This manuscript is contextually identical with the following paper: Apari P., Müller V. (2019). Paradoxes of tumour complexity: somatic selection, vulnerability by design, or infectious aetiology? Biol Rev Camb Philos Soc 94(3):1075-1088. doi: 10.1111/brv.12490

Paradoxes of tumour complexity: somatic selection, vulnerability by design, or infectious aetiology?

Péter Apari[1,2] and Viktor Müller[1,2,*]

[1]*Institute of Biology, Eötvös Loránd University, Pázmány P. s. 1/C, 1117 Budapest, Hungary*

[2]*Evolutionary Systems Research Group, MTA Centre for Ecological Research, Klebelsberg Kuno u. 3, 8237 Tihany, Hungary*

[*]Author for correspondence at address 1 (Tel: (+36) 1 381 2187; Fax: (+36) 1 381 2188; E-mail: mueller.viktor@gmail.com).

ABSTRACT

The aetiology of cancer involves intricate cellular and molecular mechanisms that apparently emerge on the short timescale of a single lifetime. Some of these traits are remarkable not only for their complexity, but also because it is hard to conceive selection pressures that would favour their evolution within the local competitive microenvironment of the tumour. Examples include ‘niche construction’ (re-programming of tumour-specific target sites) to create permissive conditions for distant metastases; long-range feedback loops of tumour

growth; and remarkably ‘plastic’ phenotypes (e.g. density-dependent dispersal) associated with metastatic cancer. These traits, which we term ‘paradoxical tumour traits’, facilitate the long-range spread or long-term persistence of the tumours, but offer no apparent benefit, and might even incur costs in the competition of clones within the tumour. We discuss three possible scenarios for the origin of these characters: somatic selection driven by specific selection regimes; non-adaptive emergence due to inherent vulnerabilities in the organism; and manipulation by putative transmissible agents that contribute to and benefit from these traits. Our work highlights a lack of understanding of some aspects of tumour development, and offers alternative hypotheses that might guide further research.

Key words: evolutionary medicine, cancer, tumour evolution, paradoxical tumour traits, somatic selection, oncogenic agents.

CONTENTS

I. INTRODUCTION

The complexity of tumour phenotypes poses a puzzle from the perspective of evolutionary biology (Gatenby & Gillies, 2008; Davies & Lineweaver, 2011; Thomas *et al*., 2017). In the time span of at most a few decades, the descendants of a normal cell of a multicellular organism can apparently develop complex ways to manipulate neighbouring (Whiteside, 2008; Hanahan & Coussens, 2012) or even distant (Kaplan *et al*., 2005; Peinado *et al*., 2017) cells to promote their growth and spread, to form organized structures that have been likened to 'organs' (Egeblad, Nakasone & Werb, 2010), and to break out of the strict control of growth in both space and time. The current paradigm of 'internal Darwinism' (Davies & Lineweaver, 2011) holds that these complex traits emerge by somatic evolution, driven by predominantly cell-level selection, at the expense of the multicellular organism (Nowell, 1976; Merlo *et al*., 2006; Greaves & Maley, 2012). However, it has been argued that the apparent 'ease' of evolving these traits must require special conditions that predispose to the convergent evolution of cancer in the many independent instances (Gatenby & Gillies, 2008; Davies & Lineweaver, 2011; Thomas *et al*., 2017).

The power of evolution often defies intuition, and tends to be underestimated. However, in the evolution of cancer within the host, it is not only the rate, but also the nature of adaptation that needs to be explained. While somatic evolution towards 'selfish' traits, such as clonal immortality and unlimited cell division, is so evident that it is the general success of the

control mechanisms that requires explanation (Michod, 2003; Pradeu, 2013), the origin of some cancer traits cannot be explained easily by the simple forces of cell-level selection, or even by the evolution of cooperation between tumour cell clones (Axelrod, Axelrod & Pienta, 2006; Cleary *et al*., 2014).

We structure our paper by presenting first a list of phenomena that we think pose a problem within the established paradigm of somatic selection, and then proceeding to discuss three possible, not mutually exclusive, explanations that might resolve the problems.

II. PARADOXICAL TUMOUR TRAITS: AN EVOLUTIONARY CONUNDRUM

Below we list some examples of the tumour traits that we term 'paradoxical', because they appear to contradict the logic of somatic selection. We discuss the difficulty of explaining each of these traits with local selection pressure.

(1) Distant niche construction

Kaplan *et al*. (2005) introduced the concept of the 'pre-metastatic niche' as a "tumour-conducive microenvironment at distant sites that arises in response to factors released by the primary tumour" (Murgai *et al*., 2017, p. 1176), often displaying characteristic site specificity according to tumour type (Hoshino *et al*., 2015; Peinado *et al*., 2017). For example, pancreatic cancer has been observed to initiate pre-metastatic niche formation in the liver (Costa-Silva *et al*., 2015), breast cancer in the bones (Cox *et al*., 2015), renal cancer in the lung (Grange *et al*., 2011), and melanoma in multiple tissues (Kaplan *et al*., 2005; Peinado *et al*., 2012). The effect of the tumour cells on distant tissues is mediated typically by exosomes released by the tumour (Costa-Silva *et al*., 2015; Grange *et al*., 2011; Peinado *et al*., 2012; Hoshino *et al*., 2015). The evolutionary conundrum arises because sharing the benefit of the effect (having daughter cells that successfully colonize the constructed permissive

environment) is not linked to contributing to its cost (production of exosomes) (Fig. 1A). No mechanism is known at the moment that would restrict the colonization of the affected distant sites to the progeny of those cells that 'paid the cost' for distant niche construction at the site. However, if the cells producing the effect enjoy the same benefit as 'free-riding' competitors, then the cost of the effect incurs a selective disadvantage, and the capacity for distant niche construction will be selected against in the within-host somatic evolution of the tumour (Tabassum & Polyak, 2015). Therefore, this trait is not only complex, but appears to contradict the expected direction of selection in tumour evolution. The systemic suppression of anti-metastatic immune responses by a tumour-induced mechanism, observed in a mouse model of breast cancer (Coffelt *et al*., 2015), follows the same evolutionary logic.

(2) Long-range positive feedback loops of tumour growth

In mouse models of prostate (Park *et al*., 2013) and lung (Engblom *et al*., 2017) cancer, primary tumour cells were able remotely to activate bone-resident osteoblasts, which then triggered the production of specific tumour-infiltrating myeloid cells that promoted the growth of the primary tumour. Similar to distant niche construction, the benefits of these long-range feedback effects seem to be shared equally between cells that contribute to the cost and cells that do not, thus presenting the same evolutionary conundrum (Fig. 1B): if producer and non-producer cells compete with each other, the cost of production burdens only the former, and all else being equal, these cells should be lost.

Both distant niche construction and long-range positive feedback loops pose a qualitative problem for explanations by somatic selection: not only do they require intricate mechanisms that might be hard to evolve, they are expected to be selected against in the within-host competition of tumour cell clones. In the following we list some further cancer traits that pose a quantitative problem: while they do confer a selective advantage within the host, their

evolution might require specific selection regimes or a timescale longer than that of within-host tumour progression.

(3) Local niche construction

The growth of tumours is often facilitated by the re-programming of neighbouring cells towards tumour-promoting phenotypes to create a supportive local microenvironment (Whiteside, 2008; Hanahan & Coussens, 2012). Tumour-associated macrophages (TAMs) (Pollard, 2004) and carcinoma-associated fibroblasts (CAFs) (Kalluri & Zeisberg, 2006) are characteristic examples of the re-programmed cell types. Because the benefits are indirect (non-cell-autonomous), the evolution of these mechanisms is also affected by the problem of shared 'public goods' (Tomlinson & Bodmer, 1997; Nagy, 2004), although to a lesser extent than in the case of the long-range mechanisms listed above. The diffusion of both those secreted factors that re-program the microenvironment, and of the tumour-promoting factors produced by the manipulated cells, weakens the association between paying the cost and reaping the benefit of the niche construction mechanisms (Fig. 1C). This partial decoupling reduces, or, depending on the balance of cost and shared benefit, might even fully abrogate the selective advantage of these traits. In addition, the ability to re-program other cells (of several cell types) is likely to be more difficult to evolve than altering the metabolism, growth or motility of the cancer cell itself, as is associated with the general hallmarks of cancer (Hanahan & Weinberg, 2000). As the non-tumour cells in the microenvironment are typically not mutated (Qiu *et al*., 2008), re-programming must rely on epigenetic mechanisms, while the transformation of the cancer cells can take advantage of genetic changes as well. Furthermore, all manipulations must occur through cell-to-cell communication, which is inevitably constrained compared with regulatory pathways within the cell. We finally note that there is a smooth gradient between 'local' and 'distant' niche construction, e.g. the

suppression of immune cells by tumour-derived exosomes can act both locally and at the systemic level (Whiteside, 2016).

(4) Metastatic potential – phenotypic plasticity

In addition to the non-cell-autonomous tumour mechanisms discussed above, metastasis, the ability of tumours to colonize distant tissues (and a hallmark of malignant progression), is also in apparent contradiction with the selection forces that act in the local competitive microenvironment (Bernards & Weinberg, 2002). The traits required for cellular motility and efficient invasion are likely to differ from, and might well conflict with those that aid competition within the local microenvironment. It has been argued (Gatenby & Gillies, 2008), and also supported by simulations (Aktipis, Maley & Pepper, 2012), that in advanced stages of cancer, metastasis might confer a selective advantage by enabling cells to move away from a local site where tumour growth has depleted local resources. However, there are two potential problems with this reasoning. First, the development of micrometastases and circulating tumour cells seems to occur already in the early, asymptomatic stages of cancer progression (Pantel, Alix-Panabières & Riethdorf, 2009; Friberg & Nystrom, 2015), when the local microenvironment might not yet be depleted. Second, while selection for local growth occurs continuously through all cell divisions in a tumour, the traits that promote distant metastases are exposed to selection only once in each cycle of metastasis formation. If there is conflict (evolutionary trade-off) between the traits required for local growth and metastasizing, the former is likely to dominate the evolutionary dynamics.

