PERSPECTIVE

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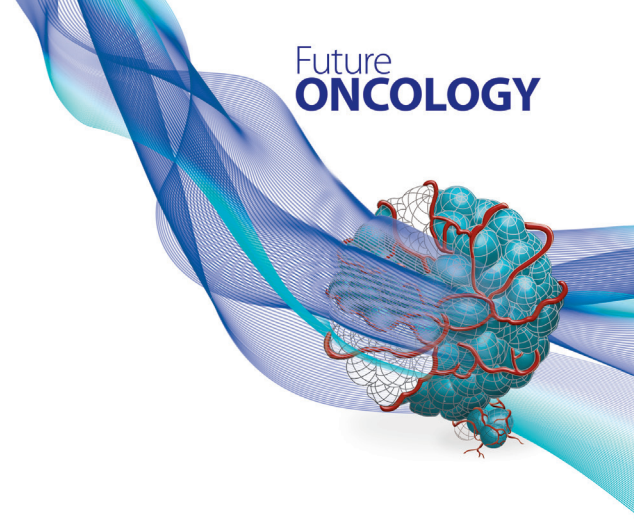

A periodic table for cancer

Richard J Epstein*

ABSTRACT Cancers exhibit differences in metastatic behavior and drug sensitivity that correlate with certain tumor-specific variables such as differentiation grade, growth rate/extent and molecular regulatory aberrations. In practice, patient management is based on the past results of clinical trials adjusted for these biomarkers. Here, it is proposed that treatment strategies could be fine-tuned upfront simply by quantifying tumorigenic spatial (cell growth) and temporal (genetic stability) control losses, as predicted by genetic defects of cell-cycle-regulatory gatekeeper and genome-stabilizing caretaker tumor suppressor genes, respectively. These differential quantifications of tumor dysfunction may in turn be used to create a tumor-specific 'periodic table' that guides rational formulation of survival-enhancing anticancer treatment strategies.

KEYWORDS
• apoptosis • genetic instability • personalized medicine • tumor suppressor genes

Scientific progress depends on the discovery of homologies and continuities [1]. Breakthroughs tend not to be made by the description of novel phenomena *per se*, but by the perception of hitherto unrecognized patterns that unite datasets and thus generate testable hypotheses [2]. Darwin's use of the fossil record to infer the basis of evolution by natural selection was one such continuous paradigm [3], while the recognition of structural regularities underlying the attributes of elements was another [4]. Acceptance of such transformational paradigms tends to be slow, as proof of principle is cautiously awaited by skeptics [5]. For example, the postulation of Mendeleev's periodic law in 1869 was merely a first step in changing the alchemical view of immutable elements [6] to that of connected entities sharing the subatomic continuity of quantum physics, as was only proven beyond doubt decades later by Rutherford, Bohr *et al.* [7,8].

It has taken over a century since Mendeleev's era for this same data-driven process to begin modernizing biological research – illustrated, for instance, by attempts to devise a 'periodic table' for protein structures [9]. In parallel with this trend, the study of cancer has been inching away from its surgical roots as a group of organ-based diseases, and toward a continuous spectrum of molecular pathologies [10]. For cancer researchers this has been an exciting time to witness the birth of a new era in which phenotypic and therapeutic predictions can sometimes be made on the basis of genotypic aberrations [11]. For clinicians, however, this tumoricentric vision of personalized oncology (or precision medicine [12]) has so far failed to yield a generalizable solution to the adaptive and heterogeneous challenges of anticancer therapeutics [13–15].

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Moderating molecular expectations

To understand the limitations of personalized oncology based on targets alone, it is necessary to appreciate that cancer growth is subject to at least three qualitatively distinct sets of variables **(Figure 1)** [16]:

- Tumor-specific activation or upregulation of oncogenic 'pushers' (drivers, drug targets);
- Tumor-specific functional defects in control-regulatory 'pullers' (suppressor gene products);
- Extratumoral defects in systemic regulatory 'holders' (environmental control factors).

One key tripwire for personalized oncology is thus camouflaged by normality – namely, the tumor microenvironment [17]. During early growth, carcinomas do not survive autonomously [18]; rather, such cells evolve in symbiosis with a multicellular network of regulatory inputs [19] including those from stromal cells and extracellular matrix [20], hormones and growth factors [21], metabolic modifiers of cell survival and inflammation [22] and immune surveillance [23]. Since none of these extrinsic constraints are measurable by direct tumor analysis, the predictivity of assays based solely on the latter can only ever be partial [24]. Although the central importance of oncoproteins and the tumor microenvironment to human cancer development and progression is unarguable, the following discussion focuses on the far more neglected therapeutic implications of suppressor gene dysfunctionality.

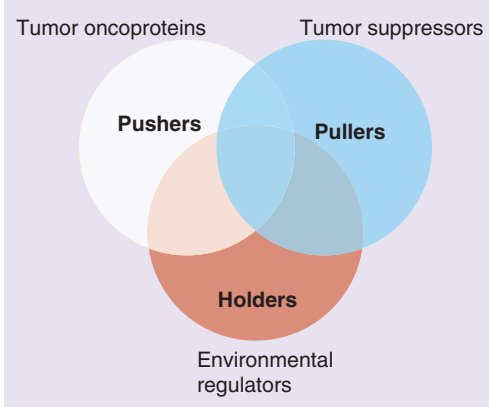

Figure 1. The 'tumor trinity' of factors modifying cancer growth and progression: activated oncoproteins ('pushers', top left), functional losses of suppressor gene function ('pullers', top right); and defects in environmental control factors such as immunity, metabolism or stromal–epithelial constraints ('holders', bottom). Of these, tumor-based analyses are restricted to the first two, while 'actionable' clinical decision-making is influenced solely by the first.
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Another tripwire for the unwary molecular oncopathologist is tumor heterogeneity [25], a phenomenon originating in part from mutations causing (and/or caused by) genetic instability [26,27]. The spatial heterogeneity of a tumor is a snapshot of genetic instability over time, posing a challenge to clinicians who seek a single drug panacea for a given patient. Awareness of this problem has popularized the use of multiple biopsies to define the multiclonal genotypes of tumors [28], and has encouraged sequential tumor sampling to detect evolving changes in cancers progressing to resistance [28]. Yet it remains unclear when such labor-intensive investments in tumor analysis will yield a commensurate return in terms of longer and better patient survival [29].

Some experts have suggested that the honeymoon period for personalized oncology may already be over [30], as few paradigm-shifting insights [31] or transdisciplinary models [32] of clinical benefit have yet materialized from the Big Data revolution. Moreover, the rising costs of personalized medicine have highlighted a need for 'precision prescribing' that enhances patient survival in an affordable manner [33]. Tumor exome sequencing [34] is now being marketed direct to patients, but the value of clinical benefit remains controversial; prescribing decisions remain influenced by just a handful of actionable mutations, amplifications or fusions that predict *de novo* response or resistance to targeted drugs [35].

