



## Review

## Cancer regeneration: Polyploid cells are the key drivers of tumor progression

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## ABSTRACT

In spite of significant advancements of therapies for initial eradication of cancers, tumor relapse remains a major challenge. It is for a long time known that polyploid malignant cells are a main source of resistance against chemotherapy and irradiation. However, therapeutic approaches targeting these cells have not been appropriately pursued which could partly be due to the shortage of knowledge on the molecular biology of cell polyploidy. On the other hand, there is a rising trend to appreciate polyploid/ multinucleated cells as key players in tissue regeneration. In this review, we suggest an analogy between the functions of polyploid cells in normal and malignant tissues and discuss the idea that cell polyploidy is an evolutionary conserved source of tissue regeneration also exploited by cancers as a survival factor. In addition, polyploid cells are highlighted as a promising therapeutic target to overcome drug resistance and relapse.

## 1. Introduction

A nearly 20% mortality rate has rendered cancer the most problematic health issue in the world. Despite huge efforts and budgets allocated to studies of cancer over the past century, efficient therapeutic approaches to inhibit cancer progression are still missing. One of the main bottlenecks in cancer treatment is tumor recurrence post chemo/radiotherapy, the exact mechanisms of which remains to be further elucidated. In agreement with the clonal expansion theory that describes the evolving feature of cancer [1], the cancer stem cell theory proposes that tumor growth, maintenance and drug resistance could be assigned to immortalized, self-renewing stem cells in the tumor [2,3]. Although, fate-mapping strategy has determined the presence of cancer stem cells in the tumors [4], little is known about the origin of these cells.

Although the first evidence on the presence of polyploid giant cancer cells dates back to almost two centuries ago [5], discoveries on their relationship to cancer stem cells have been accomplished in the recent decades [6,7]. Indeed, it is shown that polyploid giant cancer cells (PGCCs) could be assigned as oncogenerative units of the tumor which produce cancer stem cells via budding/bursting and are capable

of repopulating the whole tumor tissue in recurrence phase [8–10]. In agreement with this assumption, systematic analysis on histopathologic sections of cancerous lesions have reproducibly shown the presence of polyploid cells in more than 40% of tumors [11]. Also, the presence of polyploid cells is revealed to be positively correlated with grade of malignancy [12–16].

Polyploidy is not only a pathologic process restricted to cancers, but also a physiologic evolutionary conserved mechanism for the maintenance of tissue hemostasis [17]. Polyploidy is observed in the development of a wide range of eukaryotes from protozoans and fungi to mammals. On the other hand, polyploid cells are critical components of tissue regeneration and potential origins of tissue resident stem cells which exert a highly tolerable phenotype in response to stressors [18].

Based on the functional similarity of polyploid cells in development, tissue regeneration and cancer, it could be assumed that polyploidy is a beneficial cellular mechanism that is hijacked by cancer for adopting rapid growth and resistance to treatment. In this review, we propose an analogy between the role of polyploid cells in tissue regeneration and cancer progression. Also, cancer polyploid cells are underscored as a promising drug target. We start this argument with an overview on the history of the key findings in this field.

*Abbreviations:* PGCC, polyploid giant cancer cell; MSC, mesenchymal stem/ stromal cells; NAFLD, nonalcoholic fatty liver disease; NCI, National Cancer Institute; DBPQ, 2,3-diphenylbenzo[g]quinoxaline-5,10-dione; HPRT1, Hypoxanthine-guanine phosphoribosyltransferase 1

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## 2. Landmark investigations on PGCCs

In order to cover key findings in cancer polyploidy, we performed a search in PubMed for “polyploid giant cancer cells” OR “multinucleated giant cancer cells” followed by manual refinement which finally resulted in less than 100 articles related to cancer polyploidy.

The primary evidence on polyploid cancer cells was reported nearly two centuries ago when the cancer cells were characterized by Müller and his students [5]. His hand-drawn illustrations in the book “On the Finer Structure and Form of Morbid Tumors (Ueber den feinem Bau und die Formen der krankhaften Geschwülste)” indicate the pleomorphism of malignant cells [19]. Virchow, the father of modern pathology, defines cancer cells as “...curious bodies, provided with large nuclei and nucleoli” [20]. These findings were taken into consideration again in 1902 when the German biologist, Theodor Boveri, proposed whole genome doubling as a cellular process which is responsible for aneuploidy [21].

Considering the resemblance of polyploid cancer cells to normal multinucleated osteoclasts, several investigators referred to these cells as “osteoclast-like giant cells” [22–24]. Some other terms have also been used to describe these cells. Boyd et al. named these giant cancer cells as “pleomorphic” or “monster” cells whose presence significantly associates with histologic sub-type, ulceration and depth of invasion in melanoma [25]. These cells are now commonly referred to as Polyploid Giant Cancer cells (PGCC) [10].

The underlying mechanisms of the formation of polyploid cells have been investigated since more than fifty years ago. Although the nomenclature is still confusing [26], a variety of mechanisms are described (Box 1); the “endomitosis” firstly was described by two independent investigators in 1939 as a mechanism of genome content increment in insects [27,28]. In 1965, “endoreplication” and “cell-cell fusion” were proposed for generation of these cells by Nasjleti et al. and Okada et al. respectively [29,30]. Other terms such as “endocycling”, “cytokinesis failure”, and “entosis” were introduced as cancer polyploidy inducers in the consecutive years [31–34].

The potential inducers of polyploidy have also been the matter of investigation. Bell and Baker found leukocytes polyploidy upon exposure to x-irradiation [35]. Also, drugs such as cytochalasin, glutaraldehyde and bistramide were determined as polyploidy inducers in HL60 leukemic cells and non-small-cell bronchial carcinoma line [36–38]. Now, by application of high-throughput analysis, it is clarified that a broad range of cancer therapeutics stimulate formation of polyploid cells post administration in patients [39].

Cancer polyploid cells are known to be related to cancer progression and recurrence. In 1968, Bauke & Schöffling for the first time reported correlation between cancer progression and augmentation of polyploid cells in a 73-year old man with reticulosis malignancy [40]. Twenty years later, DNA measurements in 409 breast cancer patients indicated poor prognosis in tumors with increased DNA content [41]. Also, polyploidy was introduced as a prognostic marker of five-year survival in adenocarcinoma patients [42]. As well, correlation between incidence of polyploidy and oncogene amplification was found in gastric cancer patients [43]. Studies on colorectal cancer have indicated that tumors harboring tetraploid cells have a worse prognosis than their diploid counterparts [44]. In agreement, in studies by Dewhurst et al., a hazard ratio of 4.7 is reported for genome-doubling association with tumor relapse in colorectal cancer patients [14,45]. Moreover, the proportion of binucleated to multinucleated cells is suggested as a parameter for tumor stratification in breast cancer patients [46].

PGCCs can undergo depolyploidization through which mono-nucleated cells are formed [47]. Although since about one hundred years ago different terms such as amitosis [48,49], nuclear fragmentation [50,51], nuclear budding [52], and neosis [53] have been used to refer to the emergence of daughter cells from a polyploid/multinucleated cell, it has not received enough attention by cancer biologists until recent years. In 2000, Erenpreisa et al. observed budding of mono-nucleated descendants from P53-deficient giant tumor cells [54]. Based

on observations on cultured amniocytes and epithelial cells, Walen demonstrated the emergence of multinucleated cells through endoreplication in the presence or absence of SV40 [55,56]. These cells then originate mononucleated cells through amitosis [56]. In 2004, Rajaraman and colleagues could capture the process of depolyploidization by applying video time-lapse microscopy and named the mononucleated progenies “Raju cells” [57]. They further proposed stem cell properties and cancer-related transformation to these cells [53]. Ten years later, Zhang et al. determined that PGCCs can originate different lineages and a single polyploid cell could be the source of the whole tumor tissue [6,58]. In agreement, using time-lapse fluorescent imaging Niu et al. demonstrated similarities between blastomere development and the emergence of different lineages from PGCCs [9,59]. Based on these findings, Liu proposed a unifying theory nominated as “life code” explaining that ploidy augmentation is a key phenomenon both in embryogenesis and formation of undifferentiated and well-differentiated tumors [60,61]. The timeline of main discoveries on PGCCs is represented in Fig. 1.

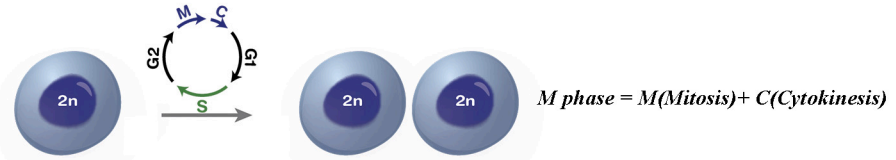
## 3. Polyploidy: a conserved basic mechanism in tissue regeneration and tumor recurrence

The beneficial aspects of polyploidy in tissue hemostasis and normal development have led to its selection over the phylogenetic tree from the simplest protozoans to multi-cellular organisms [17,62–64]. The protozoan parasite, *Entamoeba histolytica* has an oscillating life cycle between a proliferating trophozoite and a dormant cyst. It is shown that rapid proliferation of *Entamoeba* coincides with accumulation of polyploid cells up to 8 DNA contents [65]. Peculiarly, in the larvae of *Drosophila melanogaster*, approximately all tissues become polyploid through endocycling which is supposed to be mechanism of rapid growth [66]. As well, normal growth of nematode *Caenorhabditis elegans* with a fixed number of somatic cells (959) happens through augmentation of cell ploidy through endoreplication [67]. In mammalian embryos, the trophoblast giant cells are critical polyploid cells that produce a wide spectrum of cytokines and hormones [68,69]. Similarly, studies on human in vitro fertilized eggs have demonstrated 60% polyploidy in formed blastomeres [70]. Furthermore, mammalian hepatocytes, megakaryocytes, cardiomyocytes, lactating mammary cells, skin epithelial cells and osteoclasts are among the cells that polyploidy is critical for their normal development [71–74].

Not only normal growth and development, but also tissue regeneration takes advantage of programmed polyploidization. Polyploidy serves as a strategic approach for compensating cell loss post injuries which restores tissue integrity in terminally differentiated organs [75]. In follicular epithelial cells of *drosophila*, response to elimination of cells post injury happens through compensatory cellular hypertrophy and polyploidy [76]. Similarly, the injured skin epithelial cells and hindgut pylorus of *drosophila* become polyploid via endocycling or cell fusion post injury [77,78]. Also, Cao et al. found that mechanical tension in the epicardial tissue of zebrafish leads to formation of zones of multinucleated cells with regeneration capability [79]. Likewise, Pax8 positive kidney tubular epithelial cells become polyploid in response to acute kidney injury [80]. In addition, a closer look at early mouse MSC cultures established as explants from decapsulated glomeruli revealed a number of binucleated cells within nearly a week from plating (Supplementary Fig. 1). More than 70% of hepatocytes in rodents and near 50% in humans are polyploid [81,82]. Despite some contradictory findings [83], there is a bundle of evidence demonstrating that polyploidy increases upon liver injury and is a promoter of liver regeneration [84–86]. The number of polyploid cells increases in response to hepatectomy [87]. Also, these cells facilitate regeneration in chronic liver injury induced by tyrosinemia [88]. A recent study reveals that polyploid hepatocytes go through reductive division and subsequent polyploidization to regenerate the injured liver [89]. The increased number of chromosomes in the polyploid

**Box 1**  
Glossary.

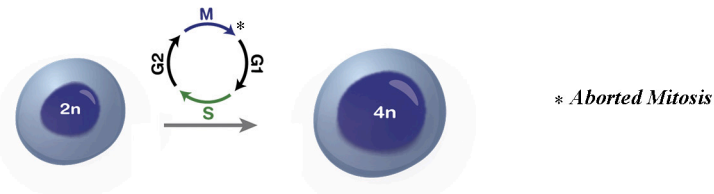
**Diploid cell cycle:** Consists of G1, S, G2 and mitotic phase finishing with cytokinesis that leads to production of diploid daughter cells.