To exacerbate the problem, the evolution of metastatic potential is often not a simple directional shift from fixed towards mobile cells, but involves the evolution of complex phenotypic plasticity. Epithelial tumour cells can undergo a reversible epithelial-to-mesenchymal transition to alternate between motile/invasive and local growth behaviour

(Batlle & Clevers, 2017), and density-dependent tumour cell migration (an efficient adaptive dispersal strategy) has been observed with several tumour cell lines (Jayatilaka *et al*., 2017). The evolution of such complex strategies is unlikely in the timespan of tumour progression within the host (Arnal *et al*., 2015). Although we note that cell lines have evolved by serial passages over extended periods; density-dependent motility remains to be demonstrated in primary tumour cells.

Thus far we have discussed why simple adaptationist explanations fail to account for the recurrent emergence of these complex tumour traits (summarized in Table 1). Below (see also Table 1) we investigate what specific circumstances might drive the somatic selection of these characters, and then we proceed to alternative explanations.

III. SPECIFIC SELECTION REGIMES

It is not impossible to conceive selection regimes that could drive the emergence of any one paradoxical tumour trait; however, each of the traits requires specific conditions to be under positive selection within the organism.

Easier to tackle are the traits that involve quantitative problems for somatic selection. The ability for local niche construction can be selected if the action radius of the tumour-promoting effect is sufficiently small, such that the net benefit of a producer cell (locally strong positive effect minus the cost of production) exceeds the benefit, decreasing with distance, that non-producer cells in the neighbourhood receive. Beyond this trivial possibility, it has been shown by mathematical modelling that producer cells can persist in a tumour if the benefit from the tumour-promoting effect scales with the concentration of producers according to a biologically plausible (sigmoid) non-linear function (Archetti, 2013). Such scaling creates frequency-dependent selection that favours producers when they are rare, which results in stable coexistence of producer and non-producer cells. This scenario has

received experimental support regarding the production of growth factors that affect both producer and non-producer cells (Archetti, Ferraro & Christofori, 2015). It remains to be seen whether the concept applies also to the more complex functions of local niche construction, including the reprogramming of neighbouring stromal cells.

Regarding metastatic potential involving adaptive phenotypic plasticity, we argued that such complex dispersal strategies are unlikely to evolve when metastases are rare events, and competition for local growth is likely to be the dominant force of selection. By contrast, repeated cycles of metastases could impose an alternating selection regime capable of selecting for adaptive dispersal. 'Metastases of metastases' have long been known to occur (Roth, Silverstein & Morton, 1976), and 'self-seeding', i.e. metastases between existing tumours (Gundem *et al*., 2015; Kim *et al*., 2009), might enable covert chains of serial metastases. Further research is needed to determine whether such chains indeed precede the development of clinically manifest metastatic cancer. Furthermore, metastatic ability might evolve as a by-product of local motility if the latter can be selected, e.g. due to metabolic demands (Aktipis *et al*., 2012). However, it then needs to be explained that at least some tumour types appear to bypass nearby dissemination, and form distant metastases from the beginning (Pantel & Brakenhoff, 2004).

Long-range mechanisms like distant niche construction and long-range positive feedback loops present even harder 'qualitative' hurdles for the somatic selection paradigm. In these cases, the production (and cost) of the promoting effects appears to be completely decoupled from the benefit that arises either at a distant anatomical site or as a systemic, and therefore equally distributed, effect. This is a fundamental difference from locally acting mechanisms, in which some of the benefit remains private to the producer cells, and sharing is incomplete. Notably, the mathematical and simulation models that implement competitive interaction events between producer and non-producer cells (Tomlinson & Bodmer, 1997; Archetti,

2013) involve incomplete sharing, and therefore do not apply in this setting. With complete sharing, non-producer cells that equally share the benefit but do not pay the cost will trivially outcompete producer cells, even though the overall growth or spread of the tumour would increase with higher proportion of the producer type.

Distant niche construction and other systemic mechanisms that promote the growth or spread of tumours could be explained by somatic selection only if one of two conditions is fulfilled. Either, (1) the benefits should be targeted preferentially to the cells that contributed to the cost (that is, sharing should be incomplete). In the case of distant niche construction, the cells that produce the re-programming factors should have 'priority' in the colonization of the constructed distant niche; in the case of positive feedback loops, the growth-promoting effect should be targeted selectively to the cells that initiate the effect. However, such linkage has not been demonstrated yet, and it is unclear how it should emerge and why it would not be vulnerable to 'cheater' variants that only carry the trait responsible for reaping the benefit. Alternatively, (2) the mechanisms of long-range effects could be selected if they conferred some local selective advantage, and the distant action emerged as a mechanistically coupled 'fortuitous' side-effect. For example, if distant niche construction depended on the same signals that contribute to the re-programming of the local tumour microenvironment in the primary tumour, then local selection for the latter also would give rise to the former as an 'accidental' by-product. Furthermore, the production of re-programming factors could confer a direct benefit in terms of the ability to metastasize, if circulating producer cells were able to facilitate their own engraftment by locally manipulating the anatomical site where they arrive. Repeated cycles of metastases could then give rise to this ability, and 'distant niche construction' could emerge as a side effect of the local manipulating function (due to systemic diffusion of the effector molecules). Such coupling cannot be excluded for the observed long-range positive feedback loops either, and it has also been proposed to explain the evolution of

metastatic potential (Bernards & Weinberg, 2002). However, as far as we are aware, there is at the moment no empirical support for this scenario. In a mouse model of melanoma, the early induction of distant pre-metastatic niches appeared to be uncoupled from lymphangiogenesis at primary lesions (Olmeda *et al*., 2017), which argues against a simple mechanistic coupling between local and distal action mechanisms.

In all, while hypothetical scenarios based on somatic selection can be devised for all 'paradoxical tumour traits', each of these scenarios requires specific conditions to apply (Table 1). In particular, the by-product scenario of long-range effects requires coupling between local and distal effects, which cannot be an evolved property of the tumour, but might exist only as an accidental 'enabling constraint', encoded in the responsiveness of non-tumour cells to molecular signals. In the next section we ask whether such reliance on non-selected features can be taken further, assuming inherent vulnerabilities in the organism that can be 'discovered' by essentially neutral evolution of the tumour.

IV. INHERENT VULNERABILITY

Wu *et al*. (2016) pointed out that neutral evolution should always be regarded as the null hypothesis in the study of evolutionary processes, including those involved in cancer. Indeed, while somatic selection clearly plays an important role in tumour progression, genomic analyses have also revealed a surprisingly strong signature of neutral genetic drift in the molecular evolution of tumours (Wu *et al*., 2016). This is not altogether surprising, considering that tumours start from normal cells 'programmed' by organismic evolution to survive in the organism, to perform some (for the organism) useful functions, and generally to exist deeply below the possible proliferative capacity of a eukaryotic cell. As it is evolved brakes, rather than internal limitations that restrict the growth of the cells (Ewald & Swain Ewald, 2013), there might be ample room for mutational divergence that does not severely

jeopardize the fitness of the cell, including the loss of functions originally provided for the organism but dispensable for the cell, but also functions gained. In the early stages of oncogenic selection *sensu* Ewald & Swain Ewald (2013), rare precancerous cells might compete not so much with each other, but with the prevalent non-mutated cells of the surrounding tissues, which could substantially mitigate the selective costs of paradoxical tumour traits. In the following we ask whether neutral evolution might contribute to the emergence of these traits in the absence of (strong) selection.

While the apparently convergent evolution of recurring cancer traits has been likened to 'reinventing the wheel' in each independent instance of cancer (Arnal *et al*., 2015), the evolution of tumour cells has, in fact, easy access to a whole 'catalogue of wheels' embedded in the host genome. Each precancerous cell starts out with the complete genetic arsenal of the multitude of cell types that comprise the host organism, and a vast landscape of cellular functions and complexity can be explored by simple regulatory mutations, or even epimutations [i.e. stable changes in gene expression in the absence of mutations (Flavahan, Gaskell & Bernstein, 2017; Grunau, 2017)].

For example, assembling the molecular machinery of local or distant niche construction 'by chance' seems *a priori* highly unlikely; however, the necessary components might pre-exist in the normal developmental program of the organism, which involves signalling between different cells and tissues to initiate commitment to alternative cell fates. For instance, the re-programming of CAFs might take advantage of a physiological epigenetic program for tissue regeneration (Orimo *et al*., 2005). In turn, phenotypic plasticity in metastases might evolve by co-opting a 'ready-made' genetic mechanism of epithelial–mesenchymal transition that plays a role in embryonic development (Cano *et al*., 2000). Some combinations of such pre-existing components, when 'stumbled upon' in the neutral evolutionary trajectories of the precancerous cell clones, might give rise to non-cell-autonomous tumour-promoting effects

while imposing negligible costs on the cells. In this scenario, selection in the local microenvironment would neither aid, nor hinder the emergence of such traits, and the evolution of tumour traits is shaped not by positive selection acting on the precancerous cells, but by the inherent ('pre-wired') vulnerabilities of the organism, resonating with Weinberg's conjecture: "Maybe the information for inducing cancer was already present in the normal cell genome, waiting to be unmasked" (Weinberg, 2013, p. 92).

Importantly, this 'accidental' scenario for the emergence of complex tumour traits is not incompatible with the recurring nature of particular traits. While the neutral evolution of tumours might encompass diverse trajectories, it is very likely that only a small fraction of these trajectories point towards complex phenotypes that happen to aid the growth or spread of the tumour, and these will then appear repeatedly in the observations. The starting point of the trajectories (the original cell type) might also restrict possibilities, resulting in tumour-type-specific effects, e.g. in the type-specific targeting of metastases that has been highlighted in Paget's classic 'seed and soil' metaphor (Paget, 1889).

To illustrate how inherent vulnerabilities can give rise to apparently complex functions without adaptive evolution, we refer to the recent finding that wound healing can induce tumour growth at both local and distant anatomical sites (Krall *et al*., 2018). Using an experimental model of breast cancer, they demonstrated that wounding triggers a systemic inflammatory reaction that involves the mobilization of myeloid cells that in turn infiltrate tumours and promote tumour growth, probably by suppressing anti-tumour adaptive immunity. All of these steps appear to occur independently of any adaptive traits of the tumour. Furthermore, if the presence of a growing tumour can induce systemic inflammation (which is again plausible without invoking adaptive tumour traits) then the circle closes to form a long-range positive feedback loop of tumour growth. Of note, in this particular case

the components of the ‘paradoxical tumour trait’ might all be pre-wired in the physiology of the organism, not even requiring a search of the trait space by neutral evolution.