Enhancing actionability

The dilemmas now facing postgenomic therapeutics are best understood in historical context. Decades ago, the discovery of oncogenes [36] raised the idea that a single genetic anomaly could account for cancer pathogenesis, diagnosis and therapeutic control alike **(Figure 2A)**. At first this seemed to be the case for many pediatric tumors and hematologic malignancies characterized by diagnostic chromosomal anomalies (e.g., the 9:22 *Bcr/Abl* translocation

of chronic myeloid leukemia) indicating a major targetable genetic aberration. For common age-related carcinomas, however, euphoria over the 'oncogene model' began to cool as it was realized that constitutive mitogenic events in epithelial tissues tend to require multiple permissive defects of upstream control genes [37]. About 100 such 'antioncogenes' [38] – later designated 'tumor suppressor genes' [39] – have since been implicated in the pathogenesis of familial cancer syndromes when mutant [40]. In 1997 Vogelstein and Kinzler observed that most of these familial cancer genes are classifiable into two (and only two) main functional groups [41], which they termed:

- Gatekeepers: cell-cycle regulatory genes that govern cell-cycle traverse, and thus control stress-induced programmed cell death, or apoptosis; e.g., *TP53, APC, CDH1, Rb*; or
- Caretakers: DNA repair or genome maintenance genes that optimize genetic stability and hence reduce mutation load [42]; e.g., *BRCA*-family, *ATM* or mismatch repair (MMR) genes such as *MLH1*. *BRCA* (homologous recombination) defects cause chromosomal instability (CIN) [43], whereas MMR defects cause microsatellite instability (MIN) [44].

The gatekeeper subclass of suppressor genes regulates diverse cell-cycle control events – including, for example, growth arrest mediated by the gatekeeper *CDH1* gene that normally encodes the epithelial adhesion molecule E-cadherin [45] – but the most cancer-protective of such events is arguably apoptosis, or programmed cell death **(Figure 2B)** [46]. Consistent with this, the low efficiency of hematogenous metastatic seeding is increased by apoptotic resistance [47–50] due to gatekeeper mutations [51–55]. The terminology 'apoptotic resistance', or similar, is used below for convenience to denote all tumorigenic phenotypes due to gatekeeper gene defects.

In contrast, caretaker gene defects predispose to tumorigenesis via more subtle and indirect acceleration of tumor progression to multicellular anarchy **(Figure 2C)** [56,57]. Caretaker gene mutations are better tolerated in the germline – that is, less often embryonic lethal – than gatekeeper gene mutations [40]. Hence, unlike gatekeeper mutations, caretaker mutations (e.g., involving the *BRCA* or MMR genes) are relatively common in the population, raising the question as to whether such mutations confer a heterozygous survival advantage during environmental stress. The latter possibility is consistent with one study reporting unexpectedly fewer miscarriages in *BRCA* mutation carriers than in noncarriers (25.2% vs 29.1%) [58].

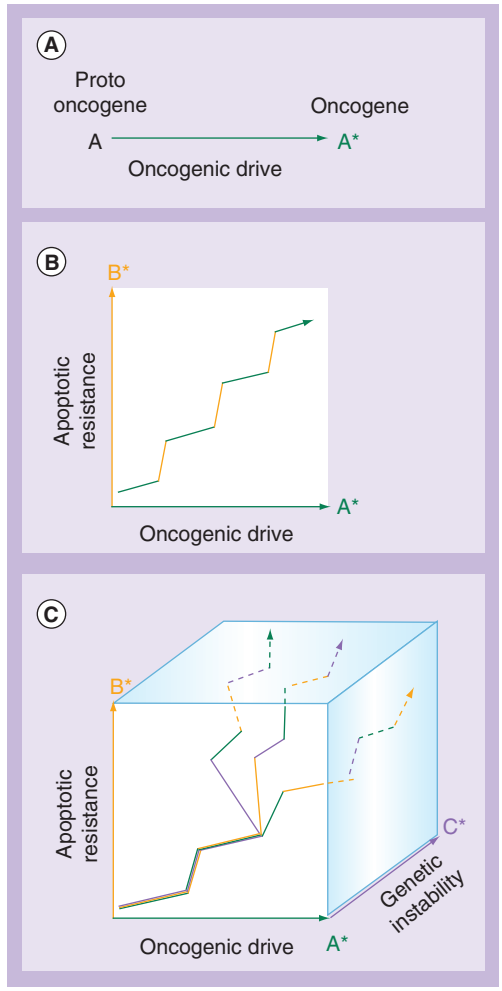

Figure 2. Genetic models of tumor growth in order of complexity. (A) One-dimensional (linear) model of tumor growth, in which constitutive activation of a single oncogene is considered the exclusive 'driver' event. **(B)** 2D model of tumor growth mediated both by oncogenic drive (horizontal dimension) and a prerequisite gatekeeper loss-of-function event (vertical dimension) permitting clonal apoptotic failure in response to oversignaling selection pressures. **(C)** 3D model of tumor progression incorporating the above two molecular events, but adding a third dimension of genetic instability due to a caretaker gene loss-of-function event.

Despite recent advances in genome sequencing capacity, there remains little software to assess

the integrity of these tumor-suppressive 'pullers', nor to quantify the myriad unactionable exome mutations, signaling molecule aberrations, unusual amplicons and deletions, intronic and/or splice variants, epimutations, transcriptomic and miRNA alterations, superenhancer anomalies and so on that comprise the dark matter of cancer cell nuclei. Conversely, as pathogenetic models of cancer become more elaborate [59], there is a risk that disease understanding becomes not easier but harder for cancer physicians. Yet, evolution has ensured that biology (of which cancer is one derangement) remains simple in principle, being distinguished from physics and chemistry by two main features:

- The selecting mechanism of death, which controls and moulds self-replicating populations;
- The interaction between organisms and the environment, which mandates adaptation.

These two 'hallmarks of life' ensure the concurrent conservation and change of the germline genome, as mediated by heritable adjustments to cell death thresholds and damage responses [60]. In general, the more complex the organism, the more cell death genes required to generate and maintain that complexity [40], and the greater the risk of species extinction ('genomic brittleness') due to reduced adaptability [61]. This balance between immediate cell survival and future genome adaptivity is thus intrinsic to evolution. It follows that most biological decisions may be reduced to two outcomes – stop/go, on/off, live/die, persist/transform – the advantage of which is selected by their effects on genome survival. Indeed, even so-called pluripotent (stem) cells are restricted at any one time to binary decisions, such as whether to undergo a symmetric or asymmetric division [62].

Certain dualities flow from this inherent binary nature of living processes. First, death controls the spatial dimension of life (whether by replication or survival/growth), whereas adaptivity controls its temporal dimension (genomic plasticity); these two control mechanisms mediate the balancing evolutionary forces of negative (purifying) and positive (adaptive) selection [63]. Second, as noted above, the normal phenotypes of cell growth and genome plasticity are constrained by two functionally distinct (gatekeeper and caretaker) gene sets [41,64]. Third, (epi) genetic defects of the latter gene classes give rise to the two most pivotal problems in clinical oncology: drug resistance, whether primary or acquired, and genetic disease progression [65]. These pairings support the view that both normal and neoplastic biology are regulated by these two properties of growth control and genome stability. If so, a 2D roadmap of tumor evolution should be derivable from the (epi)mutation spectra of the gene sets governing these properties [66], enabling creation of a predictive data tool that could assist both oncologists and their patients [67]. This is easier said than done, for two reasons: first, growth control and genome stability are not independent over time; and second, actionable treatment strategies to restore functional defects of these regulatory pathways are not obvious.

Challenge 1: quantifying cancer dysfunctionality

Caretaker and gatekeeper suppressors are qualitatively distinct: the two gene classes differ in terms of phylogenetic gene number, exon length, dinucleotide composition, evolutionary rate, embryonic essentiality, expression level and breadth, and susceptibility to missense *versus* nonsense mutations [40,68–69]. Yet despite these evolutionary differences, these gene groups are not independent in terms of function: just as gatekeeper mutations impair apoptosis and thus permit uncontrolled oncogene activation or upregulation [37], so do caretaker mutations permit more rapid microevolution of gatekeeper defects. The high frequency of somatic *TP53* gatekeeper mutations in tumors of *BRCA*-mutant families is a case in point [70]); on the other hand, afferent defects in the caretaker 'sensing' limb of the DNA damage response can blunt apoptotic responsiveness and functionality [71]. Moreover, genetic instability due to primary caretaker gene defects is increased during disease progression by stepwise accumulation of gatekeeper mutations that enable cells with disorganized (unstable) genomes or aneuploidy to survive [72,73]; this is suggested by the strong association of CIN in colorectal cancer with *APC* gatekeeper gene mutations [74]. Precise quantification of genetic instability or apoptotic resistance must therefore involve development of algorithms beyond simple enumeration of caretaker or gatekeeper mutations – particularly in late-stage and/or heavily-pretreated tumors, where both dysfunctions tend to accumulate in parallel.

