Cancer as an evolutionary and ecological process

Lauren M.F. Merlo, John W. Pepper‡, Brian J. Reid§ and Carlo C. Maley**

Abstract | Neoplasms are microcosms of evolution. Within a neoplasm, a mosaic of mutant cells compete for space and resources, evade predation by the immune system and can even cooperate to disperse and colonize new organs. The evolution of neoplastic cells explains both why we get cancer and why it has been so difficult to cure. The tools of evolutionary biology and ecology are providing new insights into neoplastic progression and the clinical control of cancer.

Clone
A set of cells that share a common genotype owing to descent from a common ancestor. In some contexts a clone is more restrictively defined as a set of genetically identical cells.

Fitness
The average contribution of a genotype to future generations. Fitness is generally a function of both survival and reproduction.

*Cellular and Molecular Oncology Program, The Wistar Institute, 3601 Spruce Street, Philadelphia, Pennsylvania 19104, USA. ‡Department of Ecology and Evolutionary Biology, Biological Sciences West, University of Arizona, Tucson, Arizona 85721, USA. §Human Biology Division, Fred Hutchinson Cancer Research Center, PO BOX 19024, Seattle, Washington 98109, and Departments of Medicine and Genome Sciences, University of Washington, Seattle, Washington 98195, USA. Correspondence to C.M. e-mail: cmaley@alum.mit.edu
doi:10.1038/nrc2013
Published online
16 November 2006

Cancer is a disease of clonal evolution within the body[1–3]. This has profound clinical implications for neoplastic progression, cancer prevention and cancer therapy. Although the idea of cancer as an evolutionary problem is not new[1,4], historically, little attention has been focused on applications of evolutionary biology to understand and control neoplastic progression. That is now beginning to change[5–13].

A neoplasm can be viewed from an evolutionary perspective as a large, genetically and epigenetically heterogeneous population of individual cells. Genetic and epigenetic alterations that are beneficial to a neoplastic clone, enabling it to expand, are generally deleterious to the host, ultimately causing death to both the host and the neoplasm. Because these somatic abnormalities have differing, heritable effects on the fitness of neoplastic cells, mutant clones might expand or contract in the neoplasm by natural selection and genetic drift, regardless of any negative effects on the organism. The fitness of a neoplastic cell is shaped by its interactions with cells and other factors in its microenvironment (its ecology), including interventions to prevent or cure cancer. Clonal evolution generally selects for increased proliferation and survival, and might lead to invasion, metastasis and therapeutic resistance.

Three decades of research have broadly supported Nowell's description of cancer, in 1976 (REF. 1), as an evolutionary system. Since 1976, researchers have identified clonal expansions[14–17] and genetic heterogeneity[5,8,13,18] within many different types of neoplasms. However, many promising opportunities for the application of evolutionary biology to carcinogenesis and oncology remain unexplored. What are the rates of genetic and epigenetic changes in a neoplasm? How can we alter those rates? How do clones expand and what can we do to control such expansions? What are the relative fitnesses of various carcinogenic alterations? What are the selective effects of our therapies? Answering these questions will enable us to measure, manage and interrupt neoplastic progression and therapeutic relapse.

Here we examine cancer through the lens of evolutionary and ecological biology. We will review what is known about the evolution and ecology of neoplastic clones, examine the consequences of these dynamics and identify important missing pieces in the puzzle of neoplastic progression, its causes, prevention, and treatment of the resulting malignancies.

Levels of selection

Evolutionary forces work on many levels in biology[19]. Selection among somatic cells occurs on the timescale of a human lifetime. Selection on organisms, over millennia, has led to adaptations that constrain somatic evolution[4,20]. An analysis of the trade-offs in the conflicting levels of selection helps to reveal not only our natural defenses against cancer, but also the nature of some remaining vulnerabilities to cancer[2,21–24]. Organism-level and gene-level selection has led to the evolution of general tumour-suppression mechanisms (BOX 1) and oncogenic vulnerabilities in our genomes (BOX 2). This review will concentrate on selection and evolution in populations of cells, rather than individuals.

Mutation

Evolution requires heritable variation within the population. Various forms of mutation (defined broadly as any event that contributes to heritable variation between cells) have a role in neoplastic progression. Studies of heterogeneity in tumours clearly show that there is extensive cytogenetic, genetic and epigenetic variability in neoplastic cell populations, and the degree of variability can predict progression to malignancy[8,13,18,25]. For example, every genetically distinct clone detected in a Barrett's

At a glance

- Neoplasms are composed of an ecosystem of evolving clones, competing and cooperating with each other and other cells in their microenvironment, and this has important implications for both neoplastic progression and therapy.
- Selection at the different levels of genes, cells and organisms might conflict, and have resulted in a legacy of tumour-suppression mechanisms and vulnerability to oncogenesis in our genomes.
- Most of the dynamics of evolution have not been measured in neoplasms, including mutation rates, fitness effects of mutations, generation times, population structure, the frequency of selective sweeps and the selective effects of our therapies.
- Many of the genetic and epigenetic alterations observed in neoplasms are evolutionarily neutral.
- Cancer therapies select for cancer stem cells with resistance mutations, although various evolutionary approaches have been suggested to overcome this problem, including selecting for benign or chemosensitive cells, altering the carrying capacity of the neoplasm and the competitive effects of neoplastic and normal cells on each other.
- Dispersal theory suggests that high cell mortality and variation of resources and population densities across space might select for metastasis.
- There is evidence of competition, predation, parasitism and mutualism between co-evolving clones in and around a neoplasm.
- We will need to interfere with clonal evolution and alter the fitness landscapes of neoplastic cells to prevent or cure cancer. Evolutionary biology should be central to this endeavor.

oesophagus pre-malignant lesion was associated with an increased risk of progressing to oesophageal adenocarcinoma by a factor of 1.4, and every 10% of genetic divergence between clones was associated with a further risk factor of 1.6 (REF. 8). Because the genetic instability that generates genetic heterogeneity is a ubiquitous characteristic of neoplasms, and is fundamental to the processes of neoplastic progression, it should be recognized as a hallmark of cancer[26]. This heterogeneity poses a problem for the study and management of neoplasms because a biopsy sample might not be representative of the neoplasm, and the neoplasm continues to change after the biopsy sample is taken.