To summarize, inherent features of the (healthy) cells and tissues of the organism might present vulnerabilities that can be exploited by the tumours in surprisingly complicated forms. Some vulnerabilities might exist as ‘ready-made’ packages, others might depend on traits accessible to tumours at low or negligible cost, through constrained (high-probability) trajectories of neutral evolution. Similar to the adaptive explanations, these scenarios also require the constellation of rare conditions to apply. In the following we discuss a third possible route to the evolution of ‘paradoxical tumour traits’.

V. PUTATIVE TRANSMISSIBLE AGENTS

A recent analysis estimated that globally about 15% of new cancer cases can be attributed to known infectious agents (Plummer *et al*., 2016), and as yet unidentified oncogenic infections might contribute to further cases of cancer (Ewald, 2009). We propose that some paradoxical tumour traits might be associated with as yet unknown transmissible agents that both benefit from and contribute to at least some stages of carcinogenesis, involving, in particular, the emergence of these complex traits. Under this scenario, it is the ability of the agents to induce the traits that is under selection, involving selection pressure to maximize long-term transmission success. Such selection forces would favour traits that appear ‘altruistic’ (or neutral) at the level of within-host competition, but confer a benefit for long-term transmission from the host. Furthermore, the time frame for the evolution of these traits would extend from a single lifetime (somatic evolution within the host) to the much longer timescale of long chains of transmissions among hosts.

A number of criteria can be formulated for the agent-mediated scenario to work – these can also be regarded as predictions for the nature of the putative transmissible agents. First,

transmission of the agents must indeed be facilitated by the growth and spread of the affected tumour cell clones. In most known oncogenic infections, it is typically assumed that, except for the earliest stages of transformation, tumour cells no longer produce the pathogen that originally initiated the process, and cancer arises only as a side effect of mechanisms that promote the propagation of the infectious agent within the host (Burnet, 1970; Moore & Chang, 2010; Ewald, 2009; Ewald & Swain Ewald, 2015). In apparent contrast, the transmissible agents presumed to be responsible for paradoxical tumour traits should benefit, i.e. they should be produced from tumour cells at least up to the stage where these traits emerge. In particular, metastatic spread should also be beneficial for the agents, either simply by increasing their production, or by facilitating transmission to other individuals. These conditions are required to create selection pressure for the capacity to induce the observed paradoxical tumour traits. As we will explain below, the putative agents are likely to take advantage of the early, asymptomatic dissemination of tumour cells, which weakens the apparent contrast.

Second, the agents must possess mechanisms that code for, or facilitate the emergence of these tumour traits (going beyond the capacity of known oncogenic agents that can immortalize infected cells). Kaposi's sarcoma-associated herpesvirus (KSHV) contributes to the construction of tumour microenvironment in Kaposi's sarcoma by reprogramming adjacent (uninfected) cells *via* the exosomal transfer of regulatory microRNA (Yogev *et al*., 2017). If unknown infectious agents are responsible for the cancer types that display paradoxical tumour traits, they should also possess targeted mechanisms to induce these traits.

Third, the agents should be highly prevalent and of low virulence. Most cancer types that display paradoxical tumour traits (e.g. breast, prostate and pancreatic cancer) do not show epidemiological evidence for a transmissible cause. This apparent contradiction can be resolved if the causative infections are prevalent in the population, and are necessary, but not

sufficient to cause clinically manifest cancer (Ewald, 2000). Under this scenario, the vast majority of infections are asymptomatic, and manifest cases of cancer occur either due to rare stochastic processes, and/or facilitated by other carcinogenic co-factors. As a necessary condition, paradoxical tumour traits should emerge, and the agents should be transmitted already in the asymptomatic stages of tumour development (which seems consistent with the early development of micrometastases (Friberg & Nystrom, 2015; Pantel *et al*., 2009)). Indeed, symptomatic stages of cancer tend to be associated with progressive illness that is likely to limit the contacts of the affected individual, and would thereby strongly curtail the transmission of an infectious agent to new hosts, even if the production of the agent within the host continues. Between-host transmission is thus predicted to occur primarily in early asymptomatic infection. Finally, the requirement for high prevalence implies that the putative agents should be highly transmissible, while our failure to identify them indicates that they must be hard to detect (although low virulence in itself makes an infectious organism easier to miss).

At this point we can ask whether known infectious agents might fit the profile that we have sketched. With a few exceptions, currently known oncogenic pathogens do not seem to benefit from the stages of cancer at which paradoxical tumour traits typically emerge, and are not known to possess mechanisms that could induce these traits (see Section V.1 for details). While some known pathogens might have as yet undiscovered capabilities in this regard (the role of KSHV in the construction of the tumour microenvironment was only recently described), most cases of paradoxical tumour traits are associated with cancer types where even infectious aetiology has not been documented yet. The putative unknown agents might be undiscovered relatives of already known oncogenic organisms; e.g. the discovery of an expanding family of human polyomaviruses (some of which are associated with cancer risk) in the last ten years (Spurgeon & Lambert, 2013) illustrates that there might be a broad array

of asymptomatic infectious agents that have remained undetected. Alternatively, our failure to detect the putative agents might indicate that they are too divergent from currently known infectious organisms to be detected by current assays. In either case, if some paradoxical tumour traits are indeed induced by transmissible agents, then these agents can, as we explain below, be placed into a continuum of pathogenic life-history strategies.

(1) The adaptive continuum of pathogen-associated cancer

We argue that there is a continuum in the association between carcinogenesis and the transmission (fitness) of the various infectious agents that contribute to it. At one end of the spectrum, most of the currently known oncogenic pathogens do not seem to benefit from the cancers, which are thought to arise only as a side effect of the presence of the agents, e.g. due to mechanisms that promote the propagation of the causative organisms within the host (Burnet, 1970; Moore & Chang, 2010; Ewald, 2009; Ewald & Swain Ewald, 2015). In stark contrast, some non-human cancers are caused by transmissible tumour cells (Metzger & Goff, 2016; Ostrander, Davis & Ostrander, 2016; Ujvari, Gatenby & Thomas, 2017), implying that the tumour itself has become infectious, and metastasizing is the only way the infectious agents can spread. We think that these two extremes enclose a spectrum of gradual transitions, and propose the following classification within the continuum.

In our classification, Class 1 carcinogenic organisms include the pathogens that induce carcinogenic effects that do not affect the proliferation or survival of the infected cells; such infectious agents are regarded as ‘indirect carcinogens’ (International Agency for Research on Cancer, 2012). For example, infection by parasitic helminths is thought to increase the risk of cancer in the urinary bladder and other affected organs by promoting chronic inflammation that, *per se*, does not benefit the pathogen (Scholte, Pascoal-Xavier & Nahum, 2018). In other cases, the carcinogenic mechanism might affect uninfected cells, e.g. the cytotoxin-associated

gene A (CagA) virulence factor of *Helicobacter pylori* is able to enter and transform cells that do not harbour the bacterium (Segal *et al*., 1999) (*H. pylori* is predominantly extracellular). In these cases, transformed cells do not directly promote the spread of the pathogens.

Class 2 carcinogenic organisms can induce immortality and/or proliferation of the infected host cells, which increases the production of infectious progeny that can be transmitted to new hosts. The first stage of neoplastic transformation is thus beneficial for the pathogen, and the ability to induce such transformation is shaped by adaptive evolution of the infectious agents. However, metastatic cancer arises only occasionally, and is a 'side-effect' of relaxed cell-cycle control and (from the perspective of the pathogen) 'run-away' somatic evolution: metastatic cells typically no longer produce the pathogen. Epstein-Barr virus, human papillomaviruses (HPV), KSHV and Merkel cell polyomavirus (MCV) are probable representatives of this class, and while long considered to be indirect inflammatory carcinogens, hepatitis B and C viruses have also recently been characterized to encode direct carcinogenic mechanisms (Ewald & Swain Ewald, 2012; Irshad, Gupta & Irshad, 2017). Importantly, while the expression of certain viral proteins is required for the maintenance of the transformed phenotype in these cases, malignant tumours are generally assumed to have ceased producing intact virions in either MCV- (Shuda *et al*., 2008) or HPV- (Steenbergen *et al*., 2014) positive cases [although integrated copies of HPV might occasionally be reactivated (Woodman, Collins & Young, 2007)]. This stage likely represents a precarious balance for the pathogen: the degree of divergence from the healthy state helps it to persist and reproduce, but the weakening of the cellular control mechanisms inevitably entails the risk of further divergence towards a cancerous state in which viral replication is lost (Ewald & Swain Ewald, 2015). The ability to induce paradoxical tumour traits that aid the proliferation and spread of the infected tumour cell clones (e.g. long-range positive feedback loops or local niche construction) might evolve in this class.

We define Class 3 carcinogenic pathogens as those that retain the ability to produce offspring in (at least early-stage) metastatic infected cells. To achieve this, the pathogen must either be able to persist in the cells as the tumour acquires metastatic potential by somatic evolution, or itself must be capable of inducing metastatic characters while the tumour cell clone still preserves much of its genomic stability. In this class, the ability to induce paradoxical tumour traits that promote metastasis (distant niche construction, adaptive plasticity of metastatic characters) can also evolve.

The metastatic dissemination of infected cells can benefit a pathogen simply by increasing the production and titre of infectious progeny. Possible examples include eukaryotic *Theileria* parasites that induce metastatic dissemination of infected leukocytes by increasing the motility and invasiveness of the cells (Ma & Baumgartner, 2014); and jaagsiekte sheep retrovirus (JSRV), which causes a transmissible lung tumour of sheep, and is spread primarily by virions produced from tumour cells in the lung, but appears also in milk and colostrum, indicating some dissemination (Griffiths, Martineau & Cousens, 2010). Remarkably, JSRV appears to be able to induce neoplastic transformation in an autonomous fashion: the genetic divergence of JSRV-transformed tumour cells is therefore considerably lower compared with other tumour types, which is likely to be important for the continued production of virus from the transformed cells. Some intracellular bacteria might also follow a similar strategy: *Fusobacterium nucleatum* and other anaerobic bacteria associated with colorectal cancer appear to be maintained in distant metastases, spreading along with the cancer cells (Bullman *et al*., 2017).

In addition to increasing the production of infectious progeny, metastasis can, in some cases, also enable the pathogen to invade specific anatomical sites important for onward transmission to other hosts. This adds a qualitative dimension to the quantitative benefit (production of infectious progeny), and the fitness benefit is realized at the between-host

level. Paradoxical tumour traits that enable targeted metastasis might evolve. A possible example is Marek's disease virus, which is suspected to take advantage of transformed infected lymphocytes to disseminate to the anatomical sites of onward transmission (Boodhoo *et al*., 2016).