Genetic instability and apoptotic resistance may thus be portrayed as the x and y coordinates of a tumor dysfunctionality map, although

their quantification remains challenging. For a given organ-specified tumor histology, an initial estimate as to the severity of these dysfunctions may be inferred from data on mutation burden [66] and chemotherapy response rates [31], respectively. By estimating genetic instability as a function of mutation load [10] – thereby distinguishing stable (e.g., breast, prostate, glioblastoma) from moderately unstable (e.g., gastric, esophageal, head/neck) and highly unstable (e.g., smoking-induced lung) cancers [66] – it should be possible to estimate the risk that a tumor will acquire rapid resistance to cytotoxic interventions [75]. Similarly, the *de novo* resistance profile of a tumor type may be predictable in part from gatekeeper pathway mutation burden. Such analyses align with low-response (DNA damage resistant) tumors, such as pancreatic cancer, renal cell carcinoma and melanoma; medium-response tumors, such as colon and breast cancers; and high-response (damage-susceptible) tumors, such as hairy cell or chronic myeloid leukemias [31].

The first-generation map depicted in **Figure 3** is in effect a 2D 'vogelgram' [76] based on pathway losses of apoptotic sensitivity and genetic stability. How might these dysfunctions be systematically quantified as integral (i.e., 'periodic') grades? Consider the example of a cancer that harbors 200 functional mutations of which 10% are identified as pro-tumorigenic gene aberrations [77]. Since molecular lesions affecting the same oncogenic pathway tend to occur within single tumors on a mutually exclusive basis [78,79], it is fair to infer that non-silent mutations in different pathways have a non-identical functional significance, and hence should confer a more severe (e.g., additive) impairment of control when combined [80].

Furthermore, since there exist parallel independent pathways controlling the properties of both genetic stability (e.g., CIN vs. MIN) and apoptosis (e.g., p53-dependent vs. -independent pathways [81]), the notion that multiple heterologous lesions yield greater dysfunctionality is justified. By the same token, multiple genetic lesions identified within the same (e.g., mTOR) pathway imply a greater dysfunctionality weighting than would single lesions of unclear functional significance. Mutational microanatomy also influences the severity of functional derangement for a given gene, as measurable by alterations in *TP53* transactivation versus dominant negative effects [82], for example. Sophisticated

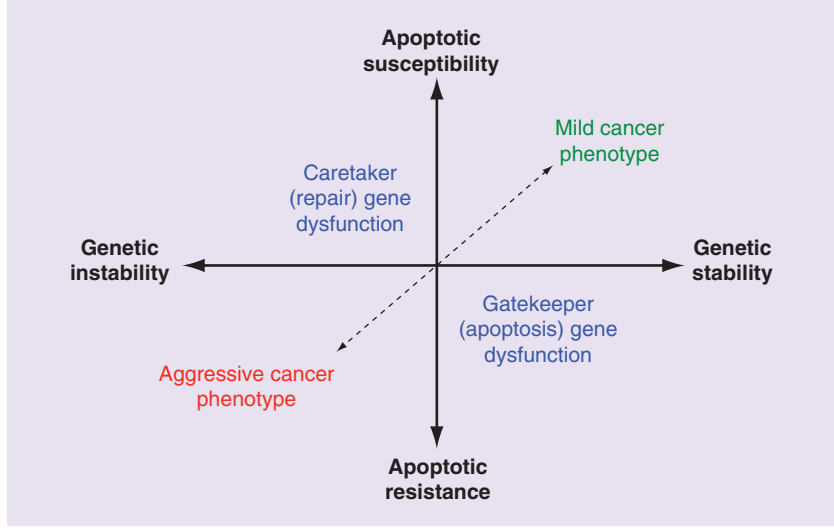

Figure 3. 2D phenotypic framework for a periodic cancer table, consisting of genetic stability (x axis, abscissa; mediated by caretaker tumor suppressor genes) mapped against apoptotic sensitivity (y axis, ordinate; mediated by gatekeeper genes). Tumor coordinates mapping to this framework are envisaged to predict, respectively, the speed of tumor progression, and the likelihood of tumor chemosensitivity or resistance.

genome-interpretive softwares may thus be able to yield an approximate digital grading (say, grade 1–4) for suppressor pathway epi/mutations affecting a given cancer.

A prototype schema as to how rising mutation loads may yield more aggressive cancer phenotypes is shown in **Figure 4**. Here, the paradigm of stepwise mutations leading to the evolution of more dysregulated tumors is illustrated by one organ-specified tumor type – breast cancer – for which several genetic subtypes are defined [83]. Without incorporating less common gene aberrations such as those affecting *RAD51C*, *BRIP1* or *PALB2*, a 2D (gatekeeper vs caretaker) molecular continuum portrays the downhill evolution of an ancestral estrogen receptor (ER)-positive breast tumor to an aggressive ER-negative, HER2-positive or triple-negative cancer subtype via serial mutations in *BRCA*-pathway caretaker genes [84], *TP53*-pathway gatekeepers [85] or both **(Figure 4A)** [86].

This slalom sequence of suppressor-deregulation events can be refined by importing additional pathways such as those mediated by *ATM*, *PTEN* (constraining the *PIK3CA–Akt–mTORC* gene network) and/or *CDH1* **(Figure 4B)**. Many nonmutational (epi)genetic or signaling aberrations may also be recruited into information-losing cascades of this type [87]. For example, loss of E-cadherin expression often occurs during