Genetic and epigenetic alterations are widespread in cancers. Stoler *et al.* estimated that there are at least 11,000 genomic alterations in the clone that generates a colon carcinoma, although many lie within non-coding regions[27]. Widespread loss of heterozygosity might be fertile ground for recessive mutations to emerge. How cells survive and even flourish with losses as large as whole chromosomes remains unresolved[28], although the sheer number of changes suggests that most are effectively neutral for the clone, and many might even increase its fitness[22,29,30]. Although it seems to be a relatively late event in neoplastic progression, the loss of *TP53* (the gene that encodes the tumour suppressor p53) normal cell cycle and apoptotic responses to chromosome breaks could confer such a large fitness advantage, by enabling cells to survive and divide, that the clone might be able to tolerate many deleterious mutations and still have a fitness advantage over p53 wild-type clones[29]. It might be that most genes in the human genome are devoted to building and maintaining a multicellular body, and are therefore irrelevant to a neoplastic cell under selection for increased survival and proliferation[22,31]. This might be analogous to organisms that switch from independent to obligate parasitic or mutualistic associations, like the ancestor of the mitochondrion, shedding genes that are no longer necessary for their new lifestyle[32].

Changes in methylation patterns can alter the expression of genes and, as the methylation rate is thought to be faster than the genetic mutation rate, epigenetic mutations might be more likely to initiate neoplasms than genetic mutations[33,34]. Hypermethylation has been shown to inactivate genes associated with DNA-damage response and repair, such as *MLH1*, *MLH3*, *MSH6* and *SFN*, in neoplasms[35,36]. In these cases, epigenetic instability probably leads to genetic instability. Therefore, the effects of many forms of (epi)genetic instability are layered on top of one another as neoplasms progress.

Rates of different types of somatic mutation have not been measured *in vivo*, although the rates themselves

Genetic drift
Random changes in allele frequencies over generations. This dynamic of random sampling has a greater effect in smaller populations.

Neutral mutation
A mutation that has no fitness effect (survival or reproductive effect).

Box 1 | Control of somatic evolution

Uncontrolled somatic evolution is a fundamental source of neoplasia, but organisms have also found ways to exploit somatic evolution to their benefit. This is most evident in the adaptive immune system, which uses controlled clonal selection to defend against cancer[136]. Somatic selection is also harnessed as a mechanism for efficiently eliminating (through apoptosis) any cells that are inappropriately proliferating or that have activated oncogenes[43,55,137].

Although some forms of somatic selection are harnessed by the organism for protection against cancer, the simplest and most wide-spread defense against cancer might be to suppress selection among cells where possible. Two primary mechanisms are thought to have key roles in the suppression of somatic selection: cellular senescence and cell differentiation.

If the number of cell generations is limited by senescence, this also limits the potential for multistage somatic evolution that underlies carcinogenesis. The dilemma faced by natural selection among organisms is how to enforce cellular senescence without creating organismal senescence that would reduce organismal fitness[23].

Similar to cellular senescence, cell differentiation limits the number of cell generations within any given cell lineage (FIG. 1). If a finite number of cell generations pass before the lineage ends in fully differentiated and non-dividing cells, this also limits the potential for multistage carcinogenesis[138]. In addition, rapidly dividing epithelia, like the skin and gastrointestinal tract, continuously shed these cells from the body. To enable the renewal and maintenance of the organism, each tissue must also include non-differentiating somatic stem cells as a sustained source of new cells[139].

Given the existence within a tissue of both reserve cells and differentiating cells, the problem arises of how to organize these cell types in such a way as to minimize the risk of tumorigenesis. This optimization problem includes the relative number of reserve cells versus differentiating cells[4,140,141]. It also involves the tissue architectures that subdivide cell populations and thereby help to limit clonal expansions[4,7,140,142].

Box 2 | **The evolution of cancer-susceptibility genes**

The maximization of fitness often involves trade-offs between different selective forces. In some cases, a germline oncogenic mutation, an allele that is particularly vulnerable to an oncogenic mutation, or an allele that disrupts tumour-suppressor gene networks, might spread in a population if the selective effects of cancer are overwhelmed by other fitness benefits of the mutation.

BRCA1 mutations seem to be more prevalent than would be expected given their carcinogenic effects on fitness and the generation of new BRCA1 germline mutations[143]. Positive selection has been detected in the RAD51-interacting domain, which is important in the response of BRCA1 to DNA damage, although why there would be diversifying selection on DNA-damage response is unknown[144]. BRCA1 alleles that predispose to breast cancer seem to have originated surprisingly recently, implying strong selection against them that probably cannot be explained by their carcinogenic effects[145]. BRCA1 is involved in the spindle checkpoint, many cell-cycle checkpoints, the DNA-damage response and development[146,147]. In addition, the high density of Alu repeats[148] increases the probability of somatic mutations in BRCA1, and might indicate conflicting selection between retrotransposons[149] and the host.

In development, cadherins contribute to epithelial differentiation, embryonic implantation and placenta formation, and in adults they form adherens junctions[150]. Cadherins, particularly E-cadherin, are commonly lost in cancer and are associated with an invasive, metastatic phenotype. A comparison of cadherins between vertebrates suggests that some members of the cadherin family, those expressed during embryonic and/or fetal development, are subject to diversifying selection in humans[150].