Finally, in rare cases, in addition to (or instead of) cell-free infectious particles, infected tumour cells can also be transmitted between hosts. The capability for metastatic dissemination is not an aid, but a strict prerequisite for this transmission route. However, the transmitted cells typically act only as vehicles for transmitting the infectious agent, and the pathogen can establish infection of the new individual by infecting resident host cells (with progeny released from the transmitted cells), rather than by the proliferation of the infected cells transmitted from the original host. The ability to induce distant spreading of the transformed cells is expected to be under strong selection in such cases. Furthermore, because the exchange of cells between individuals typically requires close contact, the pathogens are expected to evolve long persistence (and therefore low virulence) in the host, in order to adapt to the rare opportunities of transmission (similar to sexually transmitted infections) (Ewald, 2009). From the known carcinogenic pathogens, human T-cell leukemia viruses (and their non-human relatives) belong to this category (Pique & Jones, 2012).

Finally, we define Class 4 oncogenic pathogens to include transmissible cancer cell clones that are the causative organisms of transmissible cancers (Metzger & Goff, 2016; Ostrander *et al*., 2016; Ujvari *et al*., 2017). Naturally occurring transmissible cancer has been identified in three groups of hosts so far: canine transmissible venereal tumour (CTVT) is a sexually transmitted tumour in dogs that arose from a single cancer lineage thousands of years ago (Murgia *et al*., 2006); devil facial tumour disease affects Tasmanian devils (*Sarcophilus harrisii*) and consists of at least two independently evolved tumour lineages (Pearse & Swift, 2006; Pye *et al*., 2016); and disseminated neoplasia of bivalves affects multiple host species,

and evolved at least three times independently (Metzger *et al*., 2016). In these infections, it is the tumour cells themselves that are transmitted between hosts, and the infectious clones can be regarded as eukaryotic parasites [indeed, as new species in their own right (Frank, 2007; Vincent, 2010)]. In their present form, the known lineages of transmissible cancer appear to be fully autonomous: they do not harbour or require genetically independent oncogenic agents. However, an infectious aetiology cannot be excluded for the initial transformation of the ancestral tumour clone. In fact, cells transformed by Class 3 (or, to a lesser extent, by other) oncogenic pathogens might be prone to form transmissible clones, and it might be only the efficient elimination of allogeneic host cells that is likely to restrict the evolution of transmissible cancers to relatively rare occurrences. In the rare 'successful' cases, once a tumour cell clone has acquired the ability to be transmitted between hosts, there will be strong selection pressure on the cells to eliminate the original oncogenic agent (Ewald, 2009), in order to avoid the loss of resources, eliciting immune responses against the agent, and genomic instability induced by the oncogenic pathogen. This is likely to happen early after the initial transition, implying that the lack of an apparent co-infecting agent is not a strong argument against an infectious aetiology in the origin of transmissible cancers.

We emphasize that the classes in the proposed classification do not represent strict categories, but are intended only as illustrative stages of a continuous spectrum. Indeed, pathogens might evolve gradually along the spectrum, and the only abrupt transition occurs if transformed cells acquire the ability to be transmitted across consecutive host individuals, and evolve into a Class 4 oncogenic pathogen.

In this 'continuum' of the degree of linkage between the transmission of infectious agents and their oncogenic effect, putative transmissible agents responsible for paradoxical tumour traits are expected to belong mostly to Class 2 or 3, in which the causative agents benefit directly from some stages of oncogenesis. Transmissible cancers, which are clearly under selection for

efficient between-host transmission, might also evolve the ability to induce paradoxical tumour traits. However, large-scale cancer genotyping efforts should already have uncovered evidence of transmissible tumours if they were a common occurrence in human cancer, implying that the paradoxical tumour traits observed in common human cancers are unlikely to have such origins.

Finally, we note that the evolution of the mammalian placenta might have been triggered by retroviruses that can be regarded as Class 3 oncogenic pathogens. The trophoblastic cells of the placenta resemble cancer in their ability to divide rapidly, migrate, induce vascularization, suppress maternal immune rejection and, by these abilities, to effectively invade maternal tissues (Soundararajan & Rao, 2004). Remarkably, these properties rely heavily on genes of retroviral origin: on syncytins to enable cell fusion and immune suppression (reviewed in Denner, 2016), and possibly also on gene regulatory networks (Chuong, 2013). While now these genes benefit the host and can be considered to be ‘domesticated’, their expression in the placenta probably evolved originally to facilitate retroviral transmission between the mother and the foetus (Haig, 2012). The invasive properties of trophoblasts might have originated from an earlier stage of evolution in which infected cells were transformed by the retroviruses towards an invasive phenotype to create a vehicle for efficient transmission. This would have classified these viruses as Class 3 oncogenic pathogens, which would have been under selection pressure to induce paradoxical tumour traits.

VI. HOW TO DISTINGUISH?

We delineated three possible scenarios that could explain the evolution of paradoxical tumour traits: specific selection regimes, inherent vulnerabilities, and manipulation by transmissible agents. All three alternatives require specific sets of conditions, providing clues for the validation and discrimination of the scenarios. The major testable dichotomy is whether a

tumour trait arises by autonomous processes within the organism, or is induced by an acquired external element. Validation of the agent-mediated scenario requires the identification of the agent, and the demonstration of its ability to induce and to benefit from a paradoxical tumour trait.

The scenarios based on selection and inherent vulnerabilities are harder to distinguish. In both cases, the mechanisms of tumour traits can arise by mutations and/or epimutations, and there is a smooth gradient between no selection and strong selection. Signatures of positive selection in the genes responsible for paradoxical tumour traits can be revealing, as can convergent evolution in independent instances of cancer (although the latter might involve convergence in the phenotype without strong convergence in the genotype; Wu *et al*., 2016). Ultimately, the cost of the traits in the local selective microenvironment can be assessed by *in vitro* competition experiments using cells with manipulated levels of expression of the trait (Archetti *et al*., 2015). However, to proceed to this stage requires first a detailed characterization of the genetic background and molecular mechanisms of the paradoxical tumour traits. Such an understanding is needed also to recognize coupling between local and distal effects, or cases where the trait can emerge from an inherent vulnerability of the organism.

In addition, some predictions can be made also in the absence of a detailed understanding of the mechanisms. For example, if paradoxical tumour traits reflect the action of transmissible agents, then cancers that are caused by other carcinogenic effects should not display such traits, and should, in general, display less (or fewer) complex phenotypes. Testing this prediction is, however, sensitive to uncertainties regarding the contribution of infectious causes to cancer.

Another prediction can be formulated regarding genomic instability, which is a hallmark and key driver of carcinogenesis (Loeb, 1991). Genomic instability creates variation, which opens

up both adaptive and neutral evolutionary trajectories, and thereby increases the capacity and 'reach' of evolutionary processes within the lifetime of the organism. In particular, the evolution of tumour traits by somatic selection is expected to occur in the context of increasing genomic instability, which aids the exploration of new genotypes. For transmissible agents, however, genomic instability makes it harder to retain 'control' of the host cell: strong within-host selection (aided by enhanced variation) might erode the traits responsible for between-host transmission (Levin & Bull, 1994), and the agent itself might be eliminated. If feasible, the optimal strategy for the agents is rather a targeted manipulation of only those processes that directly promote their spread. As an implication, those tumour types that progress to a metastatic stage with fewer mutations (lower degree of genomic instability) might be more likely to be initiated by transmissible agents, and the search for the putative agents could focus on these types. Similarly, precancerous cells in the early stages of transformation are more promising for the search for the agents compared with late-stage cells or cell lines that have been strongly re-shaped by somatic selection and genomic instability, and are less likely to have retained the initiating agents.

VII. CONTEXT AND IMPLICATIONS

We have shown that some complex tumour traits are difficult to explain by somatic selection not only due to their complexity, but also because local selection in the tumour microenvironment is expected to act against these traits. The idea that costly 'public goods' type traits of cancer might be disfavoured by somatic selection has been brought up and analysed previously (e.g. Tomlinson & Bodmer, 1997; Nagy, 2004; Merlo *et al*., 2006; Tissot *et al*., 2016; Tabassum & Polyak, 2015; Archetti, 2013; Archetti *et al*., 2015). Most of the earlier studies addressed the production of diffusible growth factors; we have extended the

arguments to demonstrate the additional difficulties associated with more complex 'paradoxical tumour traits'.

We also note that the difficulty in explaining the origin of paradoxical tumour traits by somatic selection is not alleviated by the evolution of local cooperation between tumour cell clones, which has been predicted (Axelrod *et al*., 2006) and demonstrated (Cleary *et al*., 2014) to occur. Cooperation can evolve when distinct tumour cell clones provide mutually beneficial functions to each other that combine into synergistic fitness gain. By contrast, non-cell-autonomous paradoxical tumour traits (niche construction, long-range positive feedback loops) operate by manipulating non-evolving stromal cells, which excludes such co-evolutionary mechanisms.

We developed three possible scenarios that could explain the evolution of these traits, based on somatic selection driven by specific selection regimes, inherent vulnerabilities, or manipulation by transmissible agents. Importantly, the three scenarios are not mutually exclusive. In particular, inherent vulnerabilities can facilitate adaptive evolution by providing 'ready-made' packages of complex functionalities, e.g. phenotypic plasticity or intercellular communication. This might apply to both somatic evolution driven by specific selection regimes, and to the evolution of transmissible agents that can exploit the vulnerabilities.

Inherent vulnerability, the ease of repeatedly evolving cancer, has previously been discussed in the context of 'atavism', arguing that the ancient 'toolkit' of the ancestral unicellular lifestyle might still exist in the genome of multicellular organisms, and might be unlocked by genetic or epigenetic malfunction (Davies & Lineweaver, 2011; Davies & Agus, 2015; Vincent, 2012). This hypothesis has recently gained support by genomic and transcriptomic analyses showing that neoplastic transformation is indeed associated with a convergent loss of multicellularity-associated genes, and a return to a unicellular-like cell state (Chen *et al*., 2015; Trigos *et al*., 2017). Atavistic traits thus indeed appear to facilitate evolution towards a

'selfish' lifestyle (Thomas *et al*., 2017), involving cell-autonomous cancer traits such as replicative immortality.