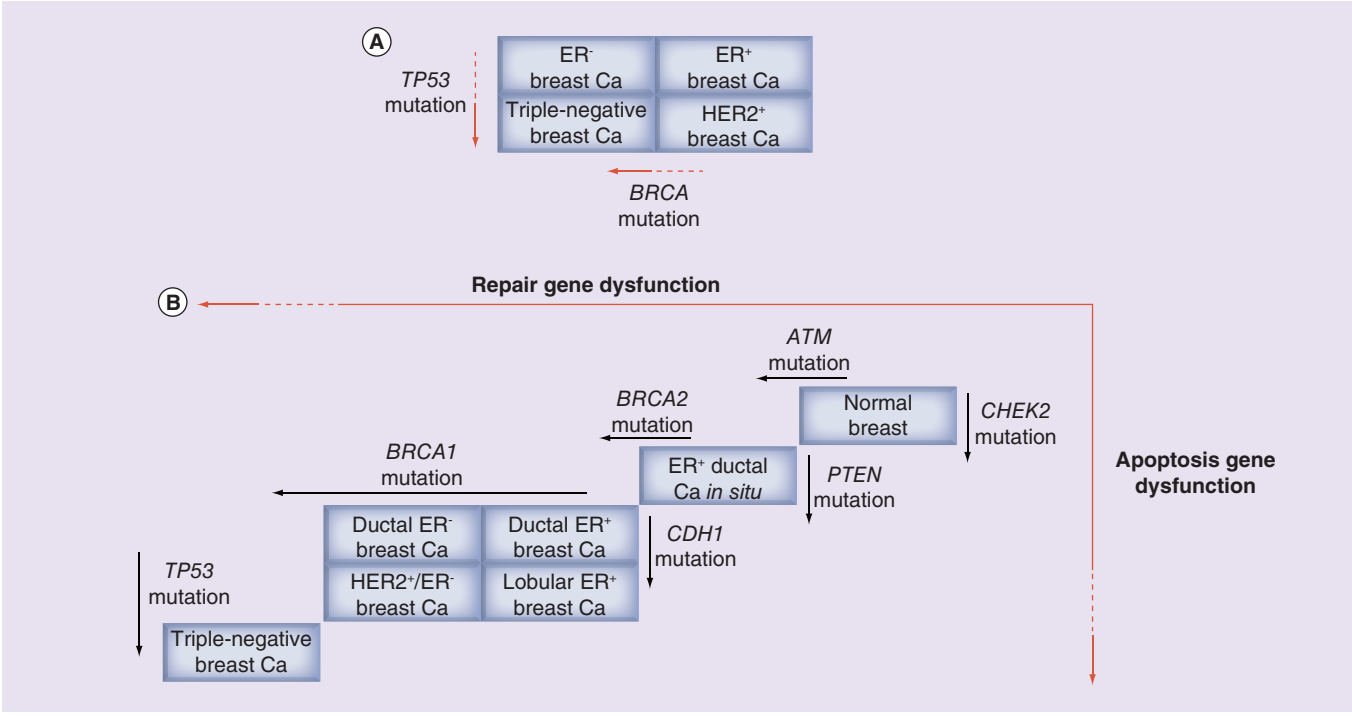

Figure 4. Microevolution schema of breast cancer subtypes through successive suppressor pathway defects. (A) Genetic transformation of an ER+ breast tumor to less differentiated ER-, HER2+, or triple-negative cancer variants, positing key pathogenetic roles of BRCA caretaker and/or p53 gatekeeper pathway dysfunctions. **(B)** More complex schema showing additional genes involved, including the *CDH1* (E-cadherin-encoding) gene locus typically mutated in classic lobular breast cancers. As in **Figure 3**, the direction of phenotypic cancer progression is from top right to bottom left.
Ca: Cancer/carcinoma; ER: Estrogen receptor.

progression of invasive ductal breast cancers as a result of epithelial-to-mesenchymal transition, even though loss-of-function *CDH1* somatic mutations are seldom seen in this (nonlobular) cancer context [88]. Similar schemas can depict the 2D mutational progression of colorectal cancer subtypes – in this context, gatekeeper defects include *APC, STK11, SMAD4, PTEN* and *TP53;* caretaker defects include the MMR genes and *MYH*; while permissive oncogene activation events affect either *BRAF* (typically with MMR gene mutations [89]) or *KRAS* (typically with *TP53* mutations [85,90]). Quantifiable mitotic biomarkers such as Ki-67 or fluorodeoxyglucose avidity may thus correlate with the burden of suppressor gene loss in such dedifferentiating tumor subtypes [91].

Challenge 2: managing cancer dysfunctionality

How might such dysfunctionality profiles assist a practising clinician? **Figure 5** depicts a second-generation cancer map based partly on the data sources cited above, and partly on historical disease behaviors [75]. This graphical representation shows that a semiquantitative rendering of two distinct tumor dysfunctions is feasible, although no claim is intended as to the validity or utility of **Figure 5** as it stands. However, the acknowledgement of broad variations in tumor instability and/or apoptotic resistance does support the possibility that treatments could be modified for individual patients on a custom basis. For example, if a tumor is deemed apoptotic-resistant but genetically stable – that is, it maps to the bottom right of the periodic table – a clinician may reasonably infer that: (1) disease ablation (say, through complete surgical extirpation with clear margins) could well prove durable; (2) cytotoxic treatments are unlikely to trigger major cytoreductions; (3) sensitization strategies to lower the apoptotic threshold may help to enhance drug sensitivity [92]; and (4) association with a low tumor cell proliferation rate on biopsy, and/or radioglucose avidity on positron scanning, suggests a relatively indolent course of metastatic progression in the absence of treatment. Conversely, if a tumor

is localized to the upper left, indicating high genetic instability with low apoptotic resistance, then marked initial responses to treatment could be complicated by rapid resistant relapses with minimal survival benefit. This could in turn suggest the desirability for close monitoring, or else for consolidation measures such as adjuvant radiotherapy (to minimize local relapse) and/or immune therapies such as vaccines (to reduce distant failure). High-stability high-sensitivity tumors located in the top right distribution, such as testicular tumors, may of course prove chemocurable. In contrast, tumors clustering in the lower left (such as anaplastic thyroid cancer) harbor heavy mutations burdens dysregulating both genetic stability and apoptosis, and may therefore be assumed to prove challenging no matter how well managed.

These recessive gene defects may remain remediable using molecule-targeted inhibitors of the two main signaling cascades relevant to anticancer drugs: the PI3K–AKT–mTOR survival pathway, and the RAS–RAF–ERK proliferation pathway. For a tumor exhibiting low-grade genetic instability but high-grade apoptotic insensitivity, whether primary or acquired [93], inhibition of the former pathway – using an mTOR or HER-family inhibitor, for example, or else by blocking a mutationally activated *PIK3CA* gene product – is currently more technically feasible than attempting tumor-specific suppressor gene replacement