A survey of evidence of recent selection in the human genome has implicated several genes that are associated with both cancer and spermatogenesis[151]. Crespi and Summers suggest that genes that are the subject of ongoing genetic conflict will both tend to show recent evolution and might be associated with cancer risk because the fitness effects of the genetic conflict overwhelm the selective effects of cancers that develop after reproduction[152]. These evolutionary conflicts might also play out through epigenetic imprinting, which has been shown to have dramatic carcinogenetic effects[153].

would be fundamental biomarkers of progression and risk stratification, as well as tools to measure the effects of interventions. Knowledge of mutation rates would enable us to develop better surveillance protocols for high-risk patients. Mutation frequency studies and measurements in cell culture put the sequence mutation rate at 10^{-6}–10^{-7} per locus per cell generation[37,38]. Although genetic instability is a hallmark of cancer, an increase in mutation rate might not always be beneficial, as most non-neutral mutations are thought to be deleterious[39]. In bacterial experiments, mutator phenotypes have emerged, although they did not evolve more quickly than non-mutator populations[40]. Breivik has shown that the type of environmental insults (for example, methylating agents or bulky-adduct-forming carcinogens) select against the checkpoints that they trigger, because cells that lose those checkpoints can reproduce more quickly than those that stop to repair the damage[29]. Therefore, the mutator phenotype might be selected owing to its effects on cell cycling rather than its generation of further advantageous mutations.

The number of mutations necessary and sufficient to cause cancer is unknown, even for retinoblastoma[41]. Estimations range from 3–12 mutations for different forms of cancer[42]. Organs with many cells and rapid turnover require more mutations[42,43]. Loeb[44] argued that the spontaneous rate of somatic mutation is not high enough to generate so many mutations in a cell. To resolve this paradox, two hypotheses have been proposed: either a genetically unstable phenotype might arise that increases the mutation rate[44], or the expansion of clones generates target populations large enough to produce the necessary subsequent mutations[45,46]. The two hypotheses are not mutually exclusive[47], and we have shown that the clonal expansion of genetically unstable clones predicts progression to oesophageal adenocarcinoma[48]. Determining exactly which mutations are necessary and sufficient to generate a cancer is important to help identify targets for cancer prevention, as well as biomarkers for risk stratification and early detection.

Neutral mutation and genetic drift

Changes in allele frequencies due to stochastic processes (BOX 3) might contribute to cancer progression. In small populations, chance might have an important role in altering allele frequencies. In general, parameters crucial for understanding the role of genetic drift in cancer progression have not been measured. These include the effective population size (the actual number of cells that contribute to future generations; N_e), cell generation times and cell turnover (the frequencies of cell division and apoptosis).

Genetic drift is intimately related to the selective advantage or disadvantage of a particular mutation and the size of the population of cells. Some mutations might have no selective effect and are considered neutral. If a particular mutation has a selective advantage much less than $1/N_e$, genetic drift is still the predominant force. Therefore, the definition of a neutral mutation is related to the type of mutation, the selective advantage and the population size[49].

Crucial to determining the effective population size, N_e, is an understanding of the role of cancer stem cells[50–52] and normal stem cells[53] during carcinogenesis. Intestinal crypts seem to contain only a few stem cells, making the effective population size very small[54] (FIG. 1). Therefore, neutral and even deleterious (for example, genetic instability) mutations in stem cells might drift to fixation in a crypt[55].

The random loss or fixation of alleles might occur through reductions in cell population sizes ('population bottlenecks'). This can occur normally in the body, for example, through the apoptosis of breast epithelium during the menstrual cycle, in disease processes such as repeated wounding in ulcerative colitis and Barrett's oesophagus, and in cancer therapies. Mutations early in development can also generate large clones ('jackpots'), and these are predicted to have a significant affect on cancer incidence[56,57].

Many of the mutations seen in neoplasms seem to be neutral. There is evidence that many clones can coexist for a long time[5,6], suggesting that the mutations that distinguish these clones might be evolutionarily neutral, although there are many mechanisms that enable competitors to coexist (BOX 4). In addition, large numbers of neutral hitchhiker mutations[58] ('passengers') might be carried to fixation by adaptive mutations[16].

Determining which carcinogenic mutations are neutral versus advantageous, depending on particular contexts of the microenvironment, will help predict clonal expansions and identify how we can change the microenvironment to make a carcinogenic mutation neutral or deleterious and prevent clonal expansion.

Fixation
When an allele (or in this case a clone) reaches 100% frequency in a population.

Hitchhiker mutation
An effectively neutral mutation that expands in a population because it is linked to a selectively advantageous allele. Sometimes called a 'passenger mutation' in cancer biology.

Box 3 | **The theory of genetic drift**

In genetic drift, individuals can leave different numbers of offspring by chance rather than fitness differences. Given enough time in a population of constant size, one clone will go to fixation and all others will go extinct. Therefore, if there are N cells, representing N clones, each clone has $1/N$ chance of reaching fixation. Furthermore, assuming a Moran model[154] in which cells divide and die asynchronously, the expected time it takes for a clone to expand from a single cell to fixation is $N(N\text{-}1)$ total cell divisions[155]. Clinically detected neoplasms are often 10^9–10^{12} cells, so the chance of fixation by genetic drift is vanishingly small, and the time that would take is far longer than a human lifetime.

These results assume populations of constant size and overlapping generations (Moran model[154]) with no recombination, no population sub-structure and no fitness effects of mutations. Populations that violate some of these assumptions often behave as idealized populations of a different size N_e, called the effective population size. In a neoplasm, the total number of cells can be much larger than the effective population size owing to differentiation, limited replicative potential, changes in population size and the occurrence of selective sweeps.