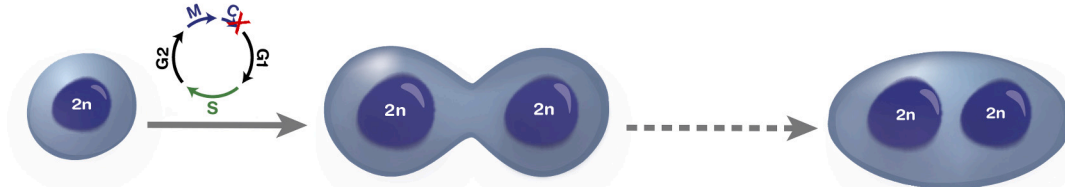
**Ploidy:** A state in which cells have more than diploid DNA content. Different mechanisms lead to ploidy:

● **Endoreplication:** Also known as endoreduplication, is a general term that refers to an incomplete cell cycle process leading to production of polyploid cells. Endomitosis, cytokinesis failure, and endocycling are three mechanisms leading to endoreplication.

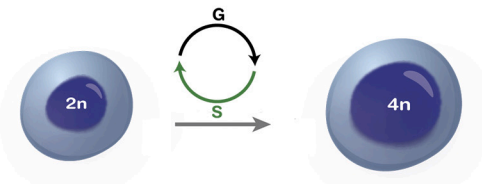
- **Endomitosis:** An initiated cell cycle which is aborted in the mitosis step which leads to formation of a cell with one polyploid nucleus.



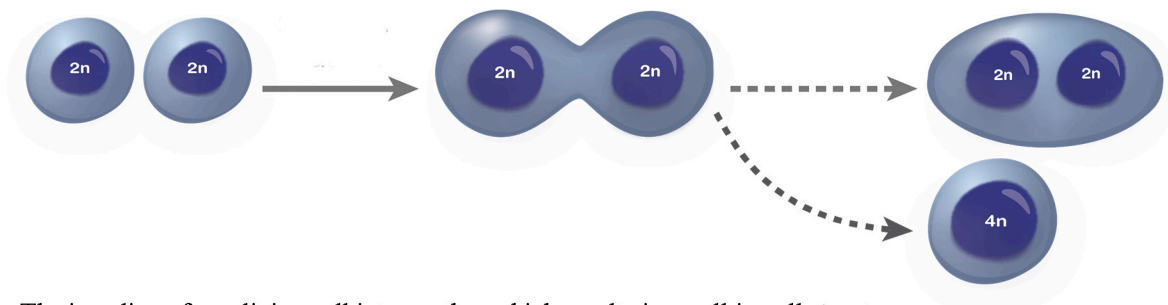
- **Cytokinesis failure:** Although mitosis is successfully finished and nuclei are formed, the cleavage furrow is aborted and daughter cells do not separate, leading to formation of a bi/multi-nucleated cell.



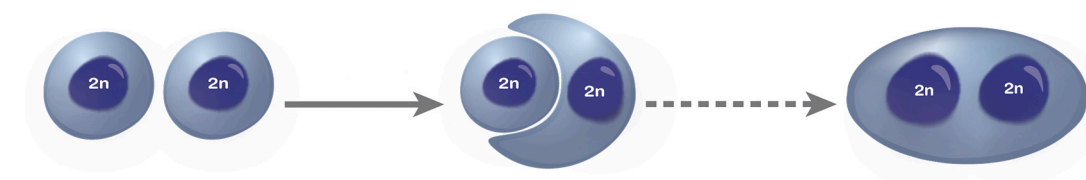
- **Endocycling:** The cell completely bypasses mitotic step and sequential cycles of G and S happens resulting in a nucleus with multiple copies of the genome. The polytene chromosomes in drosophila salivary gland are formed through this mechanism.



● **Cell-cell fusion:** Happens when two cells merge through membrane specific fusion proteins.



● **Entosis:** The invading of one living cell into another which results in a cell in cell structure.



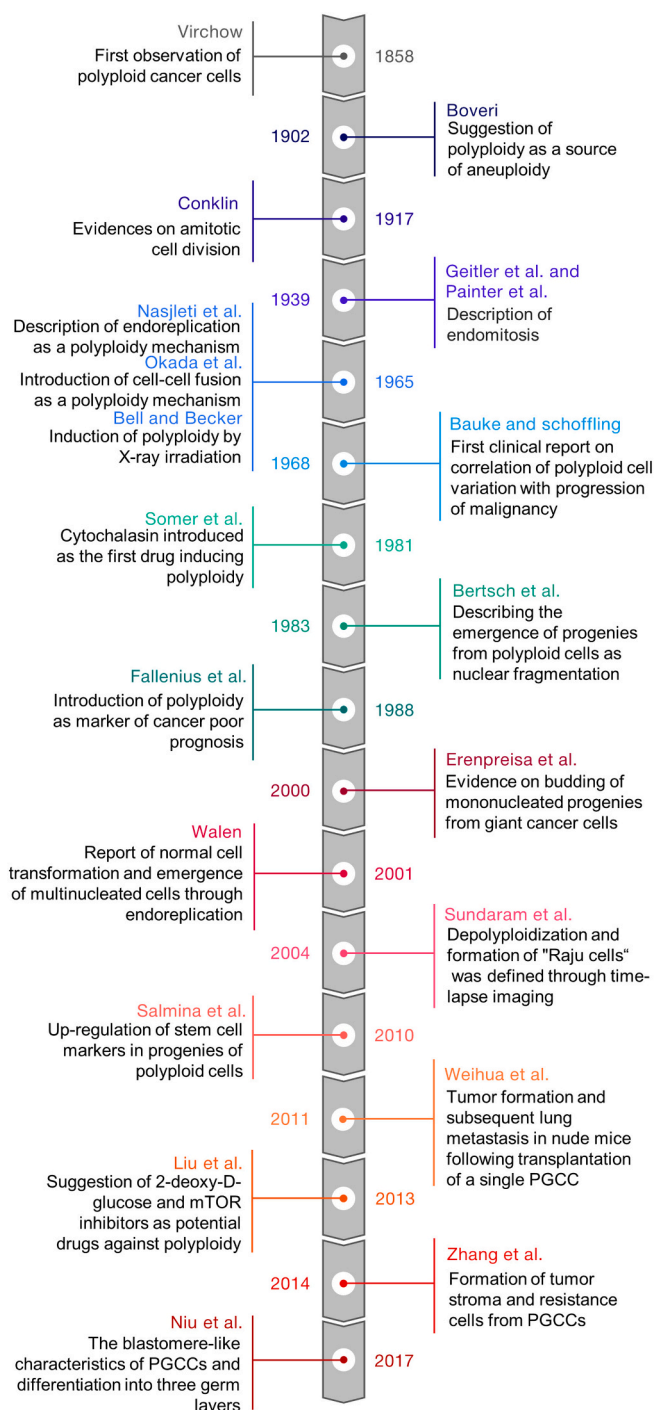


Fig. 1. Timeline of landmark discoveries in cancer polyploidy.

hepatocytes buffers against tumorigenesis caused by loss of heterozygosity of tumor suppressor genes [90]. The above-discussed findings on the role of polyploidy on tissue regeneration indicate that it is much more common in physiological states than one may assume. Yet, the mechanisms making such huge chromosomal alternations tolerable remain to be elucidated.

On the other side, polyploidy is shown to be increased with aging in tissues such as heart, eye, brain, and liver and is correlated with occurrence of some of the age-associated diseases like atherosclerosis [91], Fuchs' endothelial corneal dystrophy [92,93], nonalcoholic fatty liver disease (NAFLD) [94], and undoubtedly various neoplasms [64]. Malignant tissues as autonomous units in the body, utilize normal cell

mechanisms to maintain their function. Similarly, polyploidy is exploited in the context of cancer to entail requirements of the tumor lesion such as angiogenesis, cancer stem and stromal cell production, genomic instability and drug-resistance all of which promote tumor evolution and relapse [6,58,95,96]. Investigating the role of polyploid cells in tissue regeneration and cancer, taken us to the concept that polyploid cells behave similarly in both paradigms. In each of the four following sections, similar aspects of polyploid cells functions in tissue regeneration and malignancies are comparatively discussed (Fig. 2).

### 3.1. Polyploid cells are a potential source of stem cells

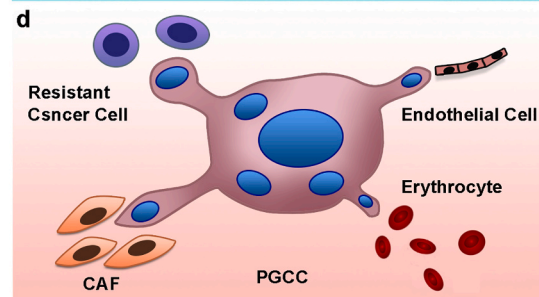
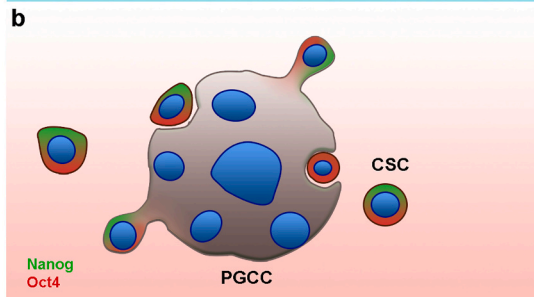
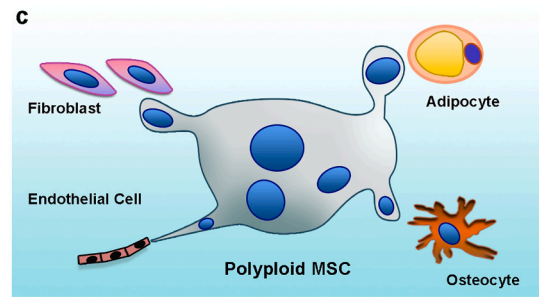
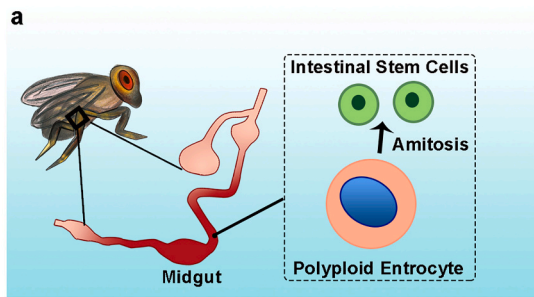
Polyploid cells are demonstrated to be potential origins of stem cells in various organisms; the intestine of *Drosophila melanogaster* contains polyploid enterocytes (8 to 32C) that are formed through endocycling of undifferentiated enteroblasts and are critical for epithelial absorption [97]. The study of Lucchetta and Ohlstein determined that upon loss of intestinal stem cells post food starvation, these polyploid enterocytes go through amitosis and originate tissue specific stem cells [98]. Likewise, it is recently found that post myocardial infarction, tetraploid cardiac cells go through reductive division and produce diploid cardiac interstitial cells in the murine heart [99]. In the liver, nearly 50% of the cells are polyploid that markedly increase upon hepatectomy, cytotoxic injury or aging [81]. Although previously it was believed that polyploid hepatocytes are terminally differentiated, recent evidence shows that they are capable of undergoing reductive division through which mononucleated hepatocytes are produced [100]. Furthermore, our studies on cultured bone marrow cells revealed a transient dormant phase during which large multinucleated cells are formed that could be the origin of mono-nucleated mesenchymal stem/ stromal cells (MSCs) [101–103]. Also, our time-lapse imaging of these cells represents vast cytoplasmic contractions that leads to budding of stem cells or a bursting phenomenon through which the polyploid cell is broken into mono-nucleated cells (Supplementary Fig. 2).