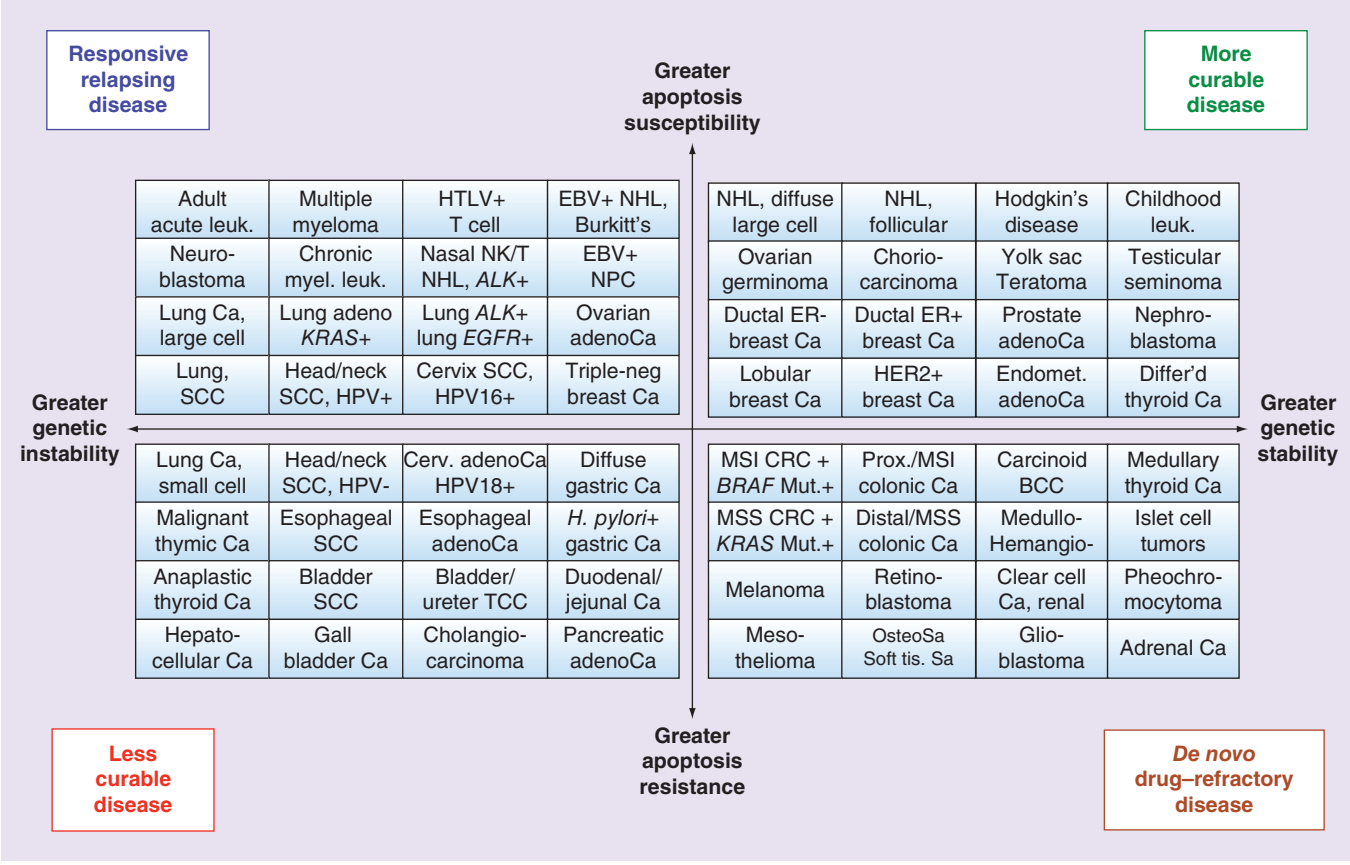

Figure 5. Prototype periodic table of typical organ-specified human cancers classified as a function of genetic stability (x axis) versus apoptotic susceptibility (y axis). Relative positioning of individual tumor types is based on multiple factors, including characteristic clinical behavior with or without treatment [75], net mutation load typical of the tumor subtype [66] and historical evidence of chemosensitivity as inferred by response rates in clinical trials [31].
AdenoCa: Adenocarcinoma; BCC: Basal cell carcinoma; Ca: Cancer/carcinoma; Cerv.: Cervix; CRC: Colorectal carcinoma; Differ'd: (Well) differentiated; EBV: Epstein–Barr virus; Endomet.: Endometrial; ER: Estrogen receptor; HPV: Human papillomavirus; Leuk.: Leukemia; MSI: Microsatellite instability (high); MSS: Microsatellite-stable; Mut.: Mutation; Myel: Myeloid; NHL: Non-Hodgkin's lymphoma; NPC: Nasopharyngeal carcinoma; Prox.: Proximal; Sa: Sarcoma; SCC: Squamous cell carcinoma; TCC: Transitional cell (urothelial) carcinoma; Tis.: Tissue; Triple-neg: Triple-negative, carcinoma *in situ*.

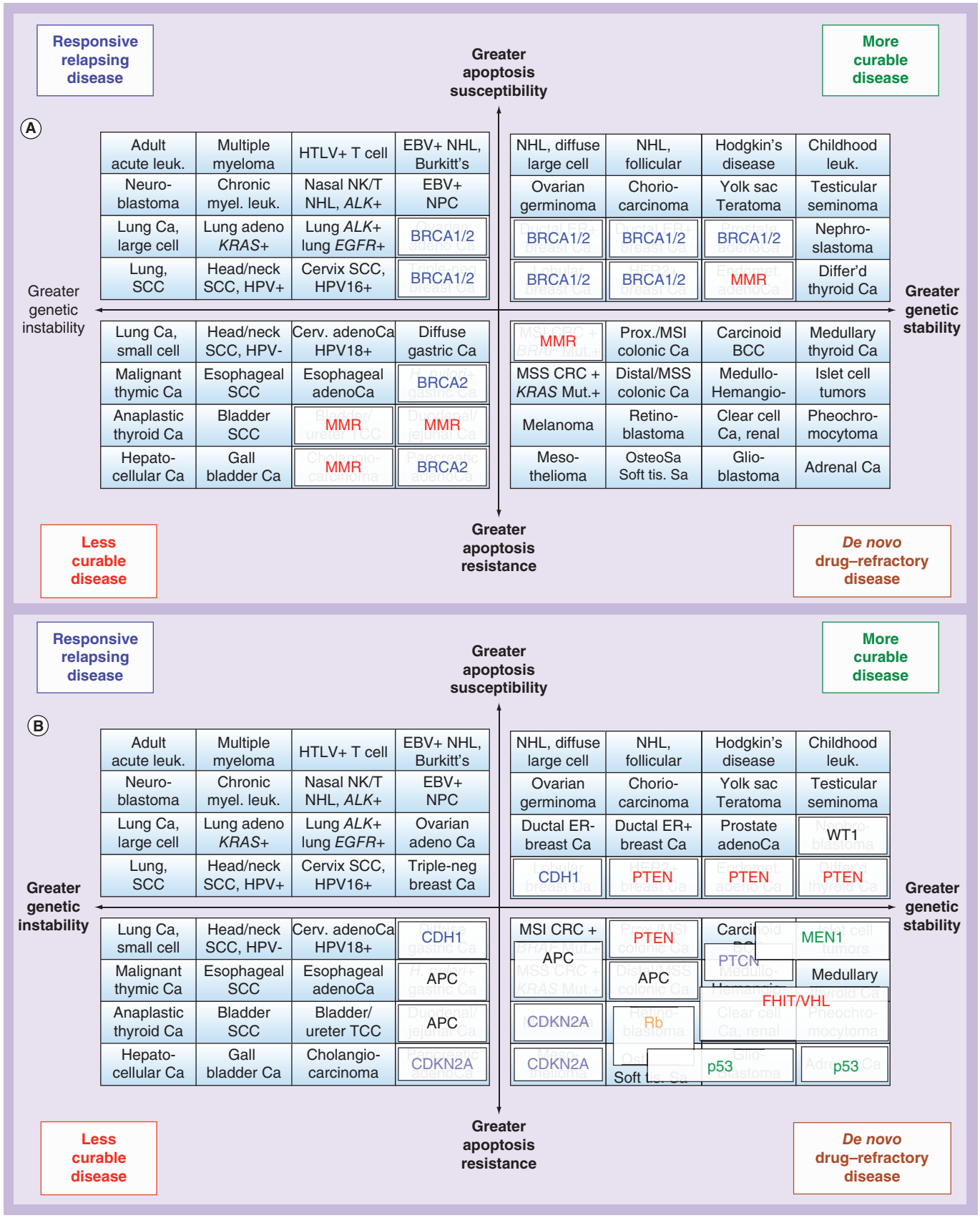
Responsive relapsing disease
Greater apoptosis susceptibility
More curable disease
A
Adult acute leuk.
Multiple myeloma
HTLV+ T cell
EBV+ NHL, Burkitt's
Neuro-blastoma
Chronic myel. leuk.
Nasal NK/T NHL, ALK+
EBV+ NPC
Lung Ca, large cell
Lung adeno KRAS+
Lung ALK+ lung EGFR+
BRCA1/2
Lung, SCC
Head/neck SCC, HPV+
Cervix SCC, HPV16+
BRCA1/2
NHL, diffuse large cell
NHL, follicular
Hodgkin's disease
Childhood leuk.
Ovarian germinoma
Chorio-carcinoma
Yolk sac Teratoma
Testicular seminoma
BRCA1/2
BRCA1/2
BRCA1/2
Nephro-blastoma
BRCA1/2
BRCA1/2
MMR
Differ'd thyroid Ca
Greater genetic instability
Greater genetic stability
Lung Ca, small cell
Head/neck SCC, HPV-
Cerv. adenoCa HPV18+
Diffuse gastric Ca
Malignant thymic Ca
Esophageal SCC
Esophageal adenoCa
BRCA2
Anaplastic thyroid Ca
Bladder SCC
MMR
MMR
Hepato-cellular Ca
Gall bladder Ca
MMR
BRCA2
MMR
Prox./MSI colonic Ca
Carcinoid BCC
Medullary thyroid Ca
MSS CRC + KRAS Mut.+
Distal/MSS colonic Ca
Medullo-Hemangio-
Islet cell tumors
Melanoma
Retino-blastoma
Clear cell Ca, renal
Pheochro-mocytoma
Meso-thelioma
OsteoSa Soft tis. Sa
Glio-blastoma
Adrenal Ca
Less curable disease
Greater apoptosis resistance
De novo drug–refractory disease
Responsive relapsing disease
Greater apoptosis susceptibility
More curable disease
B
Adult acute leuk.
Multiple myeloma
HTLV+ T cell
EBV+ NHL, Burkitt's
Neuro-blastoma
Chronic myel. leuk.
Nasal NK/T NHL, ALK+
EBV+ NPC
Lung Ca, large cell
Lung adeno KRAS+
Lung ALK+ lung EGFR+
Ovarian adeno Ca
Lung, SCC
Head/neck SCC, HPV+
Cervix SCC, HPV16+
Triple-neg breast Ca
NHL, diffuse large cell
NHL, follicular
Hodgkin's disease
Childhood leuk.
Ovarian germinoma
Chorio-carcinoma
Yolk sac Teratoma
Testicular seminoma
Ductal ER-breast Ca
Ductal ER+ breast Ca
Prostate adenoCa
WT1
CDH1
PTEN
PTEN
PTEN
Greater genetic instability
Greater genetic stability
Lung Ca, small cell
Head/neck SCC, HPV-
Cerv. adenoCa HPV18+
CDH1
Malignant thymic Ca
Esophageal SCC
Esophageal adenoCa
APC
Anaplastic thyroid Ca
Bladder SCC
Bladder/ ureter TCC
APC
Hepato-cellular Ca
Gall bladder Ca
Cholangio-carcinoma
CDKN2A
MSI CRC + APC
PTEN
MEN1
PTCN
MSS CRC + KRAS Mut.+
APC
Medullary
FHIT/VHL
CDKN2A
Rb
CDKN2A
p53
p53
Less curable disease
Greater apoptosis resistance
De novo drug–refractory disease