A mutant is likely to go extinct even if it has a selective advantage. Therefore carcinogenic mutations might appear and go extinct many times before one is lucky enough to attain a population size that is no longer in danger of going extinct by genetic drift alone. For example, a mutation in a stem cell with a 10% fitness advantage over its competitors (fitness advantage: $s = 0.1$) would have a 91% chance of going extinct by genetic drift before it could sweep to fixation in a population of $N_e = 10^6$ stem cells[155]. In a neoplasm with $N = 10^9$ cells and $N_e = 10^6$ stem cells, there is only a 1 in 1,000 chance that the mutation occurs in a stem cell, and so the chance of extinction increases to 99.99%. If a new mutation has a relative fitness advantage, the chance of extinction is shown by equation 1 (adapted from REF. 155).

$$1 - P = 1 - \frac{1 - (1 + s)^{-1}}{1 - (1 + s)^{-N_e}}\left(\frac{N_e}{N}\right) \quad (1)$$

In addition, identifying neutral mutations might enable us to use them as a molecular clock to determine the time since the initiation of a neoplasm[5,6].

Natural selection

The heritable variation of reproductive success in a population is necessary and sufficient to cause natural selection[59]. Natural selection occurs in neoplasms because (epi)genetic mutations generate heritable variation, and some mutations confer a selective advantage or disadvantage on the cell. All the hallmarks of cancer[26] lead to the differential reproductive success of a clone. These fitness advantages will be amplified in tissues with repeated wounding, in which repeated cycles of cell death and proliferation enable a mutant clone with a survival or reproductive advantage to expand.

The presence of proliferating and apoptotic cells in neoplasms implies that clones can expand and contract. Mutations that increase the fitness of a clone might lead to a selective sweep through the population of cells, eventually reaching fixation in the neoplasm (FIG. 2). In most cases, it is unknown how clones expand through a neoplasm and if there are population sub-structures that inhibit those expansions. Both clonal expansions[14–17] and carcinogenic exposures might explain field effects in carcinogenesis[60]. The expansion of a pre-malignant clone that seems histologically normal can predispose a large region to further progression and result in multi-focal and locally recurrent cancers[15]. Clonal expansions driven by epigenetic mutations have not yet been established. If a clonal expansion is driven by the mutation of a tumour suppressor or oncogene (a hypothesis often tested *in vitro* but rarely *in vivo*[14,16]), then those lesions are good candidates for biomarkers of progression because they are causally related to cancer outcome and can be easily sampled.

A crucial unresolved question is why patterns of gene loss and/or gain differ between cancers in different organs and cell types? It will be important to understand selective pressures in different organ environments. In addition, whether or not a gene is used in a normal cell type will affect the fitness of the cell with mutations in that gene[61].

Mutations in some genes are only advantageous to the clone after there has been a lesion in another gene. For example, in Barrett's oesophagus, the inactivation of *TP53* is almost always observed after the inactivation of *CDKN2A* (the gene that encodes the tumour suppressor INK4a)[16]. It is possible, in a case like this, that a mutation that is neutral on its own could expand by genetic drift before a second mutation in that clone makes the first mutation selectively advantageous. However, it is more probable that the mutation that is selectively advantageous on its own (for example, in *CDKN2A*) will initiate a clonal expansion that creates many opportunities for the other mutation (for example, in *TP53*) to occur, sparking a second clonal expansion within the first. Such genetic dependencies (BOX 5) lead to regularities in the order in which mutations appear. Linear[62] and tree models[63] of progression that implicitly rely on genetic dependencies and their predictive value[64] might be improved by testing the implied dependencies.

Artificial selection

Cancer therapies often select for resistance, caused by various mechanisms, which is the central problem in cancer therapy. At relapse, mutant clones have been discovered in lung cancer with point mutations in epidermal growth factor receptor (EGFR) that cause resistance to anilinoquinazoline EGFR inhibitors[65]. In chronic myeloid leukaemia, an amino-acid change in BCR-ABL confers resistance to imatinib (Glivec)[66], and amplification of the thymidine synthase gene causes resistance to 5-fluorouracil in colorectal cancer[67]. This shows that therapies do not simply select for cancer stem cells[68], but also cancer stem cells with resistance mutations[69].

The number of cell divisions (and the potential for mutational events) before therapy far outweighs those after therapy. A classic early experiment in evolutionary biology[70] tested whether the exposure of a bacterial population to a selective pressure (the presence of a phage) caused new mutations, or if applying the pressure selected for pre-existing mutants. The second case proved to be true. The same principle is expected to apply to cancer, although mutagenic therapies might generate resistance mutations[71]. There is evidence for resistance mutations before the application of Glivec[72]. The implication is that the earlier we intervene in progression, the less probable it is that a resistant mutant will emerge[69].

Cancers that develop without selection for genetic instability or enough time to produce much genetic heterogeneity should be unlikely to harbour a resistant clone[73].

Molecular clock
When mutations occur at a constant rate, the number of mutations that have accumulated between two different lineages is representative of the time since the lineages diverged.

Selective sweep
The process of an adaptive mutation spreading through a population, typically ending in fixation.