Alike to the ability of polyploid cells in production of progenies involved in tissue hemostasis and regeneration, PGCCs are capable of generating mononucleated cells through the process of neosis described by Rajaraman et al. [53]. Studies show that post chemo/radio therapy administration, most of the cells die, but a small proportion of survived cells enter a lag phase with senescent and multinucleated phenotype which subsequently become reactivated and produce mononucleated cells [104,105]. In 2008, it was determined that cisplatin-induced polyploid cells generate small para-diploid cells after a long dormant phase [106]. Furthermore, polyploid cells have been described as "pregnant cells" capable of parturition of progenies via crevices- or funiculus-like structures [8]. Importantly, in 2010, Salmina et al. have proposed stemness properties to the progenies of cancer polyploid cells [107], the idea which was further validated by next studies based on functional assays as well as the expression of stem cell markers such as OCT4, NANOG, SOX2 and SSEA1 [6,8,9,107,108]. Proteomic analysis has also determined up-regulation of stem cell regulating factors in breast cancer polyploid cells [109]. Moreover, it is shown that transplantation of PGCCs into immunodeficient mice causes formation of tumors positive for CD133 and CD44 which are prominent stem cell markers [10].

### 3.2. A variety of cell types emerge from polyploid cells

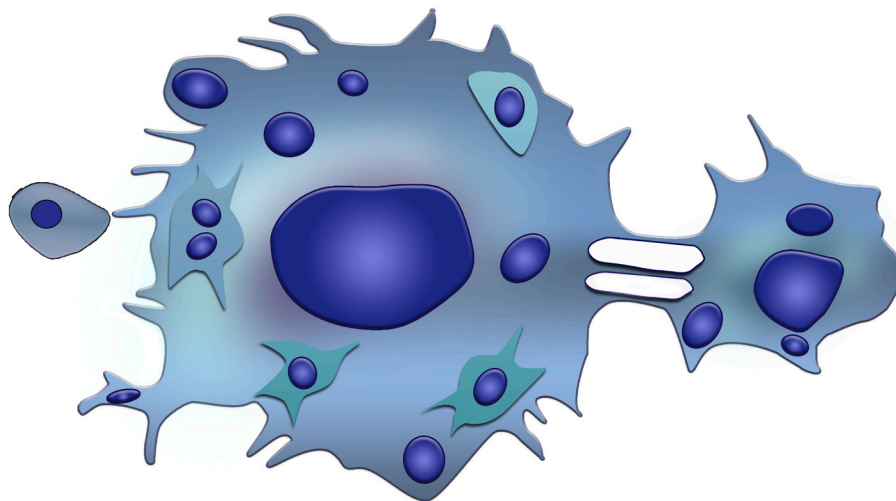
Polyploid cells with high genomic content and considerable epigenomic alterations are capable of spawning a variety of cells. Hence, they could be assumed as factories assembling and producing multiple celltypes of tissues. In agreement with this assumption, we have observed that polyploid cells in cultures of mouse bone marrow are the origin of MSCs with three lineage differentiation capacity [103]. Also, we have provided preliminary evidence that these cells could be the origin of endothelial-like CD31 and VEGFR positive cells [110].



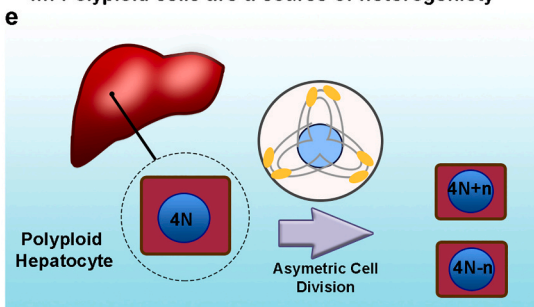


**I. Polyloid cells are a potential source of stem cells**

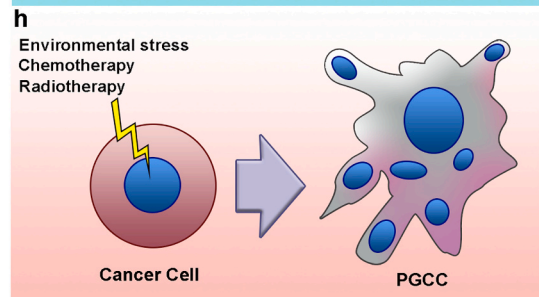
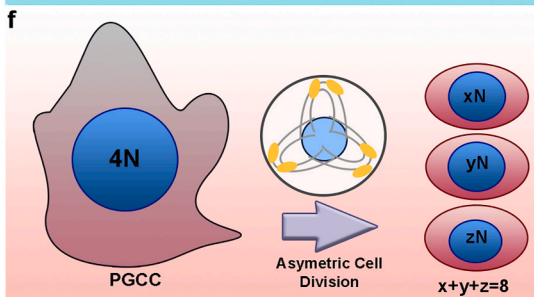
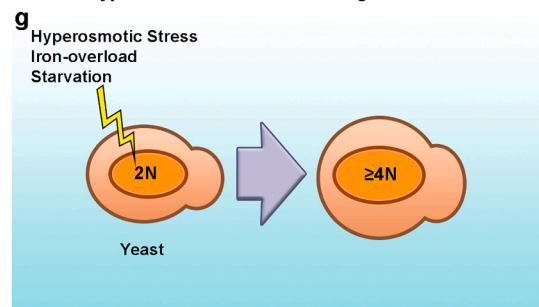
**II. A variety of cell types emerge from polyloid cells**



**III. Polyloid cells are a source of heterogeneity**



**IV. Polyloid cells are resistant against stressors**



(caption on next page)

**Fig. 2.** Polyploid cells have similar functions in tissue regeneration and tumor progression. In each panel, the top image schematically represents a role of these cells in normal tissue regeneration and the bottom one depicts a similar performance in malignant tissues. a) In *Drosophila* midgut, post ablation of stem cells, polyploid enterocytes go through amitosis to produce intestinal stem cells. b) PGCCs can produce stem cells expressing cancer stem cell (CSC) markers. c) Cultured MSCs go through a multinucleation phase upon which produce cell types such as chondrocytes, adipocytes, osteocytes, endothelial and fibroblasts. d) PGCCs remaining after chemotherapy not only generate resistant cancer cells, but also tumor vasculature, erythrocytes and cancer associated fibroblasts (CAF). e) Polyploid hepatocytes have multiple pairs of centrosomes and multi-polar spindles resulting in aneuploid daughter hepatocytes. f) PGCCs are shown to be the main source of aneuploid cancer cells. g) Yeasts tolerate environmental insults through acquiring a polyploid phenotype. h) Cancer cells acquire a resistant polyploid phenotype in response to both environmental stresses and therapeutics.

Accordingly, it is demonstrated that embryos in the blastomere stage contain polyploid cells which their presence is necessary for embryonic organ development [70,111].

A growing body of evidence highlights the concept that PGCCs are the origin of different types of tumor parenchymal and stromal cells. It is shown that a single polyploid cancer cell is capable of spheroid formation and multilineage differentiation [6,9]. In agreement, study by Zhang et al. showed that PGCCs give rise to multicellular structures mimicking the whole tumor tissue termed as “cancer-organotypic structures” [58]. Not only CD31+ endothelial, but also erythroid cells with active fetal hemoglobin are produced in the vessel-like assemblies of these structures [58,112]. This finding is further validated by other investigators which determined contribution of PGCCs in erythrocyte production as well as formation of vasculogenic mimicry and mosaic vessels in the tumor tissue [113]. Expression of macrophage, neutrophil, fibroblast and myoepithelial surface markers are demonstrated in PGCCs or their progenies [58,114–116]. Also, transdifferentiation capability of PGCCs into skeletal muscle fibers and adipocytes is determined by histomorphologic assessments [115]. Notably, the first evidence on potential of PGCCs to form a variety of cell lineages is provided by Weihua et al. in 2011, showing the generation of all tumor elements post single PGCC transplantation into nude mice [117].

### 3.3. Polyploid cells are a source of heterogeneity

Aneuploidy is commonly considered a pathogenic phenotype. However, this general belief may not always be true; around 10% of brain neural cells become aneuploid during normal development. This process allows generation of a variety of cell types to cover wide spectrum of functions of the neural system [118]. Also, a considerable fraction of mammalian liver cells is aneuploid which is a source of heterogeneity and adaptive phenotype in both normal and disease conditions [62]. Polyploid cells are a critical source for the appearance of these aneuploid cells [100]; in this paradigm, polyploid hepatocytes have an increased number of centrosomes which lead to formation of multiple spindle assemblies during anaphase. As attachment of microtubules to kinetochores is random, multiple microtubules from different poles could bind to one chromosome resulting in merotelic chromosomes that fail to orient over distinct poles and subsequently formation of aneuploid cells [86]. We have observed that multinucleated cells in the culture of normal bone marrow, generate MSCs which are mostly near-tetraploid but not tumorigenic post transplantation into nude mice [103]. In agreement, while studying MSCs from various murine tissues, our group found that most MSC cultures develop a tetraploid or near-tetraploid DNA content after several passages [119]. This finding is linked to the hypothesized behavior of these cells during wound healing in vivo, as tetraploid cells have been observed in wound-healing of rodents and human [120,121].

Aneuploidy is a well-known feature of malignancies [63]. It is shown that in Barrett's esophagus tetraploid cells are preliminaries into formation of aneuploid cells [122,123]. Likewise, whole-genome doubling is an early event in glioblastoma and associates with chromosomal instability (CIN) and aneuploidy post treatment [124]. Hence, polyploid cells are a main source of aneuploid cancer cells [14,15,125,126]. It is demonstrated that PGCCs have multiple sets of centrosomes and unusual spindle structures which subsequently lead to aneuploid

offsprings during reductive divisions [127]. Beside chromosome mis-segregation during depolyploidization which results in aneuploidy, instability in the genome content is a hallmark of PGCCs. This feature speeds up mutational changes and karyotypic variations needed for cancer cell surveillance [11,14]. Vast chromosomal rearrangements in one or a few of the chromosomes, known as chromothripsis, is one of the outcomes of CIN in PGCCs [128]. There is a line of evidence highlighting a positive correlation between polyploidy and chromothripsis in different malignancies [129–131]. Genomic changes resulting from polyploidization/depolyploidization are a cost-beneficial strategy for buffering against deleterious mutations and also a source of high heterogeneity which allows malignant cells to evolve in response to selection pressure [127,132].

### 3.4. Polyploid cells are a source of resistance against stressors

The relationship between increased genome content and fitness advantage is seen in a variety of living organisms. Yeast sub-species are shown to go through polyploidization in response to hyperosmotic, metal and starvation stresses [133–135] to augment gene expression capacity and tolerate loss of function mutations [136]. In plants, polyploidy not only results in tolerance to environmental stressors and adaptive responses, but also more product yield [137]. Also, to preserve liver function, mammalian hepatocytes take advantage of polyploidization when face with stressors such as aging, hepatectomy, iron-overload, and toxins [138–140].

Tumors are naturally exposed to different kinds of environmental stresses such as hypoxia, mechanical tension, and viruses. Polyploidy is an adaptation mechanism that allows cancers to tolerate such conditions. Initial clues on the role of hypoxia in formation of PGCCs dates back to 1996 in a study by Rofstad et al. on a human melanoma cell line [141]. This finding was later reproduced in different cancer cell lines using either hypoxic culture conditions or chemical inducers of hypoxia [6,7]. Post CoCl<sub>2</sub> treatment as a mimic of hypoxia, HIF1 $\alpha$  (hypoxia-inducible transcription factor 1 $\alpha$ ) is stabilized and overexpressed in PGCCs which through collaboration with Notch signaling promotes stem cell-like phenotype and tumor development [6,105,142]. Furthermore, lactic acid as a metabolite of tumor hypoxic milieu is shown to induce tetraploidization [143]. More recently, Arun et al. simulated physical pressures of cancer microenvironment and found that cancer cells undergo endoreplication [144]. In addition, human papilloma virus E6 and E7 proteins augment polyploidy and aneuploidy in cancer cell lines mediated through p53 and pRb inactivation [145] or Plk1, Aurora-A, Nek2, and cdk1 modulation [146].