However, the predisposition to revert to atavistic cell states cannot contribute to the paradoxical tumour traits that involve non-cell-autonomous effects, and are 'paradoxical' because they appear to contradict selection for selfish traits. Instead, the inherent vulnerabilities relevant to these traits are more likely to stem from functions that evolved in conjunction with the multicellular lifestyle. To list some examples, several pathways of tumour growth exploit genetic circuitry that is normally active during embryogenesis (Ma *et al*., 2010; Cano *et al*., 2000) or tissue regeneration (Orimo *et al*., 2005; Krall *et al*., 2018); cell motility plays an important role in immune surveillance in animals (Ewald & Swain Ewald, 2015); and placental mammals have evolved invasive properties of trophoblasts to enable placentation (Haig, 2015). These capabilities are not atavisms, but have evolved and are maintained for functions required for multicellular organisms. As a result, they are more amenable to exploitation for 'cooperative' functions in cancer.

On a general note, the diversity of inherent vulnerabilities is likely to scale with the complexity of the organism. While cancer-like phenomena occur almost everywhere across the tree of life (Aktipis *et al*., 2015; Albuquerque *et al*., 2018), complex tumour traits (aided by emergent vulnerabilities) are more likely to occur in the organisms with the most complex body plans and developmental flexibility. Of note, Burnet (1968) proposed 50 years ago that vertebrates, in particular, might pay a price of increased susceptibility to cancer for the increased developmental flexibility associated with this lineage.

How do the possibilities tackled in this paper affect the way we should (or could) treat malignant disease? Some implications of somatic selection and inherent vulnerabilities have been discussed elsewhere (Thomas *et al*., 2017). In general, tumour traits that arise by somatic selection might be more amenable to selective treatment, as these represent features divergent

from the normal physiology of the organism. By contrast, traits arising from inherent vulnerabilities comprise components that participate in some normal function, and targeting these might run a greater risk of adverse reactions by interfering with essential processes. In fact, vulnerabilities that were not coupled to some important function by structural constraints are expected to have been weeded out by selection, leaving only those that are protected by such coupling.

Obviously, if some cancer traits are induced by transmissible agents, then identifying and eliminating these agents could prevent the development of the affected cases of cancer. Ewald (2009, p. 24) estimated that "a causal role for parasitism can be excluded for less than 5% of all cancer". The lack of epidemiological signatures of a contagious cause can be explained if the causative organisms are common in the population, and cancer arises only when the infectious agent is present in combination with some other predisposing factor, as seems be the case with Epstein-Barr virus- and Merkel cell polyomavirus-associated cancers (Moore & Chang, 2017). In Ewald's nomenclature, the infectious agents should comprise an 'essential' cause of cancer, breaking down key initial barriers to oncogenesis, while further 'exacerbating' factors should be needed for the development of malignant tumours (Ewald & Swain Ewald, 2013). In this scenario, most infected individuals never develop clinically manifest cancer; however, eliminating transmission would nonetheless eliminate all cases of cancer that involve the action of the agent (Fig. 2).

A potential further implication for treatment might arise if transmissible agents can mediate a cumulative evolution of resistance against cancer drugs or radiation therapy. If cancer patients were a potential source of transmission, and the evolution of resistance in cancer treatment could involve heritable (genetic or epigenetic) traits of the transmissible agent, then these changes could be transmitted to new patients, and resistance could increase across the chains of transmission, abrogating the efficiency of widely used treatment modalities at the

population level. This effect is limited if, as we deem likely, most transmissions occur in the pre-clinical stage from asymptomatic carriers.

Conversely, if ‘paradoxical tumour traits’ turn out to arise by internal evolution, then mapping the precise conditions that generate specific selection pressures or inherent vulnerabilities might also point to new treatment strategies.

VIII. CONCLUSIONS

(1) We established that some complex tumour traits are difficult to explain by somatic selection not only due to their complexity, but also because they are expected to confer a fitness cost, rather than a benefit in the local competitive microenvironment of a tumour. Some traits (non-cell-autonomous effects of niche construction and long-range positive feedback loops) offer a benefit that is expected to be shared equally by cells contributing to the trait, and other clones that did not pay the ‘cost’, excluding the spread of the trait within the tumour. Other traits that aid metastases (e.g. adaptive phenotypic plasticity of metastatic characters) are expected to be under weak selection, as within-tumour selection is likely to dominate the life history of tumour cell clones. We formulated three scenarios that could explain, possibly in combination, the recurrent emergence of such ‘paradoxical tumour traits’.

(2) First, somatic selection can be ‘rescued’ under specific selection regimes that we described for each problematic trait. In the case of non-cell-autonomous effects, paradoxical tumour traits can evolve if their benefit affects producer cells preferentially, or if the distributed long-range effect is coupled (by structural constraints) to some local beneficial effect that is focused on the producer cells. Complex traits that aid metastases might evolve if metastases occur repeatedly, possibly in a ‘ping-pong’ fashion between different tumour foci.

(3) Alternatively, (some of) the paradoxical traits might be induced by putative (potentially, as yet unknown) transmissible agents that are disseminated by or from infected tumour cells.

In this case, the selection pressures moulding these traits would act not (only) at the within-host level and timescale, but also at the level and timescale of serial transmissions among hosts. A pathogen that is able to spread from tumour cells is under selection to evolve traits that aid the transformation, growth and dissemination of the infected cells, including traits that would incur a net cost in the intra-tumour competition of tumour cell clones. We predicted several characteristics of the hypothetical agents, including low virulence, i.e. low propensity to cause clinically manifest cancer without additional co-factors; instead, their transmission should mostly be aided by the early dissemination of tumour cells, resulting in the lack of an epidemiological signature in the diagnosed cases of malignant tumours. The existence of such transmissible agents would nonetheless increase the contribution of infectious causes to cancer.

(4) In conjunction with the possible infectious causes of complex tumour traits, we developed the framework of an adaptive continuum of pathogen-associated cancer. The continuum refers to degrees of association between stages of cancer and the transmission of oncogenic pathogens. The spectrum extends from pathogens that cause cancer as an indirect effect, but are not propagated in tumour cells, to transmissible tumours in which it is the tumour cell clones that are transmitted between hosts. In between are pathogens that contribute to the transformation of the infected cells, and are produced in either the local tumour growth or, in some cases, also from metastatic cells. Putative transmissible agents responsible for paradoxical tumour traits are expected to belong to these intermediate categories.

(5) Finally, inherent structural, genomic and regulatory components of the organism might present vulnerabilities that offer ‘easy’ evolutionary trajectories to complex tumour traits, possibly even by neutral drift, or in combination with either somatic selection or transmissible agents. The vulnerabilities relevant for complex tumour traits are likely to stem from characters originally selected for multicellular lifestyles, rather than from atavisms that

resurrect unicellular characters. The level of developmental complexity and plasticity might be correlated with the risk and severity of such vulnerabilities, exposing organisms with higher developmental complexity (e.g. vertebrates) to a higher burden of cancer.

(6) We have thus extended the dichotomy of somatic selection and atavism as the explanatory framework of tumour evolution (Thomas *et al*., 2017) in two ways: (1) by widening the scope of predisposing factors from atavistic traits to a broader category of inherent vulnerabilities (including those emerging from multicellular adaptations), and (2) by adding a third scenario based on manipulation by transmissible agents.

IX. ACKNOWLEDGEMENTS

We thank Sebastian Bonhoeffer, Paul W. Ewald, András Hubai, Imre Kacskovics and Krisztina Takács for feedback and insightful comments on some of the ideas presented herein. We are deeply grateful to Steve Frank for a critical reading of an earlier version, and for stimulating discussions that have considerably shaped our views of cancer evolution.

This work was completed in the ELTE Institutional Excellence Program (783-3/2018/FEKUTSRAT) supported by the Hungarian Ministry of Human Capacities, and has also been funded by the grant GINOP-2.3.2-15-2016-00057 (“Az evolúció fényében: elvek és megoldások”). P.A. was supported by the ÚNKP-17-3 New National Excellence Program of the Hungarian Ministry of Human Capacities; V.M. was supported by a Bolyai János Research Fellowship of the Hungarian Academy of Sciences and by a Bolyai+ Fellowship of the New National Excellence Program of the Hungarian Ministry of Human Capacities.

X. REFERENCES

AKTIPIS, C. A., BODDY, A. M., JANSEN, G., HIBNER, U., HOCHBERG, M. E., MALEY, C. C. & WILKINSON, G. S. (2015). Cancer across the tree of life: cooperation and cheating in multicellularity. *Philosophical Transactions of the Royal Society B: Biological Sciences* **370**, 20140219.

AKTIPIS, C. A., MALEY, C. C. & PEPPER, J. W. (2012). Dispersal evolution in neoplasms: the role of disregulated metabolism in the evolution of cell motility. *Cancer Prevention Research* **5**, 266–275.

ALBUQUERQUE, T. A. F., DRUMMOND DO VAL, L., DOHERTY, A. & DE MAGALHAES, J. P. (2018). From humans to hydra: patterns of cancer across the tree of life. *Biological Reviews Cambridge Philosophical Society* **93**, 1715-1734.

ARCHETTI, M. (2013). Evolutionary game theory of growth factor production: implications for tumour heterogeneity and resistance to therapies. *British Journal of Cancer* **109**, 1056–1062.

ARCHETTI, M., FERRARO, D. A. & CHRISTOFORI, G. (2015). Heterogeneity for IGF-II production maintained by public goods dynamics in neuroendocrine pancreatic cancer. *Proceedings of the National Academy of Sciences* **112**, 1833–1838.

ARNAL, A., UJVARI, B., CRESPI, B., GATENBY, R. A., TISSOT, T., VITTECOQ, M., EWALD, P. W., CASALI, A., DUCASSE, H., JACQUELINE, C., MISSE, D., RENAUD, F., ROCHE, B. & THOMAS, F. (2015). Evolutionary perspective of cancer: myth, metaphors, and reality. *Evolutionary Applications* **8**, 541–544.

AXELROD, R., AXELROD, D. E. & PIENTA, K. J. (2006). Evolution of cooperation among tumor cells. *Proceedings of the National Academy of Sciences* **103**, 13474–13479.

BATLLE, E. & CLEVERS, H. (2017). Cancer stem cells revisited. *Nature Medicne* **23**, 1124–1134.

BERNARDS, R. & WEINBERG, R. A. (2002). Metastasis genes: A progression puzzle. *Nature* **418**, 823.

BOODHOO, N., GURUNG, A., SHARIF, S. & BEHBOUDI, S. (2016). Marek's disease in chickens: a review with focus on immunology. *Veterinary Research* **47**, 119.