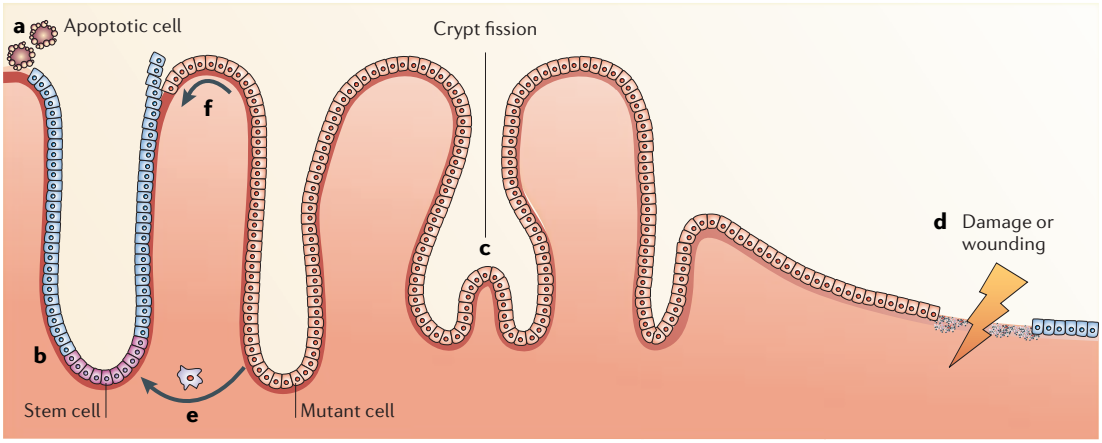

Figure 1 | **Intestinal tissue architecture and sub-population structure. a** | Cells differentiate as they move up the crypt, and eventually initiate apoptosis and slough into the lumen. **b** | Each crypt is continually renewed by a small number of long-lived stem cells that reside near the bottom of the crypt. Therefore, crypts sub-divide the epithelium into isolated sub-populations. In some conditions[16], mutant clones (red) can expand over many crypts, although the mechanism of this expansion is unknown. **c–f** | Hypotheses include: crypt fission (**c**); wounding with epithelial restitution (**d**); dispersal through the basement membrane and stroma into the base of neighbouring crypts, perhaps through epithelial–mesenchymal–epithelial transitions (**e**); and, more speculatively, dispersal over the surface of the epithelium (**f**), along the basement membrane, and then down into neighbouring crypts against the flow of cells emerging from the crypt (which might require differentiation and then dedifferentiation).

This is probably the case for most pre-clinical models of cancer. If only a few mutations are required to produce a clinically detectable neoplasm, then the neoplasm is less likely to be genetically diverse, and so is less likely to harbour a resistant clone compared with a neoplasm that must accumulate many mutations before it is detected. Many childhood cancers seem to require few mutations[42,74]. In cases such as retinoblastoma, there are few cells vulnerable to progression, and they are only vulnerable for a short period of time. Therefore, only a few tumour-suppressor genes are required to prevent retinoblastoma in most children[43]. The importance of detecting a neoplasm before wide-spread genetic heterogeneity develops is consistent with clinical experience that shows increased survival with the detection of early-stage disease[75].

There are several possible evolutionary approaches to cancer therapy and prevention that could address the problem of therapeutic resistance. These include multi-drug therapies[76], therapies that work to alter competition between cancerous and non-cancerous cells by boosting the fitness of benign cells[10], selection for chemosensitivity[10], selection for genetic stability[77] and the induction of crippling bottlenecks. Of these strategies, only multi-drug therapies have been explored experimentally and/or clinically[76]. The way a therapy is applied might also affect the evolutionary dynamics in a neoplasm. Evolutionary experiments show that the application of selective pressures in pulse versus continuous treatment can alter the outcome of competition[78]. Traditional chemotherapies are applied in large pulsed doses, but evolutionary theory, and evidence from anti-angiogenic therapy[79], suggests that lower, continuous doses might work better. Neoplastic cell populations that expand between doses might generate new resistance mutations[79]. In addition, under pulses of a therapy, the fitness of a neoplastic cell is the average of its fitness during therapy and its fitness between doses, weighted by the duration of those conditions. This is likely to be higher than the fitness under a lower but continuous dose, although pulses of extremely high doses have also been shown to be efficacious in some cases[80].

The population bottleneck caused by cancer therapy might be able to cripple a neoplasm. Following therapy, many patients with leukaemia show minimal residual disease, in which a very small population of leukaemic cells remain as a stable subpopulation, and do not grow exponentially as would be characteristic of cancer[81]. One hypothesis for the population stability is that the characteristics selected by chemotherapy might also interfere with proliferation. For example, if a cancer drug only kills proliferating cells, then quiescent cells might survive the treatment and remain quiescent thereafter[81]. Alternatively, if the bottleneck is small enough, cells with fitness disadvantages can become fixed in the neoplasm by genetic drift. Because the rate of evolution is very slow in small populations, it might take a very long time before a leukaemic clone acquires mutations that enable it to expand again.

Dispersal and colonization

Allele frequencies can change (evolution can occur) through dispersal. There are at least three ways in which dispersal can be important in cancer: the movement of cells between the partially isolated sub-populations of proliferative units, local invasion of neighbouring tissues and emigration of metastatic cells from the primary tumour.

The epithelium of most organs is organized into proliferative units such as crypts in the intestine (FIG. 1), acini in liver and breast, proliferative units in squamous epithelium and so on. These proliferative units form

Box 4 | **Mechanisms of coexistence**

Cells in a neoplasm seem to compete for the same resources, space and nutrients, and so we would predict that a clone with a fitness advantage should drive other clones extinct as it sweeps to fixation. However, there is evidence that clones can coexist for many years[5,6] (FIG. 2), and that clonal diversity might increase with progression[8]. How can more than one clone stably coexist in a neoplasm? Ecology and evolution suggest various mechanisms:

- Mutations might be evolutionarily neutral, providing no fitness advantage, and therefore no selective sweep.
- Fitness might be density dependent, so that as a clone becomes more frequent in the population, its fitness decreases. This might be caused by an immune reaction (predation), one clone gaining a fitness benefit by proximity to another clone (parasitism), or pollution of its environment by metabolic byproducts.
- Niches: clones might specialize on different resources or different microenvironments, and thereby reduce their competition[107].
- If the environment fluctuates faster than any one clone can reach fixation, then clones adapted to the different environments could coexist in non-equilibrium.
- Clones might be physically separated, and therefore unable to invade each other's territory[4] (FIG. 1).
- The total population might be expanding, therefore reducing competition for space[13].

