010) Subtle variations in Pten dose determine cancer susceptibility. Nat Genet 42:454–458. https://doi.org/10.1038/ng.556
82. Tang YC, Amon A (2013) Gene copy-number alterations: a cost-benefit analysis. Cell 152:394–405. https://doi.org/10.1016/j.cell.2012.11.043
83. Sheltzer JM et al (2017) Single-chromosome gains commonly function as tumor suppressors.

Cancer Cell 31:240–255. https://doi.org/10.1016/j.ccell.2016.12.004
84. Adams DJ et al (2004) Mutagenic insertion and chromosome engineering resource (MICER). Nat Genet 36:867–871. https://doi.org/10.1038/ng1388
85. Cong L et al (2013) Multiplex genome engineering using CRISPR/Cas systems. Science 339:819–823. https://doi.org/10.1126/science.1231143
86. Mali P et al (2013) RNA-guided human genome engineering via Cas9. Science 339:823–826. https://doi.org/10.1126/science.1232033
87. Shalem O et al (2014) Genome-scale CRISPR-Cas9 knockout screening in human cells. Science 343:84–87. https://doi.org/10.1126/science.1247005
88. Petitjean A, Achatz MI, Borresen-Dale AL, Hainaut P, Olivier M (2007) TP53 mutations in human cancers: functional selection and impact on cancer prognosis and outcomes. Oncogene 26:2157–2165. https://doi.org/10.1038/sj.onc.1210302
89. Johnson EJ et al (1996) Molecular definition of a narrow interval at 7q22.1 associated with myelodysplasia. Blood 87:3579–3586
90. Schwartz S et al (2003) Human-mouse alignments with BLASTZ. Genome Res 13:103–107. https://doi.org/10.1101/gr.809403
91. Wasserman WW, Palumbo M, Thompson W, Fickett JW, Lawrence CE (2000) Human-mouse genome comparisons to locate regulatory sites. Nat Genet 26:225–228. https://doi.org/10.1038/79965
92. Sanchez-Rivera FJ, Jacks T (2015) Applications of the CRISPR-Cas9 system in cancer biology. Nat Rev Cancer 15:387–395. https://doi.org/10.1038/nrc3950
93. Tschaharganeh DF, Bosbach B, Lowe SW (2016) Coordinated tumor suppression by chromosome 8p. Cancer Cell 29:617–619. https://doi.org/10.1016/j.ccell.2016.04.011
94. Kotini AG et al (2017) Stage-specific human induced pluripotent stem cells map the progression of myeloid transformation to transplantable leukemia. Cell Stem Cell 20:315–328 e317. https://doi.org/10.1016/j.stem.2017.01.009
95. Papapetrou EP (2016) Patient-derived induced pluripotent stem cells in cancer research and precision oncology. Nat Med 22:1392–1401. https://doi.org/10.1038/nm.4238
96. Xue W et al (2012) A cluster of cooperating tumor-suppressor gene candidates in chromosomal deletions. Proc Natl Acad Sci U S A 109:8212–8217. https://doi.org/10.1073/pnas.1206062109
97. Davoli T et al (2013) Cumulative haploinsufficiency and triplosensitivity drive aneuploidy patterns and shape the cancer genome. Cell 155:948–962. https://doi.org/10.1016/j.cell.2013.10.011
98. Nijhawan D et al (2012) Cancer vulnerabilities unveiled by genomic loss. Cell 150:842–854. https://doi.org/10.1016/j.cell.2012.07.023
99. Muller FL et al (2012) Passenger deletions generate therapeutic vulnerabilities in cancer. Nature 488:337–342. https://doi.org/10.1038/nature11331
100. Davoli T, Uno H, Wooten EC, Elledge SJ (2017) Tumor aneuploidy correlates with markers of immune evasion and with reduced response to immunotherapy. Science 355. https://doi.org/10.1126/science.aaf8399