BULLMAN, S., PEDAMALLU, C. S., SICINSKA, E., CLANCY, T. E., ZHANG, X., CAI, D., NEUBERG, D., HUANG, K., GUEVARA, F., NELSON, T., CHIPASHVILI, O., HAGAN, T., WALKER, M., RAMACHANDRAN, A., DIOSDADO, B. *ET AL*. (2017). Analysis of *Fusobacterium* persistence and antibiotic response in colorectal cancer. *Science* **358**, 1443–1448.

BURNET, F. M. (1968). Evolution of the immune process in vertebrates. *Nature* **218**, 426–30.

BURNET, F. M. (1970). *Immunological Surveillance*. Pergamon, Oxford.

CANO, A., PÉREZ-MORENO, M. A., RODRIGO, I., LOCASCIO, A., BLANCO, M. J., DEL BARRIO, M. G., PORTILLO, F. & NIETO, M. A. (2000). The transcription factor Snail controls epithelial–mesenchymal transitions by repressing E-cadherin expression. *Nature Cell Biology* **2**, 76–83.

CHEN, H., LIN, F., XING, K. & HE, X. (2015). The reverse evolution from multicellularity to unicellularity during carcinogenesis. *Nature Communications* **6**, 6367.

CHUONG, E. B. (2013). Retroviruses facilitate the rapid evolution of the mammalian placenta. *Bioessays* **35**, 853–861.

CLEARY, A. S., LEONARD, T. L., GESTL, S. A. & GUNTHER, E. J. (2014). Tumour cell heterogeneity maintained by cooperating subclones in Wnt-driven mammary cancers. *Nature* **508**, 113–117.

COFFELT, S. B., KERSTEN, K., DOORNEBAL, C. W., WEIDEN, J., VRIJLAND, K., HAU, C. S., VERSTEGEN, N. J. M., CIAMPRICOTTI, M., HAWINKELS, L., JONKERS, J. & DE VISSER, K. E. (2015). IL-17-producing gammadelta T cells and neutrophils conspire to promote breast cancer metastasis. *Nature* **522**, 345–348.

COSTA-SILVA, B., AIELLO, N. M., OCEAN, A. J., SINGH, S., ZHANG, H., THAKUR, B. K., BECKER, A., HOSHINO, A., MARK, M. T., MOLINA, H., XIANG, J., ZHANG, T., THEILEN,

T. M., GARCIA-SANTOS, G., WILLIAMS, C. *ET AL*. (2015). Pancreatic cancer exosomes initiate pre-metastatic niche formation in the liver. *Nature Cell Biology* **17**, 816–826.

COX, T. R., RUMNEY, R. M. H., SCHOOF, E. M., PERRYMAN, L., HOYE, A. M., AGRAWAL, A., BIRD, D., LATIF, N. A., FORREST, H., EVANS, H. R., HUGGINS, I. D., LANG, G., LINDING, R., GARTLAND, A. & ERLER, J. T. (2015). The hypoxic cancer secretome induces pre-metastatic bone lesions through lysyl oxidase. *Nature* **522**, 106–110.

DAVIES, P. C. & AGUS, D. B. (2015). Stochasticity and determinism in cancer creation and progression. *Convergent Science Physical Oncology* **1**, 026003.

DAVIES, P. C. & LINEWEAVER, C. H. (2011). Cancer tumors as Metazoa 1.0: tapping genes of ancient ancestors. *Physical Biology* **8**, 015001.

DENNER, J. (2016). Expression and function of endogenous retroviruses in the placenta. *APMIS* **124**, 31–43.

EGEBLAD, M., NAKASONE, E. S. & WERB, Z. (2010). Tumors as organs: complex tissues that interface with the entire organism. *Developmental Cell* **18**, 884–901.

ENGBLOM, C., PFIRSCHKE, C., ZILIONIS, R., DA SILVA MARTINS, J., BOS, S. A., COURTIES, G., RICKELT, S., SEVERE, N., BARYAWNO, N., FAGET, J., SAVOVA, V., ZEMMOUR, D., KLINE, J., SIWICKI, M., GARRIS, C. *ET AL*. (2017). Osteoblasts remotely supply lung tumors with cancer-promoting SiglecF^{high} neutrophils. *Science* **358**, eaal5081.

EWALD, P. W. (2000). *Plague time: How stealth infections cause cancers, heart disease, and other deadly ailments*. Simon and Schuster.

EWALD, P. W. (2009). An evolutionary perspective on parasitism as a cause of cancer. *Advances in Parasitology* **68**, 21–43.

EWALD, P. W. & SWAIN EWALD, H. A. (2012). Infection, mutation, and cancer evolution. *Journal of Molecular Medicine* **90**, 535–541.

EWALD, P. W. & SWAIN EWALD, H. A. (2013). Toward a general evolutionary theory of oncogenesis. *Evolutionary Applications* **6**, 70–81.

EWALD, P. W. & SWAIN EWALD, H. A. (2015). Infection and cancer in multicellular organisms. *Philosophical Transactions of the Royal Society B: Biological Sciences* **370**, 20140224.

FLAVAHAN, W. A., GASKELL, E. & BERNSTEIN, B. E. (2017). Epigenetic plasticity and the hallmarks of cancer. *Science* **357**.

FRANK, U. (2007). The evolution of a malignant dog. *Evolution & Development* **9**, 521–522.

FRIBERG, S. & NYSTROM, A. (2015). Cancer metastases: early dissemination and late recurrences. *Cancer Growth Metastasis* **8**, 43–49.

GATENBY, R. A. & GILLIES, R. J. (2008). A microenvironmental model of carcinogenesis. *Nature Reviews Cancer* **8**, 56–61.

GRANGE, C., TAPPARO, M., COLLINO, F., VITILLO, L., DAMASCO, C., DEREGIBUS, M. C., TETTA, C., BUSSOLATI, B. & CAMUSSI, G. (2011). Microvesicles released from human renal cancer stem cells stimulate angiogenesis and formation of lung premetastatic niche. *Cancer Research* **71**, 5346–5356.

GREAVES, M. & MALEY, C. C. (2012). Clonal evolution in cancer. *Nature* **481**, 306–313.

GRIFFITHS, D. J., MARTINEAU, H. M. & COUSENS, C. (2010). Pathology and Pathogenesis of Ovine Pulmonary Adenocarcinoma. *Journal of Comparative Pathology* **142**, 260–283.

GRUNAU, C. (2017). The epigenetic component in cancer evolution. In *Ecology and Evolution of Cancer* (ed. B. Ujvari, B. Roche and F. Thomas), pp. 87–98. Academic Press.

GUNDEM, G., VAN LOO, P., KREMEYER, B., ALEXANDROV, L. B., TUBIO, J. M. C., PAPAEMMANUIL, E., BREWER, D. S., KALLIO, H. M. L., HÖGNÄS, G., ANNALA, M., KIVINUMMI, K., GOODY, V., LATIMER, C., O'MEARA, S., DAWSON, K. J. *ET AL*. (2015). The evolutionary history of lethal metastatic prostate cancer. *Nature* **520**, 353.

HAIG, D. (2012). Retroviruses and the placenta. *Current Biology* **22,** R609–R613.

HAIG, D. (2015). Maternal–fetal conflict, genomic imprinting and mammalian vulnerabilities to cancer. *Philosophical Transactions of the Royal Society B: Biological Sciences* **370**, 20140178.

HANAHAN, D. & COUSSENS, L. M. (2012). Accessories to the crime: functions of cells recruited to the tumor microenvironment. *Cancer Cell* **21**, 309–322.

HANAHAN, D. & WEINBERG, R. A. (2000). The hallmarks of cancer. *Cell* **100**, 57–70.

HOSHINO, A., COSTA-SILVA, B., SHEN, T.-L., RODRIGUES, G., HASHIMOTO, A., TESIC MARK, M., MOLINA, H., KOHSAKA, S., DI GIANNATALE, A., CEDER, S., SINGH, S., WILLIAMS, C., SOPLOP, N., URYU, K., PHARMER, L. *ET AL*. (2015). Tumour exosome integrins determine organotropic metastasis. *Nature* **527**, 329–335.

INTERNATIONAL AGENCY FOR RESEARCH ON CANCER (2012). Biological agents. Volume 100B. A review of human carcinogens. In *IARC Monographs on the Evaluation of Carcinogenic Risks to Humans*, pp. 40.

IRSHAD, M., GUPTA, P. & IRSHAD, K. (2017). Molecular basis of hepatocellular carcinoma induced by hepatitis C virus infection. *World Journal of Hepatology* **9**, 1305–1314.

JAYATILAKA, H., TYLE, P., CHEN, J. J., KWAK, M., JU, J., KIM, H. J., LEE, J. S. H., WU, P. H., GILKES, D. M., FAN, R. & WIRTZ, D. (2017). Synergistic IL-6 and IL-8 paracrine signalling pathway infers a strategy to inhibit tumour cell migration. *Nature Communications* **8**, 15584.

KALLURI, R. & ZEISBERG, M. (2006). Fibroblasts in cancer. *Nature Reviews Cancer* **6**, 392.

KAPLAN, R. N., RIBA, R. D., ZACHAROULIS, S., BRAMLEY, A. H., VINCENT, L., COSTA, C., MACDONALD, D. D., JIN, D. K., SHIDO, K., KERNS, S. A., ZHU, Z., HICKLIN, D., WU, Y., PORT, J. L., ALTORKI, N. *ET AL*. (2005). VEGFR1-positive haematopoietic bone marrow progenitors initiate the pre-metastatic niche. *Nature* **438**, 820–827.

KIM, M.-Y., OSKARSSON, T., ACHARYYA, S., NGUYEN, D. X., ZHANG, X. H. F., NORTON, L. & MASSAGUÉ, J. (2009). Tumor self-seeding by circulating cancer cells. *Cell* **139**, 1315–1326.

KRALL, J. A., REINHARDT, F., MERCURY, O. A., PATTABIRAMAN, D. R., BROOKS, M. W., DOUGAN, M., LAMBERT, A. W., BIERIE, B., PLOEGH, H. L., DOUGAN, S. K. & WEINBERG, R. A. (2018). The systemic response to surgery triggers the outgrowth of distant immune-controlled tumors in mouse models of dormancy. *Science Translational Medicine* **10**, eaan3464

LEVIN, B. R. & BULL, J. J. (1994). Short-sighted evolution and the virulence of pathogenic microorganisms. *Trends in Microbiology* **2**, 76–81.