## 4. Polyploidy: a major challenge in fight against cancer

Through above discussed mechanisms polyploidy allows cancers to resist against a wide range of stresses including most available chemotherapy and radiotherapy regimens. The polyploidy/aneuploidy reversion in response to therapy was first proposed in 1960 [147] and since then, several investigators have shown formation of polyploid cells upon therapy administration. The genotoxic insults raised by chemo- and radiotherapies cause cell cycle arrest, mitotic catastrophe and death of most of the cancerous cells. Nevertheless, a small fraction of surviving cells acquire a polyploid phenotype that following a

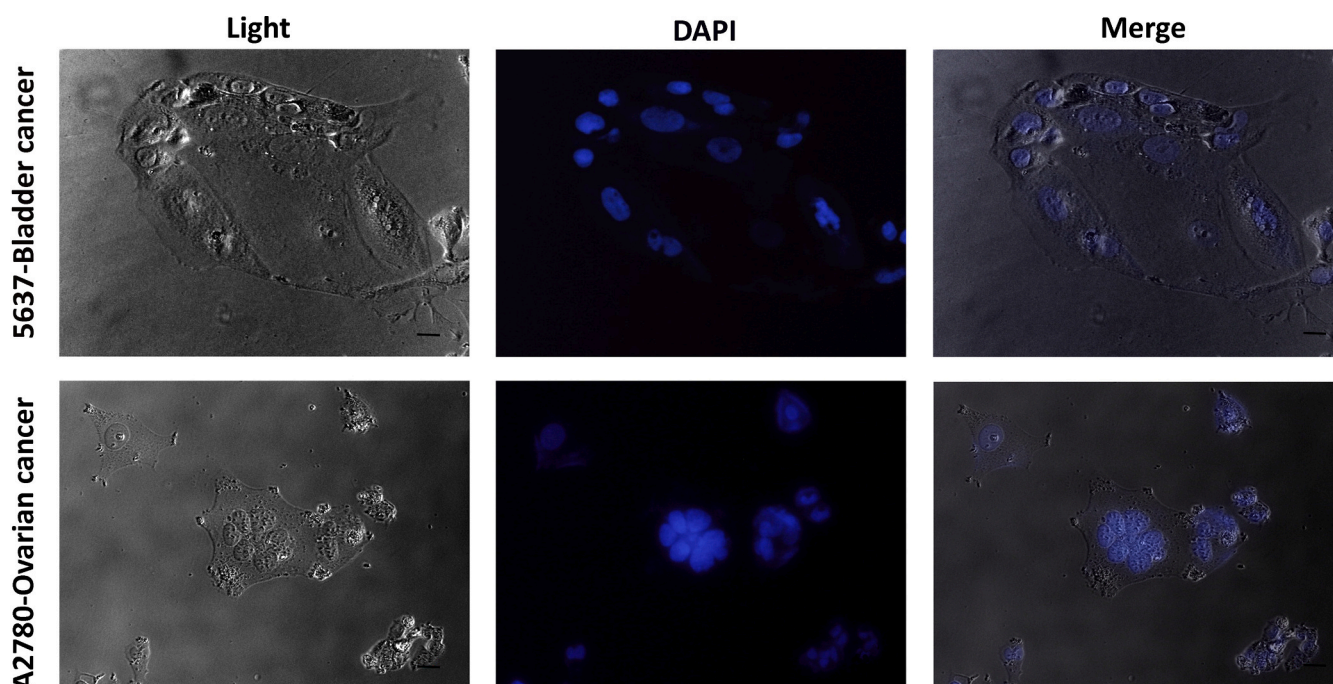


Fig. 3. Multinucleated cells enrich in 5637 bladder cancer and A2780 ovarian cancer cell lines post exposure to cisplatin. Both cell types were treated with 6  $\mu$ M cisplatin for 72 h followed by two weeks recovery. Scale bars: 50  $\mu$ m.

dormant phase produce heterogenic resistant progenies whenever favorable conditions are available [148].

Cisplatin, as a chemotherapy agent, targets DNA and makes double-strand breaks. In line with other investigators [106,149], our unpublished data indicates formation of polyploid cells in response to cisplatin treatment (Fig. 3). Also, polyploidy is observed in response to administration of purine/pyrimidine antagonists, 5-fluoro uracil and reversine in different cancer cell lines [150–153]. Studies show that multinucleation and spawning of mono-nucleated resistant stem cells happen post treatment with paclitaxel and docetaxel that are microtubule-stabilizing agents [154,155]. As well, vincristine, doxorubicin, and bortezomib are shown to induce polyploidy [156,157]. Cell cycle kinases like Aurora and Polo-like kinase which are vastly over-expressed in high-grade malignancies are the other cancer drug targets whose inhibition has been repeatedly shown to be correlated with genomic instability and formation of polyploid cells [158–160].

Evidence on the role of radiation in induction of cell polyploidy was firstly described by Bell and Baker [35]. The double-strand breaks induced by ionizing radiation lead to G2/M arrest in cancer cells. Although most of the arrested cells die through activation of DNA repair mechanisms, a small proportion of cells survive, become polyploid and serve as the source of new cancer cells [161]. Notably, it is shown that radiation exposure of non-tumorigenic breast cancer cells, causes reprogramming of differentiated cells into “induced breast cancer stem cells” with polyploid and resistance characteristics [162]. Despite convincing evidence on PGCC formation upon environmental and therapeutic stressors, no targeted treatment strategy is clinically validated yet to combat against this smart choice of the tumors. A few drugs are shown to partially target cancer polyploid cells in preclinical settings which are reviewed in the next section.

## 5. Struggling with PGCCs

Most of the current cancer therapies target rapidly dividing mono-nucleated cells in order to reduce the bulk of tumor. Nevertheless, the survived polyploid cells, which due to their resistant phenotype and capability of regenerating the whole tumor tissue can be assumed as “roots of cancer”, have been considered as therapeutic targets just in a

Table 1

Chemicals efficient for the inhibition of PGCCs.

Drug	Function	Reference
2-Deoxy-d-glucose	Glycolysis inhibitor	[163,164]
Rapamycin	mTOR inhibitor	[163]
AZD8055	mTOR inhibitor	[165]
ABT-263	Apoptosis induction through Bcl-xL Inhibition	[166]
Resveratrol	Inhibition of cell cycle progression	[167]
Salicylate	Apoptosis induction through AMPK pathway	[167]
Trolox	Targeting reactive oxygen species	[168]
DPBQ	HIF1a- and p53-dependent apoptosis	[39]
8-Azaguanine	Antimetabolite produced by HPRT1	[39]
Belinostat	HDAC inhibitor	[169]
Isoamapimod	MAPK inhibitor	[170]
Flavopiridol	CDK inhibitor	[171]
Griseofulvin	Centrosome-declustering agent	[172]

few investigations (Table 1).

Polyploid cells shift to more cost-effective metabolic pathways to survive in the extreme microenvironment of cancer lesions. In both acute myeloid leukemia and brain tumors, polyploid cells are shown to be more sensitive to the toxic effects of 2-deoxy-d-glucose, as an inhibitor of glycolysis, compared to euploid cancer cells [163,164]. mTOR which is an important regulator of cell cycle and metabolism in megakaryocytes is another proposed drug target [173]. Rapamycin, the inhibitor of mTOR, promotes apoptosis and autophagy of polyploid leukemic cells [163]. Similarly, in the study of Sharma et al. reduced formation of breast cancer polyploid cells was demonstrated in the presence of mTOR inhibitor AZD8055 [165].

As polyploid cells exert antiapoptotic effects through upregulation of pro-survival components, this feature can also be considered as an opportunity for targeting polyploid cells. Zhou et al. demonstrated that inhibition of antiapoptotic protein Bcl-xL by ABT-263 small molecule causes lethality of polyploid cells [166]. Lissa et al. have performed a pharmacological screening on an established tetraploid HCT116 cancer cell line and found that resveratrol and salicylate reduce polyploid cell survival by inhibition of cell cycle progression as well as apoptosis induction through AMPK pathway [167]. Targeting the high rate of



reactive oxygen species seen in cancer polyploid cells by Trolox, the analog of vitamin E, has also been beneficial in eradication of doxorubicin-induced HCT 116 polyploid cells [168].

The unknown and complex nature of polyploid cells is a main barrier for drug target identification which could be addressed by application of high throughput methodologies. Employing the big data from National Cancer Institute (NCI) developmental therapeutic program on the sensitivity of 45'342 chemicals on 60 cancer cell lines adjunct to the polyploidy state of these cell lines, Choudhary et al. discovered 8-azaguanine and 2,3-diphenylbenzo[g]quinoxaline-5,10-dione (DPBQ) as specific inhibitors of polyploid cells. DPBQ acts through HIF1 $\alpha$ - and p53-dependent augmentation of oxidative stress and apoptosis. Also, 8-azaguanine is an antimetabolite produced by Hypoxanthine-guanine phosphoribosyltransferase1 (HPRT1) enzyme [39].

Altogether, PGCCs cannot efficiently be triggered by currently available anti-mitotic cancer therapeutics. Not only the historical ignorance of polyploidy in cancers and consequently insufficient knowledge on the underlying molecular mechanisms, but also the limitations of cancer drug development pipelines have led to the scarcity of PGCC-targeting therapies. Indeed, current preclinical assays employed to assess the efficacy of candidate drugs are based on short term outcomes without continues follow-ups, which hinders getting insight into resistant polyploid cells [174]. In this regard, we propose further investigations focused on regulatory circuits of cell polyploidy as well as improvement of in vitro drug screening systems in order to develop PGCC-targeting agents to be administrated in combination with current available therapeutics.

## 6. Conclusion

Polyploidy, a fundamental property for organism development and tissue regeneration, is exploited by cancers as a cost-beneficial surviving strategy. Formation of heterogenic resistant cancer stem cells and tumor tissue stromal cells from PGCCs would give rise to a more aggressive counter-attack post cancer treatment. Despite near two centuries background of observing cancer polyploid cells and their outstanding effects on tumor resistance and relapse, they are not yet effectively targeted by current anti-cancer therapeutics. To design more efficient therapeutics, it is critical to investigate the molecular biology aspects of these cells such as their gene regulatory circuits and metabolic machinery, mechanisms of DNA propagation and segregation, and intercellular communication routs. The emergence of advanced techniques such as in vivo fate-mapping, single-cell imaging and expression profiling as well as quantitative and holistic approaches of systems biology raise hopes for a deeper understanding of these mysterious cells.