Figure 6. Distribution of causal suppressor gene mutations within the periodic table (see facing page). (A) Tumor distribution of germline caretaker gene mutations in familial cancer syndromes. BRCA1/2, *BRCA1* or *BRCA2* gene (mutations); MMR, mismatch repair gene, for example, *MLH1* or *MSH2,* gene (mutations). **(B)** Tumor distribution of germline gatekeeper gene mutations in familial cancer syndromes.

AdenoCa: Adenocarcinoma; Ca: Cancer/caricinoma; Cerv.: Cervix; CRC: Colorectal cancer; Differ'd: (Well) differentiated; EBV: Epstein–Barr virus; Endomet.: Eendometrial; Hemangio: Hemangioendotheliomatosis; HPV: Human papillomavirus; Leuk.: Leukemia; Medullo: Medulloblastoma; MSS: Microsatellite-stable; MSI: Microsatellite-unstable; Myel.: Myeloid; NHL: Non-Hodgkin's lymphoma; Osteo Sa: Osteosarcoma; Prox.: Proximal; Sa: Sarcoma; SCC: Squamous cell carcinoma; TCC: Transitional cell carcinoma; Tis. Tissue; Triple-neg: Triple-negative, carcinoma *in situ*.

therapy. For a tumor characterized by low-grade apoptotic insensitivity but high-grade genetic instability, on the other hand, a logical strategy could involve sustained blockade of the RAS–RAF–ERK pathway, given that reduction of DNA replication (whether by specific blockade of an ERK-upregulating mutation, or by more general induction of dormancy) should slow fixation of new mutations driving disease progression. Of these ERK-inhibitory options, dormancy (replicative arrest without apoptosis) may remain the holy grail of survival-prolonging strategies, unlike cytotoxic [94] and kinase-inhibitory drugs which select rapidly for resistance via mutation or transmodulatoion [95]. For certain responsive subsets of melanoma and renal cell carcinoma, immunotherapies now appear to represent such a nonselecting (that is, survival-enhancing) approach [96]. Inhibition of targetable non-mutant signaling pathways downstream of dysfunctional suppressors, such as the activated Smo pathway downstream of loss-of-function *PTCH* mutations, may also be useful strategies for effecting a response, though it remains unclear whether durable survival benefits (that is, lack of rapid selection for resistance) can be expected when permissive genetic dysfunctionality lies upstream of a target pathway.

Central to the vision of personalized oncology is the expectation that molecular genotyping will prove over time to be of more predictive therapeutic value than is traditional morphology alone. Consider, for example, the still futuristic prospect of rational combination targeted drug therapy, the plausibility of which has been strengthened in recent times by some clinical trials [97] and case reports [98]. Such approaches may come to be best designed upfront by supplementing a tumor's oncogenic target with its suppressor dysfunctionality profile. For example, tumors with both *HER2* amplification and *PTEN* deletion – whether in the adjuvant or relapsed clinical settings – could in theory be associated with longer survival outcomes if treated with a HER2-targeted therapy supplemented by a low-dose mTOR inhibitor. On the other hand, tumors with major genetic defects affecting both suppressor pathways (e.g., an *hDM2* amplification indicating p53 pathway dysfunction, plus a *PTEN* deletion – such as might be expected to select for additional driver mutations such as *KRAS*) may identify patient subgroups that uniquely benefit from more aggressive use of combination targeted regimens.

Problems of the prototype

It was noted in the introduction to this article that scientific progress is often made through the observation of homologies or continuities [1]. The validity or otherwise of 2D tumor profiling as depicted in **Figure 5** may thus be tested in the first instance by asking whether genotypic lesions of the same functional class exhibit corresponding phenotypic (e.g., apoptotic) continuities. Since it is known that certain inherited mutations of caretaker or gatekeeper suppressor genes predispose to a defined spectrum of familial tumor types, and that the same suppressor gene mutations are often acquired in sporadic nonfamilial tumors of the same histology [99], a tissue-specific carcinogenic driver role for these mutations is implied. Consistent with this, the distribution of organ-specific tumors causally linked to caretaker (genetic stability) gene mutations – such as *BRCA1/2*, MMR genes, *FANC* or *ATM* – forms a contiguous band bisecting the Table from top right to bottom left **(Figure 6A)**.

As also predicted, tumors linked to familial gatekeeper defects cluster in the lower right (apoptosis defective) corner **(Figure 6B)**. However, this localization raises a query as to the lack of symmetry between the two gene-tumor sets – that is, why does the grouping of 'caretaker tumors' not polarize to the opposite top-left, as would be anticipated for high-instability tumors? This anomaly suggests that even