LOEB, L. A. (1991). Mutator phenotype may be required for multistage carcinogenesis. *Cancer Research* **51**, 3075–3079.

MA, M. & BAUMGARTNER, M. (2014). Intracellular *Theileria annulata* promote invasive cell motility through kinase regulation of the host actin cytoskeleton. *PLoS Pathogens* **10**, e1004003.

MA, Y., ZHANG, P., WANG, F., YANG, J., YANG, Z. & QIN, H. (2010). The relationship between early embryo development and tumourigenesis. *Journal of Cellular and Molecular Medicine* **14**, 2697–2701.

MERLO, L. M. F., PEPPER, J. W., REID, B. J. & MALEY, C. C. (2006). Cancer as an evolutionary and ecological process. *Nature Reviews Cancer* **6**, 924–935.

METZGER, M. J. & GOFF, S. P. (2016). A Sixth modality of infectious disease: contagious cancer from devils to clams and beyond. *PLoS Pathogens* **12**, e1005904.

METZGER, M. J., VILLALBA, A., CARBALLAL, M. J., IGLESIAS, D., SHERRY, J., REINISCH, C., MUTTRAY, A. F., BALDWIN, S. A. & GOFF, S. P. (2016). Widespread transmission of independent cancer lineages within multiple bivalve species. *Nature* **534**, 705–709.

MICHOD, R. E. (2003). Cooperation and conflict mediation during the origin of multicellularity. In *Genetic and cultural evolution of cooperation* (ed. P. Hammerstein), pp. 291–307. MIT press, Cambridge MA.

MOORE, P. S. & CHANG, Y. (2010). Why do viruses cause cancer? Highlights of the first century of human tumour virology. *Nature Reviews Cancer* **10**, 878–889.

MOORE, P. S. & CHANG, Y. (2017). Common Commensal Cancer Viruses. *PLoS Pathogens* **13**, e1006078.

MURGAI, M., JU, W., EASON, M., KLINE, J., BEURY, D. W., KACZANOWSKA, S., MIETTINEN, M. M., KRUHLAK, M., LEI, H., SHERN, J. F., CHEREPANOVA, O. A., OWENS, G. K. & KAPLAN, R. N. (2017). KLF4-dependent perivascular cell plasticity mediates pre-metastatic niche formation and metastasis. *Nature Medicine* **23**, 1176–1190.

MURGIA, C., PRITCHARD, J. K., KIM, S. Y., FASSATI, A. & WEISS, R. A. (2006). Clonal origin and evolution of a transmissible cancer. *Cell* **126**, 477–487.

NAGY, J. D. (2004). Competition and natural selection in a mathematical model of cancer. *Bulletin of Mathematical Biology* **66**, 663–687.

NOWELL, P. (1976). The clonal evolution of tumor cell populations. *Science* **194,** 23–28.

OLMEDA, D., CEREZO-WALLIS, D., RIVEIRO-FALKENBACH, E., PENNACCHI, P. C., CONTRERAS-ALCALDE, M., IBARZ, N., CIFDALOZ, M., CATENA, X., CALVO, T. G., CANON, E., ALONSO-CURBELO, D., SUAREZ, J., OSTERLOH, L., GRANA, O., MULERO, F. *ET AL*. (2017). Whole-body imaging of lymphovascular niches identifies pre-metastatic roles of midkine. *Nature* **546**, 676-680.

ORIMO, A., GUPTA, P. B., SGROI, D. C., ARENZANA-SEISDEDOS, F., DELAUNAY, T., NAEEM, R., CAREY, V. J., RICHARDSON, A. L. & WEINBERG, R. A. (2005). Stromal fibroblasts present in invasive human breast carcinomas promote tumor growth and angiogenesis through elevated SDF-1/CXCL12 secretion. *Cell* **121**, 335–348.

OSTRANDER, E. A., DAVIS, B. W. & OSTRANDER, G. K. (2016). Transmissible tumors: breaking the cancer paradigm. *Trends in Genetics* **32**, 1–15.

PAGET, S. (1889). The distribution of secondary growths in cancer of the breast. 1889. *The Lancet* **133**, 571–573.

PANTEL, K., ALIX-PANABIÈRES, C. & RIETHDORF, S. (2009). Cancer micrometastases. *Nature Reviews Clinical Oncology* **6**, 339–351.

PANTEL, K. & BRAKENHOFF, R. H. (2004). Dissecting the metastatic cascade. *Nature Reviews Cancer* **4**, 448–456.

PARK, S. I., LEE, C., SADLER, W. D., KOH, A. J., JONES, J., SEO, J. W., SOKI, F. N., CHO, S. W., DAIGNAULT, S. D. & MCCAULEY, L. K. (2013). Parathyroid hormone–related protein drives a $CD11b^{+}Gr1^{+}$ cell–mediated positive feedback loop to support prostate cancer growth. *Cancer Research* **73**, 6574–6583.

PEARSE, A. M. & SWIFT, K. (2006). Transmission of devil facial-tumour disease. *Nature* **439**, 549.

PEINADO, H., ALECKOVIC, M., LAVOTSHKIN, S., MATEI, I., COSTA-SILVA, B., MORENO-BUENO, G., HERGUETA-REDONDO, M., WILLIAMS, C., GARCIA-SANTOS, G., GHAJAR, C., NITADORI-HOSHINO, A., HOFFMAN, C., BADAL, K., GARCIA, B. A., CALLAHAN, M. K., *ET AL*. (2012). Melanoma exosomes educate bone marrow progenitor cells toward a pro-metastatic phenotype through MET. *Nature Medicine* **18**, 883–891.

PEINADO, H., ZHANG, H., MATEI, I. R., COSTA-SILVA, B., HOSHINO, A., RODRIGUES, G., PSAILA, B., KAPLAN, R. N., BROMBERG, J. F., KANG, Y., BISSELL, M. J., COX, T. R., GIACCIA, A. J., ERLER, J. T., HIRATSUKA, S. *ET AL*. (2017). Pre-metastatic niches: organ-specific homes for metastases. *Nature Reviews Cancer* **17**, 302–317.

PIQUE, C. & JONES, K. (2012). Pathways of cell-cell transmission of HTLV-1. *Frontiers in Microbiology* **3**, 378.

PLUMMER, M., DE MARTEL, C., VIGNAT, J., FERLAY, J., BRAY, F. & FRANCESCHI, S. (2016). Global burden of cancers attributable to infections in 2012: a synthetic analysis. *The Lancet Global Health* **4**, e609–e616.

POLLARD, J. W. (2004). Tumour-educated macrophages promote tumour progression and metastasis. *Nature Reviews Cancer* **4**, 71–78.

PRADEU, T. (2013). Immunity and the Emergence of Individuality. In *From Groups to Individuals: Evolution and Emerging Individuality* (ed. F. Bouchard and P. Huneman), pp. 77–96. MIT Press, Cambridge, MA.

PYE, R. J., PEMBERTON, D., TOVAR, C., TUBIO, J. M., DUN, K. A., FOX, S., DARBY, J., HAYES, D., KNOWLES, G. W., KREISS, A., SIDDLE, H. V., SWIFT, K., LYONS, A. B., MURCHISON, E. P. & WOODS, G. M. (2016). A second transmissible cancer in Tasmanian devils. *Proceedings of the National Academy of Sciences* **113**, 374–379.

QIU, W., HU, M., SRIDHAR, A., OPESKIN, K., FOX, S., SHIPITSIN, M., TRIVETT, M., THOMPSON, E. R., RAMAKRISHNA, M., GORRINGE, K. L., POLYAK, K., HAVIV, I. & CAMPBELL, I. G. (2008). No evidence of clonal somatic genetic alterations in cancer-associated fibroblasts from human breast and ovarian carcinomas. *Nature Genetics* **40**, 650–655.

ROTH, J. A., SILVERSTEIN, M. J. & MORTON, D. L. (1976). Metastatic potential of metastases. *Surgery* **79**, 669–673.

SCHOLTE, L. L. S., PASCOAL-XAVIER, M. A. & NAHUM, L. A. (2018). Helminths and Cancers From the Evolutionary Perspective. *Frontiers in Medicine* **5**, 90

SEGAL, E. D., CHA, J., LO, J., FALKOW, S. & TOMPKINS , L. S. (1999). Altered states: Involvement of phosphorylated CagA in the induction of host cellular growth changes by *Helicobacter pylori*. *Proceedings of the National Academy of Sciences* **96**, 14559–14564.

SHUDA, M., FENG, H., KWUN, H. J., ROSEN, S. T., GJOERUP, O., MOORE, P. S. & CHANG, Y. (2008). T antigen mutations are a human tumor-specific signature for Merkel cell polyomavirus. *Proceedings of the National Academy of Sciences* **105**, 16272–16277.

SOUNDARARAJAN, R. & RAO, A. J. (2004). Trophoblast 'pseudo-tumorigenesis': significance and contributory factors. *Reproductive Biology and Endocrinology* **2**, 15.

SPURGEON, M. E. & LAMBERT, P. F. (2013). Merkel cell polyomavirus: A newly discovered human virus with oncogenic potential. *Virology* **435**, 118–130.

STEENBERGEN, R. D. M., SNIJDERS, P. J. F., HEIDEMAN, D. A. M. & MEIJER, C. J. L. M. (2014). Clinical implications of (epi)genetic changes in HPV-induced cervical precancerous lesions. *Nature Reviews Cancer* **14**, 395–405.

TABASSUM, D. P. & POLYAK, K. (2015). Tumorigenesis: it takes a village. *Nature Reviews Cancer* **15**, 473–483.

THOMAS, F., UJVARI, B., RENAUD, F. & VINCENT, M. (2017). Cancer adaptations: Atavism, de novo selection, or something in between? *Bioessays* **39**, 1700039.

TISSOT, T., ARNAL, A., JACQUELINE, C., POULIN, R., LEFEVRE, T., MERY, F., RENAUD, F., ROCHE, B., MASSOL, F., SALZET, M., EWALD, P., TASIEMSKI, A., UJVARI, B. & THOMAS, F. (2016). Host manipulation by cancer cells: Expectations, facts, and therapeutic implications. *Bioessays* **38**, 276–285.

TOMLINSON, I. P. M. & BODMER, W. F. (1997). Modelling the consequences of interactions between tumour cells. *British Journal Of Cancer* **75**, 157–160.