Which, if any, of these mechanisms are at work in neoplasms is an important open question in cancer biology.

semi-isolated sub-populations, which typically include a small number of stem cells and a larger number of transient amplifying and fully differentiated cells[4,54]. The observation of clonal expansions[16,82,83] implies that some mutants can breach the barriers between proliferative units. In most cases, we do not know how clones expand (FIG. 1). In the skin, UV light can destroy proliferative units that might be reconstituted by neighbouring mutants[14]. Does clonal expansion always require some form of wounding, or is there normal turnover of proliferative units? Mutants might also spread by dispersal between proliferative units.

Box 5 | **How to study evolution in neoplasms**

The study of evolution rests on measuring changes in the frequency of (epi)genetic variants in a population. This requires measuring different clones in a neoplasm, which in practice entails:

- Isolating the cell population of interest.
- Extracting and assaying DNA in the purified population.
- Measuring the frequency of (epi)genetic lesions in the DNA.

If clones in the neoplasm can first be separated (for example by flow cytometry), then patterns of (epi)genetic alterations can be associated with specific clones, and frequencies of those lesions can be measured by the frequencies of the clones in the neoplasm. The easiest way to separate clones is to take more than one biopsy from a neoplasm, separated by space, and to analyse each biopsy separately.

Analysing several biopsies from a neoplasm also enables the powerful but under-used technique of genetic-dependency analysis (clonal ordering) to be used, in which the order in which genetic lesions arose can be inferred from the spatial patterns of shared lesions[156]. That is, if one biopsy has lesions in loci A and B, and another biopsy only has a lesion in locus A, we can infer that the lesion in locus A probably occurred first, was associated with a clonal expansion, and the lesion in locus B occurred later. If this pattern occurs in many neoplasms, it is evidence that lesions in locus B are only selectively advantageous in cells with the locus A lesion, and so there is a genetic dependency between the lesions.

Tracking clones as they evolve over time would be even better than clonal ordering from single time points. Such studies have already been reported from several conditions, including oesophageal squamous-cell carcinoma[157], Barrett's oesophagus[8,158–160], oral leukoplakia[161,162] and ulcerative colitis[163,164]. Serial biopsies can also be obtained during randomized trials to prevent or treat some cancers, for example, gastric[165] and prostate cancer[166]. A randomized trial offers the opportunity to observe clonal adaptation to the intervention, which might provide valuable information in designing new trials even if the original was unsuccessful.

This 'local metastasis' hypothesis could explain genetically related multi-focal tumours in some tissues, but has not been rigorously tested[84].

Metastasis requires that cells leave the primary tumour, but few such cells successfully colonize a distant organ[85]. This leads to a paradox: metastatic clones should have a fitness disadvantage relative to non-metastatic clones in the primary tumour owing to the loss of the progeny that emigrate. How could a metastatic clone expand and produce enough metastatic cells to successfully colonize a distant site[86,87]? Early[87] or late in progression[88], mutations that confer a metastatic phenotype might also provide a fitness advantage within the primary tumour that can compensate for the loss of emigrating progeny. Alternatively, metastatic mutations might be hitchhikers on selective sweeps, and their phenotype might be triggered later by a change in the tumour environment. An analogous example of compensating pleiotropy can be found in the evolution of ageing, in which a mutation that increases fitness before reproduction might be advantageous to the organism even if it causes decreased fitness later in life[89]. Hitchhiker mutations that reach fixation and then become deleterious with a change in environment are difficult to observe.

Within a single population of organisms, there is selection against dispersal. The main selective advantage of dispersal is the colonization of new populations[90]. Colonizing individuals often have high fitness because they can escape from deteriorating local conditions caused by population growth and the over-consumption of resources. The high density[91] and necrotic centres[92] of most solid neoplasms suggest that space and nutrients are limited. This leads to fierce competition, so there might be selection for dispersal.

Other conditions also select for dispersal, including high mortality rates, the variation of resources across space (for example, because of neoangiogenesis) and time (for example, because of wounding), and even stochastic fluctuations in local population densities[93]. If neoplastic cells, like many organisms, face trade-offs between local competition and dispersal, then local