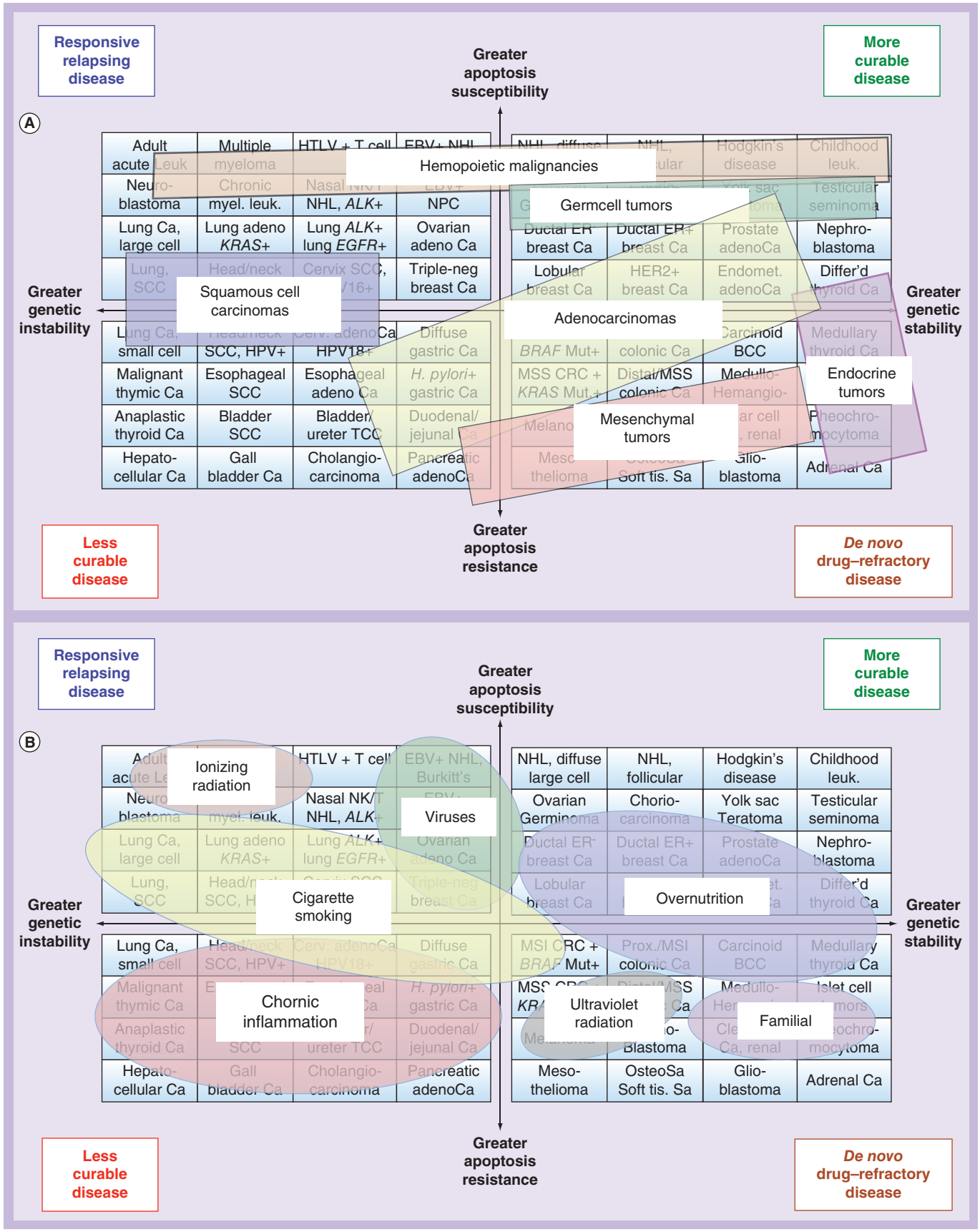
Responsive relapsing disease
More curable disease
Greater apoptosis susceptibility
A
Hemopoietic malignancies
Germcell tumors
Squamous cell carcinomas
Adenocarcinomas
Endocrine tumors
Mesenchymal tumors
Greater genetic instability
Greater genetic stability
Adult acute Leuk
Multiple myeloma
HTLV + T cell
EBV+ NHL
NHL, diffuse large cell
NHL, follicular
Hodgkin's disease
Childhood leuk.
Neuro-blastoma
Chronic myel. leuk.
Nasal NK/T NHL, ALK+
EBV+ NPC
Ovarian Germinoma
Chorio-carcinoma
Yolk sac Teratoma
Testicular seminoma
Lung Ca, large cell
Lung adeno KRAS+
Lung ALK+ lung EGFR+
Ovarian adeno Ca
Ductal ER- breast Ca
Ductal ER+ breast Ca
Prostate adenoCa
Nephro-blastoma
Lung, SCC
Head/neck SCC, HPV+
Cervix SCC, HPV16+
Triple-neg breast Ca
Lobular breast Ca
HER2+ breast Ca
Endomet. adenoCa
Differ'd thyroid Ca
Lung Ca, small cell
Cerv. adenoCa HPV18+
Diffuse gastric Ca
BRAF Mut+ colonic Ca
Carcinoid BCC
Medullary thyroid Ca
Malignant thymic Ca
Esophageal SCC
Esophageal adeno Ca
H. pylori+ gastric Ca
MSS CRC + KRAS Mut+
Distal/MSS colonic Ca
Medullo-Hemangio-
Anaplastic thyroid Ca
Bladder SCC
Bladder/ ureter TCC
Duodenal/ jejunal Ca
Melano
ar cell , renal
Pheochro-mocytoma
Hepato-cellular Ca
Gall bladder Ca
Cholangio-carcinoma
Pancreatic adenoCa
Meso-thelioma
Soft tis. Sa
Glio-blastoma
Adrenal Ca
Greater apoptosis resistance
Less curable disease
De novo drug–refractory disease
B
Ionizing radiation
Viruses
Cigarette smoking
Overnutrition
Chornic inflammation
Ultraviolet radiation
Familial
MSI CRC + BRAF Mut+
Prox./MSI colonic Ca
Islet cell
Blastoma
Ca, renal
Osteo Sa

Figure 7. Etiologic continuities within the prototype periodic table. (A) Histological and morphological continuities within the table. **(B)** Predisposing carcinogenic exposures as a function of tumor distribution, again confirming continuity within phenotypic subgroups. AdenoCa: Adenocarcinoma; Ca: Cancer/caricinoma; Cerv.: Cervix; CRC: Colorectal cancer; Differ'd: (Well) differentiated; EBV: Epstein–Barr virus; Endomet.: Eendometrial; Hemangio: Hemangioendotheliomatosis; HPV: Human papillomavirus; Leuk: Leukemia; Medullo: Medulloblastoma; MSS: Microsatellite-stable; MSI: Microsatellite-unstable; Myel.: Myeloid; NHL: Non-Hodgkin's lymphoma; Osteo Sa: Osteosarcoma; Prox.: Proximal; Sa: Sarcoma; SCC: Squamous cell carcinoma; Tis.: Tissue; TCC: Transitional cell carcinoma; Triple-neg: Triple-negative, carcinoma *in situ*.

classical caretaker-dependent tumors – such as primary breast, ovarian or colorectal cancers – incur only intermediate genetic instability as a result of caretaker mutations alone, with an additional burden of genome-destabilizing mutations (including those involving gatekeepers such as *TP53* [100]) acquired during tumor progression. The implication that caretaker mutations alone confer a mild phenotype, as expected from their low embryonic lethality [40], is consistent with the high prevalence of heterozygous defects [101]. Hence, whereas apoptotic resistance may be predicted largely by the number of gatekeeper pathway dysfunctions alone, genetic instability may be better quantified by the sum of caretaker and gatekeeper mutations. Consistent with this, a positive slant (top right to bottom left) biases the distribution of common age-dependent cancers in this working 2D dysfunctionality map.

Model-testing continuities may also be interrogated at the levels of tumor histology or etiology. **Figure 7A** shows consistent patterns united by histopathology, including endocrine (glandular) tumors localizing to the high-stability/low-apoptosis lower right; mesenchymal and sarcomatous tumors clustering along the bottom; adenocarcinomas co-localizing in a caretaker-like (see **Figure 6A**) distribution, consistent with intermediate losses of both stability and apoptotic susceptibility; and hematological malignancies forming a cluster high in apoptotic sensitivity (that is, a relatively low burden of gatekeeper mutations) but highly variable in genetic stability. Similarly, **Figure 7B** shows dysfunctionality aggregations based on tumor etiology. A lower left cluster of poor-prognosis tumors with combined defects in apoptosis and stability is linked to chronic inflammation; a smoking-inducible tumor cluster is characterized by high instability but lower apoptotic resistance; whereas a tumor cluster with intermediate apoptotic resistance but high genetic stability is attributable to overnutrition, consistent with tumorigenesis via insulin-mediated downregulation of stress-induced cell death [102].