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## Author contributions

Y.G developed the main conceptual idea of the manuscript. S.M and R.A performed literature survey and designed figures. S.M drafted the work. L.S.M and N.B.N aided in preparing experimental evidence and improving content of the manuscript. Y.G and N.B.N critically revised the manuscript. All authors made a substantial intellectual contribution to the work, and approved the submitted version for publication.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## References

- [1] P.C. Nowell, The clonal evolution of tumor cell populations, *Science* (New York, N.Y.) 194 (1976) 23–28.
- [2] T. Lapidot, C. Sirard, J. Vormoor, B. Murdoch, T. Hoang, J. Caceres-Cortes, M. Minden, B. Paterson, M.A. Caligiuri, J.E. Dick, A cell initiating human acute myeloid leukaemia after transplantation into SCID mice, *Nature* 367 (1994) 645–648.
- [3] A.Z. Ayob, T.S. Ramasamy, Cancer stem cells as key drivers of tumour progression, *J. Biomed. Sci.* 25 (2018) 20.
- [4] E. Batlle, H. Clevers, Cancer stem cells revisited, *Nat. Med.* 23 (2017) 1124–1134.
- [5] S.R. Amend, G. Torga, K.C. Lin, L.G. Kostecka, A. de Marzo, R.H. Austin, K.J. Pienta, Polyploid giant cancer cells: unrecognized actuators of tumorigenesis, metastasis, and resistance, *Prostate* 79 (2019) 1489–1497.
- [6] S. Zhang, I. Mercado-Urbe, Z. Xing, B. Sun, J. Kuang, J. Liu, Generation of cancer stem-like cells through the formation of polyploid giant cancer cells, *Oncogene* 33 (2014) 116–128.
- [7] L.M. Lopez-Sanchez, C. Jimenez, A. Valverde, V. Hernandez, J. Penarando, A. Martinez, C. Lopez-Pedraza, J.R. Munoz-Castaneda, J.R. De la Haba-Rodriguez, E. Aranda, A. Rodriguez-Ariza, CoCl<sub>2</sub>, a mimic of hypoxia, induces formation of polyploid giant cells with stem characteristics in colon cancer, *PLoS One* 9 (2014) e99143.
- [8] D. Diaz-Carballo, S. Saka, J. Klein, T. Rennkamp, A.H. Acikelli, S. Malak, H. Jastrow, G. Wennemuth, C. Tempfer, I. Schmitz, A. Tannapfel, D. Strumberg, A distinct oncogenetic multinucleated cancer cell serves as a source of stemness and tumor heterogeneity, *Cancer Res.* 78 (2018) 2318–2331.
- [9] N. Niu, I. Mercado-Urbe, J. Liu, Dedifferentiation into blastomere-like cancer stem cells via formation of polyploid giant cancer cells, *Oncogene* 36 (2017) 4887–4900.
- [10] J. Liu, I. Mercado-Urbe, N. Niu, B. Sun, J. Kuang, S. Zhang, Re-thinking the concept of cancer stem cells: Polyploid giant cancer cells as mother cancer stem cells. [abstract], *Proceedings of the 105th Annual Meeting of the American Association for Cancer Research*; 2014 Apr 5-9; San Diego, CA, AACR, Philadelphia (PA), 2014(Cancer Res 2014;74(19 Suppl):Abstract nr 1917).
- [11] T.I. Zack, S.E. Schumacher, S.L. Carter, A.D. Cherniack, G. Saksena, B. Tabak, M.S. Lawrence, C.Z. Zhsng, J. Wala, C.H. Mermel, C. Sougnez, S.B. Gabriel, B. Hernandez, H. Shen, P.W. Laird, G. Getz, M. Meyerson, R. Beroukhim, Pan-cancer patterns of somatic copy number alteration, *Nat. Genet.* 45 (2013) 1134–1140.
- [12] C.W. Rand, C.B. Courville, Multinucleation of cortical nerve cells at the margins of traumatic lesions of the human brain, *J. Neuropathol. Exp. Neurol.* 6 (1947) 1–14.
- [13] L.J. Rather, A note on the origin of multinucleated giant cells from vascular channels in tumors; tumors arising in thyroid gland, bone, and soft tissue, *Archiv. Pathol.* 52 (1951) 98–103.
- [14] S.M. Dewhurst, N. McGranahan, R.A. Burrell, A.J. Rowan, E. Gronroos, D. Endesfelder, T. Joshi, D. Mouradov, P. Gibbs, R.L. Ward, N.J. Hawkins, Z. Szallasi, O.M. Sieber, C. Swanton, Tolerance of whole-genome doubling propagates chromosomal instability and accelerates cancer genome evolution, *Cancer Discov.* 4 (2014) 175–185.
- [15] G. Lundberg, Y. Jin, D. Sehic, I. Ora, R. Versteeg, D. Gisselsson, Intratumour diversity of chromosome copy numbers in neuroblastoma mediated by on-going chromosome loss from a polyploid state, *PLoS One* 8 (2013) e59268.
- [16] E. Kaur, J.S. Goda, A. Ghorai, S. Salunkhe, P. Shetty, A.V. Moiyadi, E. Sridhar, A. Mahajan, R. Jalali, S. Dutt, Molecular features unique to glioblastoma radiation resistant residual cells may affect patient outcome - a short report, *Cell. Oncol. (Dordrecht)* 42 (2019) 107–116.
- [17] Y. Van de Peer, E. Mizrahi, K. Marchal, The evolutionary significance of polyploidy, *Nat. Rev. Genet.* 18 (2017) 411–424.
- [18] O.V. Anatskaya, A.E. Vinogradov, Somatic polyploidy promotes cell function under stress and energy depletion: evidence from tissue-specific mammal transcriptome, *Funct. Integrat. Genom.* 10 (2010) 433–446.
- [19] J. Müller, Über den feineren Bau und die Formen der krankhaften Geschwülste, G. Reimer, Berlin, 1838.
- [20] R. Virchow, Cellular Pathology as Based Upon Physiological and Pathological Histology, JB Lippincott, Place Published, 1863.
- [21] T. Boveri, Über mehrpolige Mitosen als Mittle zur Analyse des Zellkerns, *Verhandl Phys-med Ges (Wulzburg)* NF 35 (1902) 67–90.
- [22] U. Anazawa, H. Hanaoka, T. Shiraishi, H. Morioka, T. Morii, Y. Toyama, Similarities between giant cell tumor of bone, giant cell tumor of tendon sheath, and pigmented villonodular synovitis concerning ultrastructural cytochemical features of multinucleated giant cells and mononuclear stromal cells, *Ultrastruct. Pathol.* 30 (2006) 151–158.
- [23] N.A. Athanasou, C.A. Wells, J. Quinn, D.P. Ferguson, A. Heryet, J.O. McGee, The origin and nature of stromal osteoclast-like multinucleated giant cells in breast carcinoma: implications for tumour osteolysis and macrophage biology, *Br. J. Cancer* 59 (1989) 491–498.
- [24] J. Bauditz, B. Rudolph, W. Wermke, Osteoclast-like giant cell tumors of the