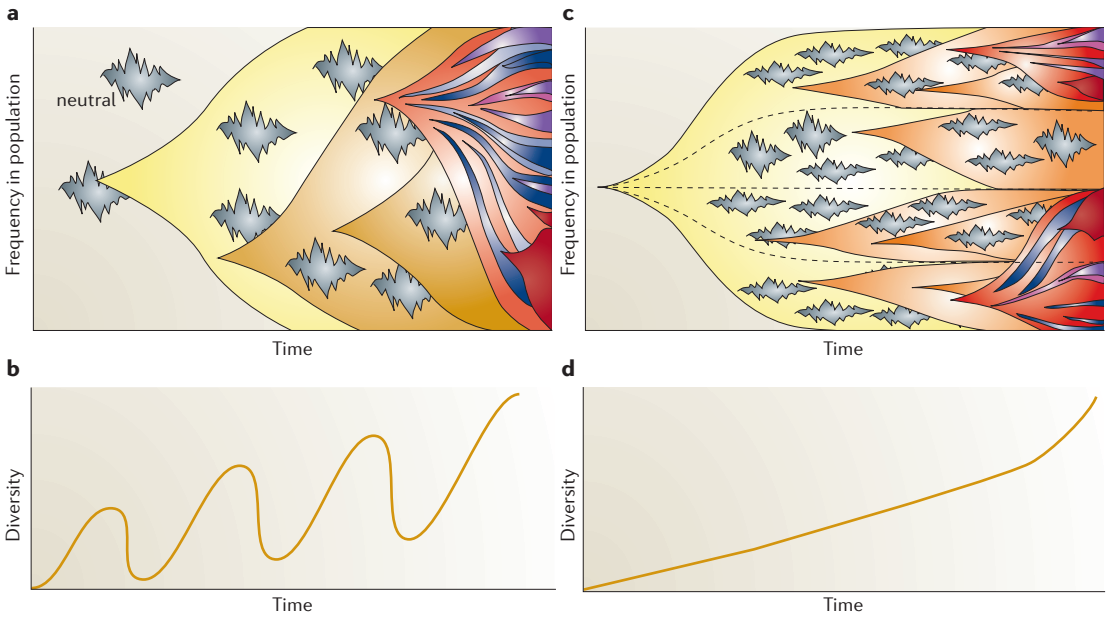

Figure 2 | **Asexual evolution in neoplastic progression.** Frequency within a neoplasm is shown on the Y-axis and time on the X-axis. **a** | If a neoplasm acts like a single population of cells, then an adaptive mutant can sweep through the population and become fixed (yellow, orange and red clones). Multistage carcinogenesis is thought to represent a series of such selective sweeps. The emergence of a clone with high levels of genetic instability (red) might accelerate the generation of new clones. **b** | Genetic diversity should fluctuate, increasing as genetic instability generates new clones and decreasing when a clone homogenizes the neoplasm in a selective sweep. **c** | If the neoplasm is divided into sub-populations (dashed lines) or there is a diversity of microenvironments that create different niches, then selective sweeps will tend to be constrained within a sub-population or niche, although they might occasionally invade a neighbouring sub-population. **d** | In a sub-divided population, total diversity might increase over time because selective sweeps cannot homogenize the entire population. Figure modified with permission from REF. 16 © (2004) American Association for Cancer Research.

therapeutic interventions that penalize cell proliferation, such as radiotherapy, will favour the ability to metastasize over the ability to compete within a neoplasm. It might even be possible to select against the emergence of metastasis (and resistance[73]) by relaxing these constraints on a neoplasm, but this remains to be tested in preclinical models and might be difficult to translate to the clinic.

The seed and soil hypothesis[94] suggests that metastasis is analogous to the colonization of a new habitat. Success at colonization of an ecosystem seems to depend on the characteristics of the invader[95], the climate[96], available space and resources in the new ecosystem and the configuration of native organisms[97]. A predictive model of metastasis might benefit from the identification and measurement of similarities in the 'climate' (microenvironment) between organs. There is some evidence that polyploidy in plants, and perhaps aneuploidy in neoplasms, is associated with an ability to invade new environments[98], perhaps owing to an increased opportunity for mutations, deletions and genetic rearrangements with the presence of extra alleles[99]. Some ecological studies have supported the hypothesis that increasing species complexity in an ecosystem facilitates further invasions[97]. The relationship between cell type complexity in an organ and its colonization by metastases has not yet been studied, but a recent experiment in *Escherichia coli* suggests that colonization by a superior competitor is more probable in a genetically diverse population than a community with few genetic variants[100].

Ecology

Ecology studies the dynamics of communities of species and their interactions. Ecological interactions can be classified by their fitness effects on the interacting individuals (FIG. 3). Examples of many different ecological interactions can be found in neoplasms, and most of these deserve further study.

Competition. For neoplastic cells in a heterogeneous population, competition exists in the form of resource consumption (oxygen for example). However, neoplastic clones can also have direct negative effects on each other through unknown soluble factors[101,102]. Neoplastic clones injected into opposite flanks of mice[103] and rats[104] can inhibit each other's growth, although in some cases the inhibition only affects one of the clones, and so is an amensal interaction (FIG. 3). Apparent competition can also occur in neoplasms in which one clone can stimulate an immune response that clears other clones and the immunogenic clone.

Carcinogenesis models based on Lotka–Volterra competition equations define conditions under which cancerous cells might be driven extinct. These include reducing the number of cancer cells that can be supported in the tissue, reducing the negative competitive effects of cancer cells on normal cells and increasing the

Amensal
An interaction between individuals that decreases the fitness of one party but has no effect on the other.

Lotka–Volterra competition equations
The Lotka–Volterra model of competition is based on logistic growth equations of two populations that negatively affect each other's growth.

negative competitive effects of normal cells on cancer cells[12]. These and other models can help define the parameters that must be targeted by therapies and the most effective methods for drug treatment regimens[11,105–107].

Predation. Predator species have negative effects on their prey, and gain some growth and reproductive benefit in return. Models of predation might be applicable to the interaction of neoplastic cells with the immune system.

Neoplasms evolve various mechanisms to escape predation from the immune system, including downregulation of the major histocompatibility complex[108]. The various mechanisms that a neoplasm can use to escape the immune system suggests that immune therapies are unlikely to work except in neoplasms with little genetic heterogeneity. Activated cytotoxic T lymphocytes do not directly benefit from the destruction of neoplastic cells, although they clonally expand in response to activation by antigen-presenting cells, and so the end result is the same. One dissimilarity here is that a predator will go extinct if its prey goes extinct. This is clearly not the case for T cells if they clear the neoplasm.

Minimal residual disease might be understood in terms of a predation model or a population bottleneck as discussed above. It is possible that residual neoplastic cells are not quiescent and are continually culled by the immune system[109]. If activated lymphocytes and neoplastic cell populations fluctuate in a typical predator–prey dynamic, we might be able to drive the neoplastic cells extinct by amplifying the fluctuations, perhaps by increasing the time lag between neoplastic clonal expansion and immune response. In populations of organisms, chaotic population fluctuations can be an effective source of local extinctions[110].

Parasitism. Parasitism is similar to predation, in that one species benefits at the expense of the other, although parasites often produce many offspring without killing their host. There is little evidence of clones within a neoplasm parasitizing each other. However, there is ample opportunity for clones to be free-riders on the metabolic investments of their neighbours, such as stimulating associated fibroblasts to release growth factors, stimulating neo-angiogenesis or the breakdown of the extracellular matrix and the release of growth factors contained within[107,111], and so on. Such parasitism between lineages is known in microbes[112] and viruses[113], and can be referred to as a 'cheater strategy'[114] because the parasitic clones gain a fitness benefit from their neighbours at no cost to themselves.