TRIGOS, A. S., PEARSON, R. B., PAPENFUSS, A. T. & GOODE, D. L. (2017). Altered interactions between unicellular and multicellular genes drive hallmarks of transformation in a diverse range of solid tumors. *Proceedings of the National Academy of Sciences* **114**, 6406–6411.

UJVARI, B., GATENBY, R. A. & THOMAS, F. (2017). Transmissible cancer: the evolution of interindividual metastasis. In *Ecology and Evolution of Cancer* (ed. B. Ujvari, B. Roche and F. Thomas), pp. 167–179. Academic Press, London.

VINCENT, M. (2012). Cancer: A de-repression of a default survival program common to all cells? *BioEssays* **34**, 72–82.

VINCENT, M. D. (2010). The animal within: carcinogenesis and the clonal evolution of cancer cells are speciation events sensu stricto. *Evolution* **64**, 1173–1183.

WEINBERG, R. A. (2013). *The Biology of Cancer*. Garland Science, New York.

WHITESIDE, T. L. (2008). The tumor microenvironment and its role in promoting tumor growth. *Oncogene* **27**, 5904–5912.

WHITESIDE, T. L. (2016). Exosomes and tumor-mediated immune suppression. *The Journal of Clinical Investigation* **126**, 1216–1223.

WOODMAN, C. B. J., COLLINS, S. I. & YOUNG, L. S. (2007). The natural history of cervical HPV infection: unresolved issues. *Nature Reviews Cancer* **7**, 11–22.

WU, C. I., WANG, H. Y., LING, S. & LU, X. (2016). The ecology and evolution of cancer: the ultra-microevolutionary process. *Annual Review of Genetics* **50**, 347–369.

YOGEV, O., HENDERSON, S., HAYES, M. J., MARELLI, S. S., OFIR-BIRIN, Y., REGEV-RUDZKI, N., HERRERO, J. & ENVER, T. (2017). Herpesviruses shape tumour microenvironment through exosomal transfer of viral microRNAs. *PLoS Pathogens* **13**, e1006524.

Table 1. Tumour traits that are problematic to explain by somatic selection.

Paradoxical tumour trait	**Definition**	**Problem with within-host selection**	**Can be selected within the host if:**
Distant niche construction	Factors released by the tumour transform a distant anatomical site to be permissive for metastases	Qualitative: 'cheater' clones of the tumour share the benefit, while only 'niche constructor' clones bear the cost	the constructed niche is preferentially accessible to the 'niche constructor' clones; or niche construction arises as a by-product of a locally beneficial trait
Long-range positive feedback loops	Factors released by the tumour activate a remote mechanism that promotes tumour growth by a long-range, spatially distributed effect	Qualitative: 'cheater' clones of the tumour receive equal share of the benefit, while only producer clones bear the cost	producer clones benefit preferentially from the growth feedback effect; or the long-range effect arises as a by-product of a locally beneficial trait
Local niche construction	Factors released by the tumour reprogram adjacent cells to form a supportive tumour microenvironment	Quantitative: 'cheater' clones share some of the fitness benefit, without paying the cost	spatial spreading of the tumour-promoting effect is strongly limited; and/or benefits are non-linear, creating frequency-dependent selection
Metastatic potential – phenotypic plasticity	Tumour cells acquire the ability to leave the tumour and colonize distant tissues, displaying adaptive phenotypic plasticity	Quantitative: antagonistic selection for efficient local growth is expected to dominate clonal evolution; few rounds of serial metastases	serial metastases occur more frequently than currently realized; and/or metastatic potential arises as a by-product of a locally beneficial trait

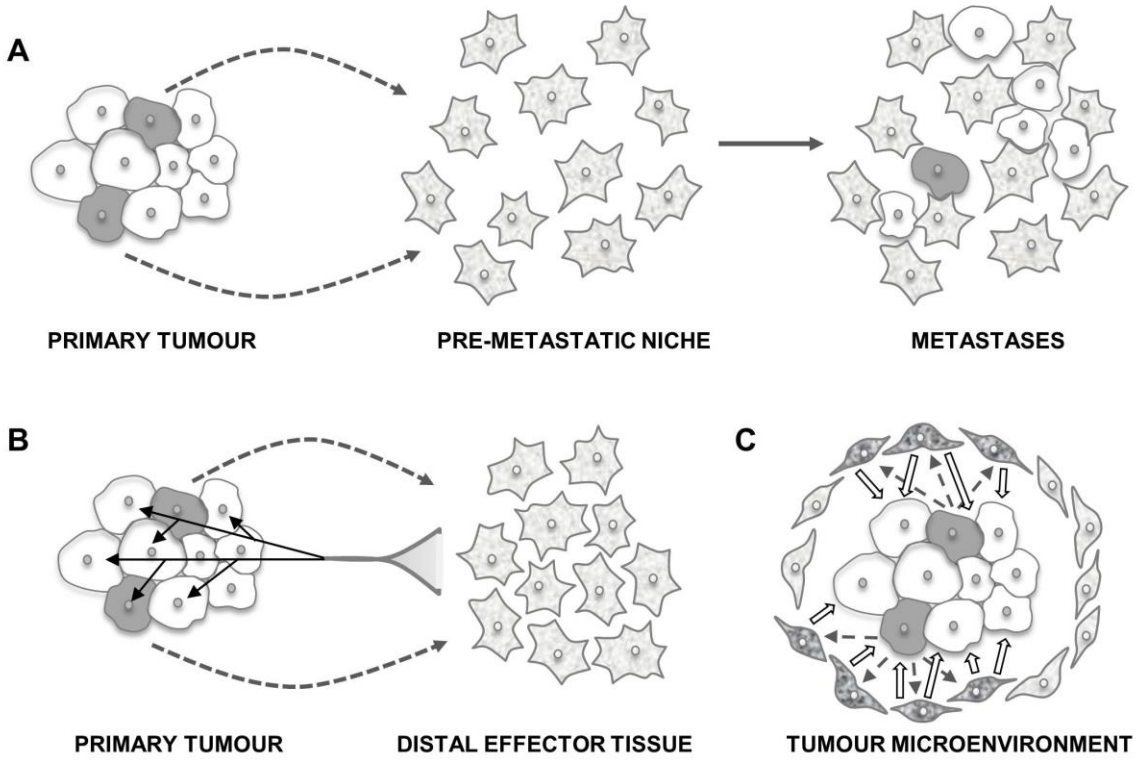

Fig. 1. Problems with somatic selection for non-cell-autonomous tumour traits. In distant niche construction (A), tumour cells release signals (dashed arrows) that target a distant anatomical site, re-programming cells to form a permissive microenvironment for the invasion of circulating tumour cells. If production of the signal imposes a metabolic cost, producer cells (shaded) are selected against in the local microenvironment, while in the colonization of the distant niche, non-producer cells receive equal benefit. Somatic selection is expected to eliminate the trait. In long-range positive feedback loops (B), tumour cells stimulate distal tissue (dashed arrows) to produce systemic factors that promote the growth of the tumour. If the cells that produce the stimulus (indicated by shading) receive the same growth benefit (solid arrows) in the tumour as non-producer cells, but bear the cost of producing the stimulus, somatic selection is expected to eliminate the trait. Finally, in the construction of the local tumour microenvironment (C), tumour cells re-program adjacent stromal cells to provide supporting functions that facilitate tumour growth. Both the re-programming signal (dashed arrows) and the growth promoting effects (block arrows) can spread in the local neighbourhood, weakening the relative benefit of the producer cells that alone bear the cost of the signals.

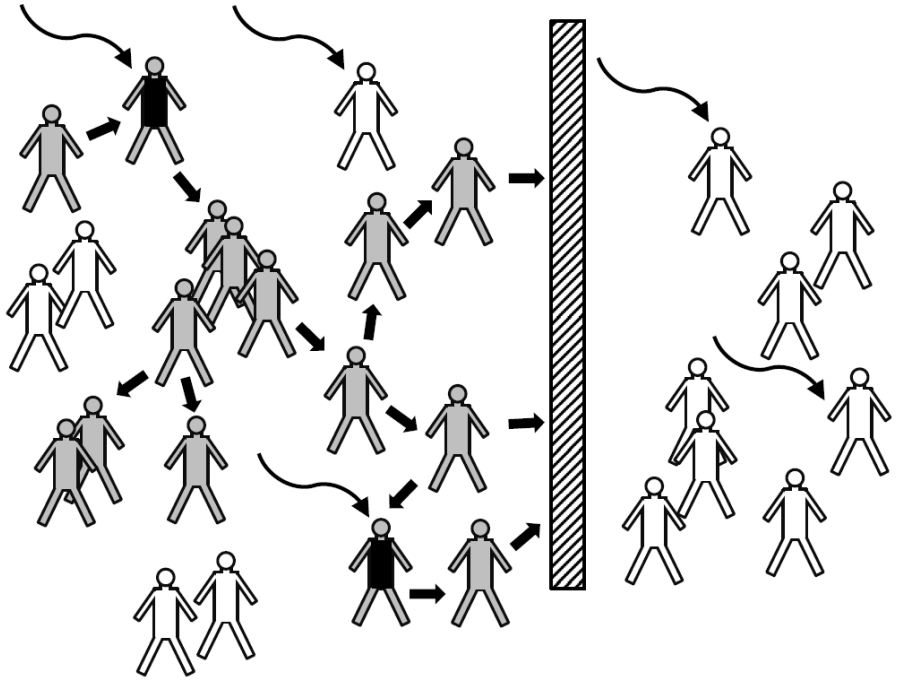

Fig. 2. Schematic representation of the epidemiology of cancer caused by the combination of a prevalent infection and additional factors. Chronically infected individuals are indicated by grey shading, block arrows denote past transmission events, and curly arrows represent the additional factor(s) necessary to trigger cancer. The rare cases of clinically manifest cancer (indicated by black shading of the body of the individual) appear only when infected individuals are exposed to the additional factor(s), while asymptomatic infections are common in the population. As a result, although infections spread along contacts, the cases of detectable disease do not cluster substantially in the network. An intervention that blocks transmissions (indicated by the striped rectangle) would nonetheless eradicate not only the infection, but also the cancer from the population, despite continued exposure to the co-factor(s). If the dependence of cancer on the infectious agent is not absolute, interventions blocking transmission reduce but do not eliminate cancer, while the linkage still remains hard to detect. The additional factors required for cancer might involve co-infections, other carcinogens, rare genetic predisposition, or low-probability mutation events.