The present model remains a test of concept, and certain limitations are clear. First, no claim is made that organ-specified cancer types – 'breast cancer' or 'ovarian cancer', for example – necessarily display greater phenotypic continuity than tumors of different organs but similar molecular pathologies [103]. In theory, individual tissue-specific tumors could map to any position on the apoptosis/stability graph, rather than remaining restricted to the 'average' organ-specific tumor coordinates shown here for convenience. Moreover, a given patient will not have a single set of tumor coordinates valid for their entire disease; the genetically defined dysfunctionalities will change (proof of which will require repeat genomic sampling) depending on whether the disease is untreated/pretreated, primary/secondary, liver metastasis/bone metastasis, and so on.

Second, there is no *a priori* reason to assume that a periodic table of human cancers must be rectangular in shape, notwithstanding its 2D coordinates; indeed, like other dual-parameter distributions (e.g., the north-south vs east-west compass), the most plausible shape for a 2D representation of the cancer universe is circular. Similarly, there is no rationâle for assuming such a Table to be homogeneously filled; a distribution encompassing all cancer genomes might be expected to be peppered with 'volcanos' (hotspots) of common molecular pathologies, contrasting with internal 'lakes' and peripheral 'bays' of coordinates lacking tumor subtypes. Any digital periodicity ascribed to tumors would be a functional approximation based on grading cut-offs and scoring criteria as noted above to guide clinical decision-making.

Third, there remain both semantic and technical hurdles in quantifying 'genetic instability' and 'apoptotic resistance' for purposes of treatment strategizing. For example, colorectal cancers with MMR defects exhibit microsatellite instability (MSI) and hence a high total number of mutations [66]; yet these tumors are characterized by a better prognosis than microsatellite-stable tumors with fewer mutations [75] – presumably due to confounding by a paradoxically higher frequency in the latter

subset of critical mutations such as those affecting *TP53* and *KRAS* [104]. A software program quantifying only mutation number, without correction for mutation impact, is therefore unlikely to suffice for clinically useful phenotyping. Similarly, the notion of 'apoptosis resistance' remains difficult to define: for example, low-grade ER-rich lobular breast cancers tend to be sensitive to hormonal manipulation yet resistant to cytotoxic therapy [105], while many other slow-proliferating well-differentiated tumors are likewise 'chemoresistant' yet have a good prognosis. Conversely, poorly differentiated tumors (e.g., small-cell lung cancer) are often chemosensitive in terms of initial response, yet confer poor survival due to rapid resistance [106]. Similarly, chemoresistant tumors such as clear-cell renal carcinoma or melanoma may respond to targeted drug therapies, indicating that DNA damage is not the only apoptotic trigger [107]. Metastatic lesions from such tumors may also be indolent for years, exhibit spontaneous regression and/or prove responsive to immunotherapies [108]. Strategies to predict damage-independent and immune-mediated apoptosis are thus needed to prevent such tumors being wrongly written off as apoptosis-resistant due only to lack of cytotoxic 'response'.

Conclusion

Like the first map of the world from ancient Babylon [109], this draft periodic table for cancer envisages ongoing refinements; the prototype is only a first approximation, analogous to the popular (but inaccurate) Mercator's projection of the world [110]. The premiss underpinning this 2D map is that just two of the postulated 'hallmarks of cancer' [59] (evading apoptosis and unstable DNA, as identified by the heritable cancer susceptibility analyses of Kinzler and Vogelstein [41]) are often primary – the remainder (such as, angiogenesis or Warburgism; direct effects of impaired apoptosis such as metastasis; or extrinsic pathologies such as inflammation) tend to be secondary phenotypic events acquired during tumor evolution. Since genetic instability and apoptotic insensitivity give rise to the key oncologic problems of tumor progression and drug resistance, quantification of these dysfunctionalities as illustrated here should prove important in predicting the severity of the associated clinical problems.

More nuanced and adaptive strategies are needed for effective anticancer drug treatment. Assuming further advances in genomic software, it should become possible in the future for oncologists to incorporate the above measures of tumor dysfunction into long-term treatment plans, and to track the dynamic trajectory of these dysfunctions by sequential tumor samplings throughout disease progression. Such dysfunction-adjusted treatment strategies may help to slow the progression of cancers to drug resistance, and thereby enhance patient survival and life quality.

Future perspective

Any human conception of cancer dysfunctionality must be simpler than the molecular reality, raising the possibility that addition of a third dimension to the present 2D cancer model could prove more accurate. If so, which 3D parameter would add the most value to future clinical decision making? **Figure 2** raises the obvious candidate of oncogenic drive, but this focus may prove of limited therapeutic actionability and patient benefit – first, because only a minority of tumors contain targetable drivers, and second, because pre-existing suppressor gene defects ensure that rapid responses to targeted drugs select for rapid resistance, perhaps yielding only trivial survival benefits. Hence, the priority of this discussion is to seek novel therapeutic approaches to hitherto neglected pathways.

A future value-adding 'third-dimensional' drug pathway could modulate the micrometastatic niche in the (neo)adjuvant or preventive settings. Cancers may metastasise to diverse tissues, but it has long been noted that a subset of tumors restricts its spread to a single nonprimary organ: for example, HER2-positive breast cancers spread more to the brain [111] where HER-family receptors and ligands are abundant [112], whereas breast cancers low in E-cadherin (whether due to primary *CDH1* mutations [113] or secondary Snail-dependent *trans*-repression [114]) often metastasize to serosal surfaces [115], mimicking ovarian cancer peritoneal metastases that upregulate α_5-integrin following E-cadherin loss [116]. These examples suggest that certain drug classes so far judged 'inactive' by response-based clinical trial criteria – such as protease inhibitors, G-protein-coupled receptor blockers and cancer vaccines – could have survival-prolonging value when used as adjuvants to inhibit micrometastasis viability in specific organs. If so, a future periodic table could conceivably include predictors of tissue homing as a third dimension.

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EXECUTIVE SUMMARY

- Current oncologic drug decision-making algorithms remain based on two variables alone – clinical observation/trials (retrospective), and oncoprotein expression (prospective) – but do not yet incorporate either tumor dysfunctionality (suppressor defects) or biomarker-defined microenvironmental dysregulation into this analysis.
- The Vogelstein–Kinzler model of heritable cancer predispositions suggests that the two most fundamental dysfunctionalities of human tumors are genetic instability (as exemplified by caretaker suppressor gene mutations) and apoptotic resistance (as exemplified by gatekeeper gene mutations). The beauty of this simple paradigm may be more readily translated to clinical decision-making than is the more complex Weinberg–Hanahan multi-hallmark model of molecular oncogenesis, which may be more useful for basic research.
- The clinical benefit (actionability) of tumor genome read-outs should be improved by developing techniques to differentially compute the relative severities of genetic instability and apoptotic resistance in specific tumor biopsies.
- Such software-based quantifications of tumor dysfunctionality may be visually represented in a periodic table that is intuitive enough to guide strategic treatment decisions aimed at circumventing the central clinical problems of drug resistance and tumor progression.
- Examples of molecularly-customized strategies to mitigate resistance and progression include maintenance drug treatments to downregulate the PI3K–AKT–mTOR (cell survival) and RAS–RAF–ERK (DNA replication) signaling pathways, respectively.

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