- pancreas and liver, *World J. Gastroenterol.* 12 (2006) 7878–7883.
- [25] A.S. Boyd, H. Wu, Y. Shyr, Monster cells in malignant melanoma, *Am. J. Dermatopathol.* 27 (2005) 208–210.
- [26] S. White-Gilbertson, C. Voelkel-Johnson, *Giants and monsters: unexpected characters in the story of cancer recurrence*, *Advances in Cancer Research*, Academic Press, Place Published, 2020.
- [27] L. Geitler, Die Entstehung der polyploiden Somakerne der Heteropteren durch Chromosomenteilung ohne Kernteilung, *Zeitschrift für Zellforschung und Mikroskopische Anatomie Abt. B Chromosoma* 1 (1939) 1–22.
- [28] T.S. Painter, E.C. Reindorp, Endomitosis in the nurse cells of the ovary of *Drosophila melanogaster*, *Zeitschrift für Zellforschung und Mikroskopische Anatomie Abt. B Chromosoma* 1 (1939) 276–283.
- [29] C.E. Nasjleti, J.M. Walden, H.H. Spencer, Polyploidy and endoreduplication in nonleukemic patients, *N. Engl. J. Med.* 272 (1965) 250–251.
- [30] Y. Okada, F. Murayama, Multinucleated giant cell formation by fusion between cells of two different strains, *Exp. Cell Res.* 40 (1965) 154–158.
- [31] E. Therman, D.A. Buchler, U. Nieminen, S. Timonen, Mitotic modifications and aberrations in human cervical cancer, *Cancer Genet. Cytogenet.* 11 (1984) 185–197.
- [32] M.J. Daniels, Y. Wang, M. Lee, A.R. Venkataraman, Abnormal cytokinesis in cells deficient in the breast cancer susceptibility protein BRCA2, *Science* 306 (2004) 876–879.
- [33] E. White, Entosis: it's a cell-eat-cell world, *Cell* 131 (2007) 840–842.
- [34] A. Janssen, R.H. Medema, Entosis: aneuploidy by invasion, *Nat. Cell Biol.* 13 (2011) 199–201.
- [35] A.G. Bell, D.G. Baker, X-irradiation-induced polyploidy in human leucocyte cultures, *Exp. Cell Res.* 38 (1965) 144–152.
- [36] K.D. Somers, M.M. Murphey, Cytochalasin B-induced multinucleation of human tumor and normal cell cultures, *Cell Biol. Int. Rep.* 4 (1980) 487–495.
- [37] P. Sentein, D. Le Coq, Enhanced polyploidy by glutaraldehyde in cultured HL60 leukemic cells, *Leuk. Res.* 10 (1986) 575–584.
- [38] C. Roussakis, N. Robillard, D. Riou, J.F. Biard, G. Pradal, P. Piloquet, C. Debitus, J.F. Verbist, Effects of bistramide A on a non-small-cell bronchial carcinoma line, *Cancer Chemother. Pharmacol.* 28 (1991) 283–292.
- [39] A. Choudhary, B. Zachek, R.F. Lera, L.M. Zasadil, A. Lasek, R.A. Denu, H. Kim, C. Kanugh, J.J. Laffin, J.M. Harter, K.B. Wisinski, S. Saha, B.A. Weaver, M.E. Burkard, Identification of selective lead compounds for treatment of high-ploidy breast cancer, *Mol. Cancer Ther.* 15 (2016) 48–59.
- [40] J. Bauke, K. Schöffling, Polyploidy in human malignancy. Hypopentaploid chromosome pattern in malignant reticulosis with secondary sideroachrestic anemia, *Cancer* 22 (1968) 686–694.
- [41] A.G. Fallenius, G.U. Auer, J.M. Carstensen, Prognostic significance of dna measurements in 409 consecutive breast cancer patients, *Cancer* 62 (1988) 331–341.
- [42] D.M. Takanishi Jr., J. Hart, P. Covarelli, R. Chappell, F. Michelassi, Ploidy as a prognostic feature in colonic adenocarcinoma, *Archiv. Surg. (Chicago. Ill.)* 131 (1996) 587–592.
- [43] H. Tsujimoto, H. Sugihara, A. Hagiwara, T. Hattori, Amplification of growth factor receptor genes and DNA ploidy pattern in the progression of gastric cancer, *Virchows Arch.* 431 (1997) 383–389.
- [44] X.F. Sun, Clinicopathological and biological features of DNA tetraploid colorectal cancers, *Cancer J. (Sudbury, Mass.)* 12 (2006) 501–506.
- [45] S. Dewhurst, C. Swanton, Tetraploidy and CIN: a dangerous combination, *Cell Cycle (Georgetown, Tex.)* 14 (2015) 3217.
- [46] L.Z. Blanco, T.A. Thurow, A. Mahajan, B. Susnik, I. Helenowski, J.S. Chmiel, M.E. Sullivan, Multinucleation is an objective feature useful in the diagnosis of pleomorphic lobular carcinoma in situ, *Am. J. Clin. Pathol.* 144 (2015) 722–726.
- [47] F. Ianzini, E.A. Kosmacek, E.S. Nelson, E. Napoli, J. Erenpreisa, M. Kalejs, M.A. Mackey, Activation of meiosis-specific genes is associated with depolyploidization of human tumor cells following radiation-induced mitotic catastrophe, *Cancer Res.* 69 (2009) 2296–2304.
- [48] E.M. Kuhn, E. Therman, B. Susman, Amitosis and endocycles in early cultured mouse trophoblast, *Placenta* 12 (1991) 251–261.
- [49] E.G. Conklin, Mitosis and amitosis, *Biol. Bull.* 33 (1917) 396–NaN-391.
- [50] E.V. Zybina, T.G. Zybina, M.S. Bogdanova, G.I. Stein, Whole-genome chromosome distribution during nuclear fragmentation of giant trophoblast cells of *Microtus rossiaemeridionalis* studied with the use of gonosomal chromatin arrangement, *Cell Biol. Int.* 29 (2005) 1066–1070.
- [51] G. Bertsch, P. J. Siemens, Nuclear fragmentation, *Phys. Lett. B* 126 (1983) 9–12.
- [52] S. Mansilla, M. Bataller, J. Portugal, A nuclear budding mechanism in transiently arrested cells generates drug-sensitizing and drug-resistant cells, *Biochem. Pharmacol.* 78 (2009) 123–132.
- [53] R. Rajaraman, M.M. Rajaraman, S.R. Rajaraman, D.L. Guernsey, Neosis—a paradigm of self-renewal in cancer, *Cell Biol. Int.* 29 (2005) 1084–1097.
- [54] J.A. Erenpreisa, M.S. Cragg, B. Fringes, I. Sharakhov, T.M. Illidge, Release of mitotic descendants by giant cells from irradiated Burkitt's lymphoma cell line, *Cell Biol. Int.* 24 (2000) 635–648.
- [55] K.H. Walen, Spontaneous cell transformation: karyoplasts derived from multinucleated cells produce new cell growth in senescent human epithelial cell cultures, in *vitro* cellular & developmental biology, *Animal* 40 (2004) 150–158.
- [56] K.H. Walen, The origin of transformed cells: studies of spontaneous and induced cell transformation in cell cultures from marsupials, a snail, and human amniocytes, *Cancer Genet. Cytogenet.* 133 (2002) 45–54.
- [57] M. Sundaram, D.L. Guernsey, M.M. Rajaraman, R. Rajaraman, Neosis: a novel type of cell division in cancer, *Cancer Biol. Ther.* 3 (2004) 207–218.
- [58] S. Zhang, I. Mercado-Urbe, J. Liu, Tumor stroma and differentiated cancer cells can be originated directly from polyploid giant cancer cells induced by paclitaxel, *Int. J. Cancer* 134 (2014) 508–518.
- [59] N. Niu, J. Zhang, N. Zhang, I. Mercado-Urbe, F. Tao, Z. Han, S. Pathak, A.S. Multani, J. Kuang, J. Yao, R.C. Bast, A.K. Sood, M.C. Hung, J. Liu, Linking genomic reorganization to tumor initiation via the giant cell cycle, *Oncogenesis* 5 (2016) e281.
- [60] J. Liu, The “life code”: a theory that unifies the human life cycle and the origin of human tumors, *Semin. Cancer Biol.* 60 (2020) 380–397.
- [61] J. Liu, The dualistic origin of human tumors, *Semin. Cancer Biol.* 53 (2018) 1–16.
- [62] K.P. Schoenfelder, D.T. Fox, The expanding implications of polyploidy, *J. Cell Biol.* 209 (2015) 485–491.
- [63] J.I. Øvrebo, B.A. Edgar, Polyploidy in tissue homeostasis and regeneration, *Development* 145 (2018) dev156034.
- [64] K.J. Gjelsvik, R. Besen-McNally, V.P. Losick, Solving the Polyploid Mystery in Health and Disease, *Trends Genet.* 35 (2019) 6–14.
- [65] S. Das, A. Lohia, Delinking of S phase and cytokinesis in the protozoan parasite *Entamoeba histolytica*, *Cell. Microbiol.* 4 (2002) 55–60.
- [66] T.L. Orr-Weaver, When bigger is better: the role of polyploidy in organogenesis, *Trends Genet.* 31 (2015) 307–315.
- [67] E. Lozano, A.G. Sáez, A.J. Flemming, A. Cunha, A.M. Leroi, Regulation of Growth by Ploidy in *Caenorhabditis elegans*, *Curr. Biol.* 16 (2006) 493–498.
- [68] A. MacAuley, J.C. Cross, Z. Werb, Reprogramming the cell cycle for endoreduplication in rodent trophoblast cells, *Mol. Biol. Cell* 9 (1998) 795–807.
- [69] D. Hu, J.C. Cross, Development and function of trophoblast giant cells in the rodent placenta, *Int. J. Dev. Biol.* 54 (2009) 341–354.
- [70] S.J. Pickering, A. Taylor, M.H. Johnson, P.R. Braude, An analysis of multi-nucleated blastomere formation in human embryos, *MHR* 1 (1995) 254–264.
- [71] Z. Liu, S. Yue, X. Chen, T. Kubin, T. Braun, Regulation of cardiomyocyte polyploidy and multinucleation by CyclinG1, *Circ. Res.* 106 (2010) 1498–1506.
- [72] J. Zanet, A. Freije, M. Ruiz, V. Coulon, J.R. Sanz, J. Chiesa, A. Gandarillas, A mitosis block links active cell cycle with human epidermal differentiation and results in endoreplication, *PLoS One* 5 (2010) e15701.
- [73] A.C. Rios, N.Y. Fu, P.R. Jamieson, B. Pal, L. Whitehead, K.R. Nicholas, G.J. Lindeman, J.E. Visvader, Essential role for a novel population of binucleated mammary epithelial cells in lactation, *Nat. Commun.* 7 (2016) 11400.
- [74] N. Takegahara, H. Kim, H. Mizuno, A. Sakaue-Sawano, A. Miyawaki, M. Tomura, O. Kanagawa, M. Ishii, Y. Choi, Involvement of receptor activator of nuclear factor- $\kappa$ B ligand (RANKL)-induced incomplete cytokinesis in the polyploidization of osteoclasts, *J. Biol. Chem.* 291 (2016) 3439–3454.
- [75] V.P. Losick, Wound-induced polyploidy is required for tissue repair, *Adv. Wound Care (New Rochelle)* 5 (2016) 271–278.
- [76] Y. Tamori, W.M. Deng, Tissue repair through cell competition and compensatory cellular hypertrophy in postmitotic epithelia, *Dev. Cell* 25 (2013) 350–363.
- [77] V.P. Losick, D.T. Fox, A.C. Spradling, Polyploidization and cell fusion contribute to wound healing in the adult *Drosophila epithelium*, *Curr. Biol.* 23 (2013) 2224–2232.
- [78] E. Cohen, S.R. Allen, J.K. Sawyer, D.T. Fox, Fuzzy-Related dictates A cell cycle switch during organ repair and tissue growth responses in the *Drosophila hindgut*, *Elife* 7 (2018) e38327.
- [79] J. Cao, J. Wang, C.P. Jackman, A.H. Cox, M.A. Trembley, J.J. Balowski, B.D. Cox, A. De Simone, A.L. Dickson, S. Di Talia, E.M. Small, D.P. Kiehart, N. Bursac, K.D. Poss, Tension creates an endoreplication wavefront that leads regeneration of epicardial tissue, *Dev. Cell* 42 (2017) 600–615 (e604).
- [80] E. Lazzeri, M.L. Angelotti, A. Peired, C. Conte, J.A. Marschner, L. Maggi, B. Mazzinghi, D. Lombardi, M.E. Melica, S. Nardi, E. Ronconi, A. Sisti, G. Antonelli, F. Becherucci, L. De Chiara, R.R. Guevara, A. Burger, B. Schaefer, F. Annunziato, H.-J. Anders, L. Lasagni, P. Romagnani, Endocycle-related tubular cell hypertrophy and progenitor proliferation recover renal function after acute kidney injury, *Nat. Commun.* 9 (2018) 1344.
- [81] Y. Miyaoka, A. Miyajima, To divide or not to divide: revisiting liver regeneration, *Cell Div.* 8 (2013) 8.
- [82] B.N. Kudryavtsev, M.V. Kudryavtseva, G.A. Sakuta, G.I. Stein, Human hepatocyte polyploidization kinetics in the course of life cycle, *Virchows Arch. B* 64 (1993) 387–393.
- [83] P.D. Wilkinson, E.R. Delgado, F. Alencastro, M.P. Leek, N. Roy, M.P. Weirich, E.C. Stahl, P.A. Otero, M.I. Chen, W.K. Brown, A.W. Duncan, The polyploid state restricts hepatocyte proliferation and liver regeneration in mice, *Hepatology* 69 (2019) 1242–1258.
- [84] T.C. Weglarz, J.L. Degen, E.P. Sandgren, Hepatocyte transplantation into diseased mouse liver: kinetics of parenchymal repopulation and identification of the proliferative capacity of tetraploid and octaploid hepatocytes, *Am. J. Pathol.* 157 (2000) 1963–1974.
- [85] K. Overturf, M. Al-Dhalimy, M. Finegold, M. Grompe, The repopulation potential of hepatocyte populations differing in size and prior mitotic expansion, *Am. J. Pathol.* 155 (1999) 2135–2143.
- [86] A.W. Duncan, Aneuploidy, polyploidy and ploidy reversal in the liver, *Semin. Cell Dev. Biol.* 24 (2013) 347–356.
- [87] S.H. Sigal, P. Rajvanshi, G.R. Gorla, R.P. Sokhi, R. Saxena, D.R. Gebhard Jr., L.M. Reid, S. Gupta, Partial hepatectomy-induced polyploidy attenuates hepatocyte replication and activates cell aging events, *Am. J. Phys.* 276 (1999) G1260–G1272.
- [88] P.D. Wilkinson, F. Alencastro, E.R. Delgado, M.P. Leek, M.P. Weirich, P.A. Otero, N. Roy, W.K. Brown, M. Oertel, A.W. Duncan, Polyploid hepatocytes facilitate adaptation and regeneration to chronic liver injury, *Am. J. Pathol.* 189 (2019) 1241–1255.
- [89] T. Matsumoto, L. Wakefield, B.D. Tarlow, M. Grompe, In vivo lineage tracing of polyploid hepatocytes reveals extensive proliferation during liver regeneration, *Cell Stem Cell* 26 (2020) 34–47.e33.

- [90] S. Zhang, K. Zhou, X. Luo, L. Li, H.-C. Tu, A. Sehgal, L.H. Nguyen, Y. Zhang, P. Gopal, B.D. Tarlow, The polyploid state plays a tumor-suppressive role in the liver, *Dev. Cell* 44 (2018) 447–459 (e445).
- [91] N.M. Borradaile, J.G. Pickering, Polyploidy impairs human aortic endothelial cell function and is prevented by nicotinamide phosphoribosyltransferase, *Am. J. Physiol.* 298 (2010) C66–C74.
- [92] H. Ikebe, T. Takamatsu, M. Itoi, S. Fujita, Age-dependent changes in nuclear DNA content and cell size of presumably normal human corneal endothelium, *Exp. Eye Res.* 43 (1986) 251–258.
- [93] G. Vedana, G. Villarreal Jr., A.S. Jun, Fuchs endothelial corneal dystrophy: current perspectives, *Clin. Ophthalmol.* 10 (2016) 321–330.
- [94] G. Gentric, V. Maillet, V. Paradis, D. Couton, A. L'Hermitte, G. Panasyuk, B. Fromenty, S. Celton-Morizur, C. Desdoutets, Oxidative stress promotes pathologic polyploidization in nonalcoholic fatty liver disease, *J. Clin. Invest.* 125 (2015) 981–992.
- [95] Z. Yang, H. Yao, F. Fei, Y. Li, J. Qu, C. Li, S. Zhang, Generation of erythroid cells from polyploid giant cancer cells: re-thinking about tumor blood supply, *J. Cancer Res. Clin. Oncol.* 144 (2018) 617–627.
- [96] G. Miesniak, M.A. Sliwiska, O. Alster, A. Strzeszewska, P. Sunderland, M. Piechota, H. Was, E. Sikora, Polyploidy formation in doxorubicin-treated cancer cells can favor escape from senescence, *Neoplasia (New York, N.Y.)* 17 (2015) 882–893.
- [97] B. Ohlstein, A. Spradling, Multipotent Drosophila intestinal stem cells specify daughter cell fates by differential notch signaling, *Science (New York, N.Y.)* 315 (2007) 988–992.
- [98] E.M. Lucchetta, B. Ohlstein, Amitosis of polyploid cells regenerates functional stem cells in the drosophila intestine, *Cell Stem Cell* 20 (2017) 609–620.e606.
- [99] K.M. Broughton, T. Khieu, N. Nguyen, M. Rosa, S. Mohsin, P. Quijada, B.J. Wang, O.H. Echeagaray, D.A. Kubli, T. Kim, F. Firouzi, M.M. Monsanto, N.A. Gude, R.M. Adamson, W.P. Dembitsky, M.E. Davis, M.A. Sussman, Cardiac interstitial tetraploid cells can escape replicative senescence in rodents but not large mammals, *Commun. Biol.* 2 (2019) 205.
- [100] A.W. Duncan, M.H. Taylor, R.D. Hickey, A.E. Hanlon Newell, M.L. Lenzi, S.B. Olson, M.J. Finegold, M. Grompe, The ploidy conveyor of mature hepatocytes as a source of genetic variation, *Nature* 467 (2010) 707–710.
- [101] N. Ahmadbeigi, M. Soleimani, M. Vasei, Y. Gheisari, Y. Mortazavi, K. Azadmanesh, A. Omidkhoda, E. Janzamin, N.B. Nardi, Isolation, characterization, and transplantation of bone marrow-derived cell components with hematopoietic stem cell niche properties, *Stem Cells Dev.* 22 (2013) 3052–3061.
- [102] N. Ahmadbeigi, M. Soleimani, F. Babaeijandaghi, Y. Mortazavi, Y. Gheisari, M. Vasei, K. Azadmanesh, S. Rostami, A. Shafiee, N.B. Nardi, The aggregate nature of human mesenchymal stromal cells in native bone marrow, *Cytotherapy* 14 (2012) 917–924.
- [103] N. Ahmadbeigi, M. Soleimani, Y. Gheisari, M. Vasei, S. Amanpour, I. Bagherizadeh, S. Ali Mohammad Shariati, K. Azadmanesh, S. Amini, A. Shafiee, V. Arabkari, N. Nardi, Dormant Phase and Multinuclear Cells: Two Key Phenomena in Early Culture of Murine Bone Marrow Mesenchymal Stem Cells, *Place Published*, 2010.
- [104] C. Leikam, A.L. Hufnagel, C. Otto, D.J. Murphy, B. Muhling, S. Kneitz, I. Nanda, M. Schmid, T.U. Wagner, S. Haferkamp, E.B. Brocker, M. Scharl, S. Meierjohann, In vitro evidence for senescent multinucleated melanocytes as a source for tumor-initiating cells, *Cell Death Dis.* 6 (2015) e1711.
- [105] J. Chen, N. Niu, J. Zhang, L. Qi, W. Shen, K.V. Donkena, Z. Feng, J. Liu, Polyploid giant cancer cells (PGCCs): the evil roots of cancer, *Curr. Cancer Drug Targets* 19 (2019) 360–367.
- [106] P.E. Puig, M.N. Gully, A. Bouchot, N. Droin, D. Cathelin, F. Bouyer, L. Favier, F. Ghiringhelli, G. Kroemer, E. Solary, F. Martin, B. Chauffert, Tumor cells can escape DNA-damaging cisplatin through DNA endoreduplication and reversible polyploidy, *Cell Biol. Int.* 32 (2008) 1031–1043.
- [107] K. Salmina, E. Jankevics, A. Huna, D. Perminov, I. Radovica, T. Klymenko, A. Ivanov, E. Jascenko, H. Scherthan, M. Cragg, J. Erenpreisa, Up-regulation of the embryonic self-renewal network through reversible polyploidy in irradiated p53-mutant tumour cells, *Exp. Cell Res.* 316 (2010) 2099–2112.
- [108] C. Lagadee, E. Vlashi, L. Della Donna, C. Dekmezian, F. Pajonk, Radiation-induced reprogramming of breast cancer cells, *Stem Cells* 30 (2012) 833–844.
- [109] S. Zhang, I. Mercado-Urbe, S. Hanash, J. Liu, iTRAQ-based proteomic analysis of polyploid giant cancer cells and budding progeny cells reveals several distinct pathways for ovarian cancer development, *PLoS One* 8 (11) (2013) e80120.
- [110] Y. Gheisari, N. Ahmadbeigi, Mesenchymal stem cells and endothelial cells: a common ancestor? *Archiv. Iran. Med.* 19 (2016) 584–587.
- [111] K. Yakin, B. Balaban, B. Urman, Impact of the presence of one or more multinucleated blastomeres on the developmental potential of the embryo to the blastocyst stage, *Fertil. Steril.* 83 (2005) 243–245.
- [112] S. Zhang, I. Mercado-Urbe, J. Liu, Generation of erythroid cells from fibroblasts and cancer cells in vitro and in vivo, *Cancer Lett.* 333 (2013) 205–212.
- [113] Y. Qu, L. Zhang, Z. Rong, T. He, S. Zhang, Number of glioma polyploid giant cancer cells (PGCCs) associated with vasculogenic mimicry formation and tumor grade in human glioma, *J. Exp. Clin. Cancer Res.* 32 (2013) 75.
- [114] L. Jia, S. Zhang, Y. Ye, X. Li, I. Mercado-Urbe, R.C. Bast Jr., J. Liu, Paclitaxel inhibits ovarian tumor growth by inducing epithelial cancer cells to benign fibroblast-like cells, *Cancer Lett.* 326 (2012) 176–182.
- [115] S. Zhang, I. Mercado-Urbe, A. Sood, R.C. Bast, J. Liu, Coevolution of neoplastic epithelial cells and multilineage stroma via polyploid giant cells during immortalization and transformation of mullerian epithelial cells, *Genes Cancer* 7 (2016) 60–72.
- [116] L. Zhang, P. Ding, H. Lv, D. Zhang, G. Liu, Z. Yang, Y. Li, J. Liu, S. Zhang, Number of polyploid giant cancer cells and expression of EZH2 are associated with VM formation and tumor grade in human ovarian tumor, *Biomed. Res. Int.* 2014 (2014) 903542.
- [117] Z. Weihua, Q. Lin, A.J. Ramoth, D. Fan, L.J. Fidler, Formation of solid tumors by a single multinucleated cancer cell, *Cancer* 117 (2011) 4092–4099.
- [118] J.W. Westra, S.E. Peterson, Y.C. Yung, T. Mutoh, S. Barral, J. Chun, Aneuploid mosaicism in the developing and adult cerebellar cortex, *J. Comp. Neurol.* 507 (2008) 1944–1951.
- [119] L. da Silva Meirelles, *Biologia de células-tronco mesenquimais pós-natais*, Genetics, Federal University of Rio Grande do Sul, Porto Alegre, 2007, p. 177.
- [120] A. Ermis, M. Oberringer, R. Wirbel, M. Koschnick, W. Mutschler, R.G. Hanselmann, Tetraploidization is a physiological enhancer of wound healing, *Eur. Surg. Res.* 30 (1998) 385–392.
- [121] M. Oberringer, D. Lothschutz, M. Jennewein, M. Koschnick, W. Mutschler, R.G. Hanselmann, Centrosome multiplication accompanies a transient clustering of polyploid cells during tissue repair, *Mol. Cell Biol. Res. Commun.* 2 (1999) 190–196.
- [122] L.M. Merlo, L.S. Wang, J.W. Pepper, P.S. Rabinovitch, C.C. Maley, Polyploidy, aneuploidy and the evolution of cancer, *Adv. Exp. Med. Biol.* 676 (2010) 1–13.
- [123] M.T. Barrett, D. Pritchard, C. Palanca-Wessels, J. Anderson, B.J. Reid, P.S. Rabinovitch, Molecular phenotype of spontaneously arising 4N (G2-tetraploid) intermediates of neoplastic progression in Barrett's esophagus, *Cancer Res.* 63 (2003) 4211–4217.
- [124] B. Boisselier, F. Dugay, M.A. Belaud-Rotureau, A. Coutolleau, E. Garcion, P. Menei, P. Guardiola, A. Rousseau, Whole genome duplication is an early event leading to aneuploidy in IDH-wild type glioblastoma, *Oncotarget* 9 (2018) 36017–36028.
- [125] A. Ogden, P.C. Rida, B.S. Knudsen, O. Kucuk, R. Aneja, Docetaxel-induced polyploidization may underlie chemoresistance and disease relapse, *Cancer Lett.* 367 (2015) 89–92.
- [126] R. Mirzayans, B. Andrais, D. Murray, Roles of polyploid/multinucleated giant cancer cells in metastasis and disease relapse following anticancer treatment, *Cancers* 10 (2018).
- [127] L. Lv, T. Zhang, Q. Yi, Y. Huang, Z. Wang, H. Hou, H. Zhang, W. Zheng, Q. Hao, Z. Guo, H.J. Cooke, Q. Shi, Tetraploid cells from cytokinesis failure induce aneuploidy and spontaneous transformation of mouse ovarian surface epithelial cells, *Cell Cycle (Georgetown, Tex.)* 11 (2012) 2864–2875.
- [128] M.C. Fontana, G. Marconi, J.D.M. Feenstra, E. Fonzi, C. Papayannidis, A. Ghelli Luserna di Rorá, A. Padella, V. Solli, E. Franchini, E. Ottaviani, A. Ferrari, C. Baldazzi, N. Testoni, I. Iacobucci, S. Soverini, T. Haferlach, V. Guadagnuolo, L. Semerad, M. Doubek, M. Steurer, Z. Racil, S. Paolini, M. Manfrini, M. Cavo, G. Simonetti, R. Kralovics, G. Martinielli, Chromothripsis in acute myeloid leukemia: biological features and impact on survival, *Leukemia* 32 (2018) 1609–1620.
- [129] I.V. Uryvaeva, A.S. Mikaelyan, N.O. Dashenkova, T.L. Marshak, Chromothripsis in hepatocarcinogenesis: the role of a micronuclear aberration and polyploidy, *Biol. Bull.* 45 (2018) 419–425.
- [130] B.R. Mardin, A.P. Drainsas, S.M. Waszak, J. Weischenfeldt, M. Isokane, A.M. Stütz, B. Raeder, T. Efthymiopoulos, C. Buccitelli, M. Segura-Wang, A cell-based model system links chromothripsis with hyperploidy, *Mol. Syst. Biol.* 11 (2015) 828.
- [131] F. Notta, M. Chan-Seng-Yue, M. Lemire, Y. Li, G.W. Wilson, A.A. Connor, R.E. Denroche, S.-B. Liang, A.M. Brown, J.C. Kim, A renewed model of pancreatic cancer evolution based on genomic rearrangement patterns, *Nature* 538 (2016) 378–382.
- [132] Z. Storchova, D. Pellman, From polyploidy to aneuploidy, genome instability and cancer, *Nat. Rev. Mol. Cell Biol.* 5 (2004) 45–54.
- [133] G. Rancati, N. Pavelka, B. Fleharty, A. Noll, R. Trimble, K. Walton, A. Perera, K. Staehling-Hampton, C.W. Seidel, R. Li, Aneuploidy underlies rapid adaptive evolution of yeast cells deprived of a conserved cytokinesis motor, *Cel* 135 (2008) 879–893.
- [134] S.L. Chang, H.Y. Lai, S.Y. Tung, J.Y. Leu, Dynamic large-scale chromosomal rearrangements fuel rapid adaptation in yeast populations, *PLoS Genet.* 9 (2013) e1003232.
- [135] R. Dhar, R. Sagesser, C. Weikert, J. Yuan, A. Wagner, Adaptation of *Saccharomyces cerevisiae* to saline stress through laboratory evolution, *J. Evol. Biol.* 24 (2011) 1135–1153.
- [136] A.M. Selmecki, Y.E. Maruvka, P.A. Richmond, M. Guillet, N. Shores, A.L. Sorenson, S. De, R. Kishony, F. Michor, R. Dowell, D. Pellman, Polyploidy can drive rapid adaptation in yeast, *Nature* 519 (2015) 349.
- [137] P.S. Soltis, D.B. Marchant, Y. Van de Peer, D.E. Soltis, Polyploidy and genome evolution in plants, *Curr. Opin. Genet. Dev.* 35 (2015) 119–125.
- [138] Y. Miyaoka, K. Ebato, H. Kato, S. Arakawa, S. Shimizu, A. Miyajima, Hypertrophy and unconventional cell division of hepatocytes underlie liver regeneration, *Curr. Biol.* 22 (2012) 1166–1175.
- [139] K. Yanger, Y. Zong, L.R. Maggs, S.N. Shapira, R. Maddipati, N.M. Aiello, S.N. Thung, R.G. Wells, L.E. Greenbaum, B.Z. Stanger, Robust cellular reprogramming occurs spontaneously during liver regeneration, *Genes Dev.* 27 (2013) 719–724.
- [140] A.W. Duncan, A.E.H. Newell, W. Bi, M.J. Finegold, S.B. Olson, A.L. Beaudet, M. Grompe, Aneuploidy as a mechanism for stress-induced liver adaptation, *J. Clin. Invest.* 122 (2012) 3307–3315.
- [141] E.K. Rofstad, N.M. Johnsen, H. Lyng, Hypoxia-induced tetraploidisation of a diploid human melanoma cell line in vitro, *Br. J. Cancer* 27 (1996) S136–S139.
- [142] E.-B. Braune, Y.L. Tsoi, Y.P. Phoon, S. Landor, H. Silva Cascales, D. Ramsköld, Q. Deng, A. Lindqvist, X. Lian, C. Sahlgren, S.-B. Jin, U. Lendahl, Loss of CSL unlocks a hypoxic response and enhanced tumor growth potential in breast cancer cells, *Stem Cell Rep.* 6 (2016) 643–651.

- [143] Z. Tan, Z.V. Chu, Y.J.A. Chan, Y.E. Lu, G. Rancati, Mammalian cells undergo endoreduplication in response to lactic acidosis, *Sci. Rep.* 8 (2018) 2890.
- [144] R.P. Arun, D. Sivanesan, B. Patra, S. Varadaraj, R.S. Verma, Simulated microgravity increases polyploid giant cancer cells and nuclear localization of YAP, *Sci. Rep.* 9 (2019) 10684.
- [145] J.J. Chen, Genomic instability induced by human papillomavirus oncogenes, *North Am. J. Med. Sci.* 3 (2010) 43–47.
- [146] D. Patel, A. Incassati, N. Wang, D.J. McCance, Human papillomavirus type 16 E6 and E7 cause polyploidy in human keratinocytes and up-regulation of G2-M-phase proteins, *Cancer Res.* 64 (2004) 1299–1306.
- [147] L. Revesz, U. Norman, Chromosome ploidy and radiosensitivity of tumours, *Nature* 187 (1960) 861–862.
- [148] L.M. Merlo, J.W. Pepper, P.S. Rabinovitch, C.C. Maley, Polyploidy, aneuploidy and the evolution of cancer, *Polyplodization and Cancer*, Springer, Place Published, 2010, pp. 1–13.
- [149] G. Cantero, N. Pastor, S. Mateos, C. Campanella, F. Cortes, Cisplatin-induced endoreduplication in CHO cells: DNA damage and inhibition of topoisomerase II, *Mutat. Res.* 599 (2006) 160–166.
- [150] J. Coward, A. Harding, Size does matter: why polyploid tumor cells are critical drug targets in the war on cancer, *Front. Oncol.* 4 (2014) 123.
- [151] T.C. Hsieh, F. Tragano, Z. Darzynkiewicz, J.M. Wu, The 2,6-disubstituted purine reversine induces growth arrest and polyploidy in human cancer cells, *Int. J. Oncol.* 31 (2007) 1293–1300.
- [152] C.H. Kuo, Y.C. Lu, Y.S. Tseng, C.S. Shi, S.H. Chen, P.T. Chen, F.L. Wu, Y.P. Chang, Y.R. Lee, Reversine induces cell cycle arrest, polyploidy, and apoptosis in human breast cancer cells, *Breast Cancer (Tokyo, Japan)* 21 (2014) 358–369.
- [153] L. Cheng, H. Wang, K. Guo, Z. Wang, Z. Zhang, C. Shen, L. Chen, J. Lin, Reversine, a substituted purine, exerts an inhibitive effect on human renal carcinoma cells via induction of cell apoptosis and polyploidy, *OncoTargets Ther.* 11 (2018) 1025–1035.
- [154] S.K. Martin, H. Pu, J.C. Penticuff, Z. Cao, C. Horbinski, N. Kyprianou, Multinucleation and mesenchymal-to-epithelial transition alleviate resistance to combined cabazitaxel and antiandrogen therapy in advanced prostate cancer, *Cancer Res.* 76 (2016) 912–926.
- [155] K. Mittal, S. Donthamsetty, R. Kaur, C. Yang, M.V. Gupta, M.D. Reid, D.H. Choi, P.C.G. Rida, R. Aneja, Multinucleated polyploidy drives resistance to Docetaxel chemotherapy in prostate cancer, *Br. J. Cancer* 116 (2017) 1186–1194.
- [156] S. Islam, A.L. Paek, M. Hammer, S. Rangarajan, R. Ruijtenbeek, L. Cooke, E. Weterings, D. Mahadevan, Drug-induced aneuploidy and polyploidy is a mechanism of disease relapse in MYC/BCL2-addicted diffuse large B-cell lymphoma, *Oncotarget* 9 (2018) 35875–35890.
- [157] P. Balsas, P. Galan-Malo, I. Marzo, J. Naval, Bortezomib resistance in a myeloma cell line is associated to PSMbeta5 overexpression and polyploidy, *Leuk. Res.* 36 (2012) 212–218.
- [158] A.A. Dar, L.W. Goff, S. Majid, J. Berlin, W. El-Rifai, Aurora kinase inhibitors—rising stars in cancer therapeutics? *Mol. Cancer Ther.* 9 (2010) 268–278.
- [159] M. Kawakami, L.M. Mustachio, L. Zheng, Y. Chen, J. Rodriguez-Canales, B. Mino, J.M. Kurie, J. Roszik, P.A. Villalobos, K.L. Thu, J. Silvester, D.W. Cescon, I.I. Wistuba, T.W. Mak, X. Liu, E. Dmitrovsky, Polo-like kinase 4 inhibition produces polyploidy and apoptotic death of lung cancers, *Proc. Natl. Acad. Sci. U. S. A.* 115 (2018) 1913–1918.
- [160] S. Sharma, J.Y. Zeng, C.M. Zhuang, Y.Q. Zhou, H.P. Yao, X. Hu, R. Zhang, M.H. Wang, Small-molecule inhibitor BMS-777607 induces breast cancer cell polyploidy with increased resistance to cytotoxic chemotherapy agents, *Mol. Cancer Ther.* 12 (2013) 725–736.
- [161] J.Y. Kim, C.H. Kim, I.J. Stratford, A.V. Patterson, J.H. Hendry, The bioreductive agent RH1 and gamma-irradiation both cause G2/M cell cycle phase arrest and polyploidy in a p53-mutated human breast cancer cell line, *Int. J. Radiat. Oncol. Biol. Phys.* 58 (2004) 376–385.
- [162] C. Lagadec, E. Vlasi, L. Della Donna, C. Dekmezian, F. Pajonk, Radiation-induced reprogramming of breast cancer cells, *Stem Cells (Dayton, Ohio)* 30 (2012) 833–844.
- [163] L.L. Liu, Z.J. Long, L.X. Wang, F.M. Zheng, Z.G. Fang, M. Yan, D.F. Xu, J.J. Chen, S.W. Wang, D.J. Lin, Q. Liu, Inhibition of mTOR pathway sensitizes acute myeloid leukemia cells to aurora inhibitors by suppression of glycolytic metabolism, *Mol. Cancer Res.* 11 (2013) 1326–1336.
- [164] P. Donovan, K. Cato, R. Legaie, R. Jayalath, G. Olsson, B. Hall, S. Olson, S. Boros, B.A. Reynolds, A. Harding, Hyperdiploid tumor cells increase phenotypic heterogeneity within Glioblastoma tumors, *Mol. BioSyst.* 10 (2014) 741–758.
- [165] S. Sharma, H.P. Yao, Y.Q. Zhou, J. Zhou, R. Zhang, M.H. Wang, Prevention of BMS-777607-induced polyploidy/senesence by mTOR inhibitor AZD8055 sensitizes breast cancer cells to cytotoxic chemotherapeutics, *Mol. Oncol.* 8 (2014) 469–482.
- [166] W. Zhou, J. Xu, E. Gelston, X. Wu, Z. Zou, B. Wang, Y. Zeng, H. Wang, A. Liu, L. Xu, Q. Liu, Inhibition of Bcl-xL overcomes polyploidy resistance and leads to apoptotic cell death in acute myeloid leukemia cells, *Oncotarget* 6 (2015) 21557–21571.
- [167] D. Lissa, L. Senovilla, S. Rello-Varona, I. Vitale, M. Michaud, F. Pietrocola, A. Boileve, F. Obrist, C. Bordenave, P. Garcia, J. Michels, M. Jemaa, O. Kepp, M. Castedo, G. Kroemer, Resveratrol and aspirin eliminate tetraploid cells for anticancer chemoprevention, *Proc. Natl. Acad. Sci. U. S. A.* 111 (2014) 3020–3025.
- [168] G. Mosieniak, M.A. Sliwinska, O. Alster, A. Strzeszewska, P. Sunderland, M. Piechota, H. Was, E. Sikora, Polyploidy formation in doxorubicin-treated cancer cells can favor escape from senescence, *Neoplasia (New York, N.Y.)* 17 (2015) 882–893.
- [169] A.P. Havas, K.B. Rodrigues, A. Bhakta, J.A. Demirjian, S. Hahn, J. Tran, M. Scavello, A.A. Tula-Sanchez, Y. Zeng, M. Schmelz, C.L. Smith, Belinostat and vincristine demonstrate mutually synergistic cytotoxicity associated with mitotic arrest and inhibition of polyploidy in a preclinical model of aggressive diffuse large B cell lymphoma, *Cancer Biol. Ther.* 17 (2016) 1240–1252.
- [170] Y.T. Yeung, S. Yin, B. Lu, S. Fan, R. Yang, R. Bai, C. Zhang, A.M. Bode, K. Liu, Z. Dong, Losmapimod overcomes gefitinib resistance in non-small cell lung cancer by preventing tetraploidization, *EBioMedicine* 28 (2018) 51–61.
- [171] M. Motwani, X. Li, G.K. Schwartz, Flavopiridol, a cyclin-dependent kinase inhibitor, prevents spindle inhibitor-induced endoreduplication in human cancer cells, *Clin. Cancer Res.* 6 (2000) 924–932.
- [172] A. Ogden, P.C.G. Rida, R. Aneja, Let's huddle to prevent a muddle: centrosome declustering as an attractive anticancer strategy, *Cell Death Differ.* 19 (2012) 1255–1267.
- [173] H. Raslova, V. Baccini, L. Loussaief, B. Comba, J. Larghero, N. Debili, W. Vainchenker, Mammalian target of rapamycin (mTOR) regulates both proliferation of megakaryocyte progenitors and late stages of megakaryocyte differentiation, *Blood* 107 (2006) 2303–2310.
- [174] R. Mirzayans, D. Murray, Intratumor heterogeneity and therapy resistance: contributions of dormancy, apoptosis reversal (anastasis) and cell fusion to disease recurrence, *Int. J. Mol. Sci.* 21 (2020).