Mutualism and commensalism. Little is known about cooperative (mutualistic and commensal) relationships (FIG. 3) within neoplasms. However, Heppner, Miller and others have shown that a mutant clone can increase the fitness of other clones in commensal interactions, and even confer a metastatic phenotype on an otherwise non-metastatic clone[3,103,115]. Axelrod *et al.* have proposed that clones in a neoplasm could cooperate through diffusible factors, and thereby circumvent the requirement that a single clone has to accumulate all the hallmarks of cancer[116]. To date, the only known case of mutualism in a human neoplasm is the relationship between neoplastic epithelium and activated fibroblasts, both of which get a fitness advantage from the association[117–119] and seem to be co-evolving[120–122].

Mutualistic
An interaction between individuals that increases the fitness of both parties.

Commensal
An interaction between individuals that increases the fitness of one party and has no fitness effect on the other.

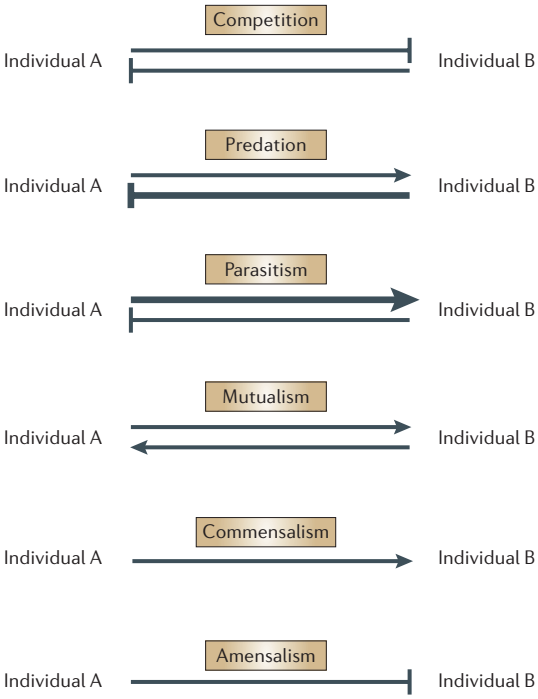

Figure 3 | **Ecological interactions.** Ecological interactions can be classified by the fitness effects of the individuals (neoplastic cells) on each other. Fitness effects can be positive (arrow) negative (closed arrow) or there might be no effect (no arrow). There are many mechanisms that can result in the different types of interactions[167], even in a neoplasm. For example, parasitism and predation are distinguished by the size not the type of the effects (sizes of the arrows), and clones might compete through the consumption of resources or by inhibiting each other through cell signalling[102]. Indirect interactions between clones might occur through direct interactions with a third type of cell, as in the case of a neoplastic clone reducing the fitness of another clone through the stimulation of an immune response[101,103,104,168].

The environment

The microenvironment of neoplastic cells has a dramatic effect on progression[123]. Placing teratocarcinoma cells in a mouse blastocyst is enough to suppress their carcinogenic phenotype[124]. Metastasis can be suppressed by the injection of a metastatic cell line into a heterotopic site[125]. Conversely, normal mammary epithelial cells can in some cases develop into invasive carcinomas in an environment that mimics activated stroma through the overexpression of hepatocyte growth factor (HGF) and/or transforming growth factor β1 (TGFβ1)[126]. Increased expression of HGF or stromal cell derived factor 1 (SDF1) by fibroblasts promotes epithelial

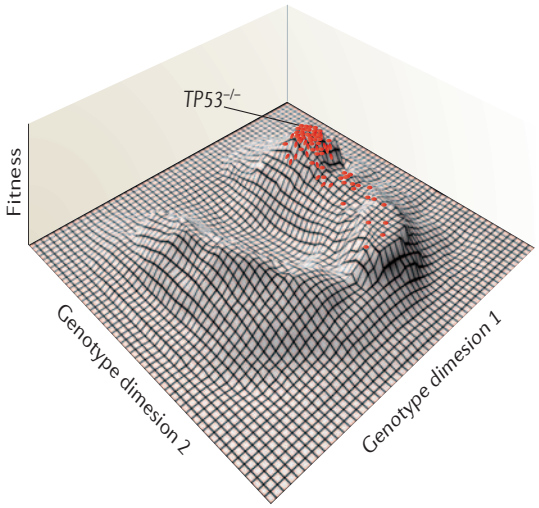

Figure 4 | **Evolution of a neoplastic population.** A highly simplified representation of a neoplastic cell population as a cloud of points evolving on a fitness landscape. Here, genotype is represented in the X and Y dimensions, and fitness is represented in the Z dimension. Locations are connected if a mutation (any possible genetic alteration) can change one genotype into a neighbour genotype. Evolving populations will typically move up fitness gradients by natural selection and only descend by mutation and genetic drift. Regions of neutral mutations are plateaux in a fitness landscape. Regularities in neoplastic progression reflect regularities in the fitness landscape. For example, if the points that represent genotypes missing *TP53* have high fitness, neoplasms will often evolve loss of *TP53*, although the paths they take to that region of the fitness landscape might differ. The fitness of genotypes, and therefore the topography of the fitness landscape, depends on the local microenvironment, including the ecology of other cells present. Interventions can be visualized as deformations of the fitness landscape.

neoplasms in mice[127,128]. These experiments show that we can modulate progression by altering the neoplastic environment.

Repeated, moderate disturbance of cell populations might select for genetic diversity and progression. With too little disturbance the environment is relatively homogenous, and the best competitors drive weaker competitors extinct. Too much disturbance wipes out populations entirely[129]. Perhaps chronic wounding promotes neoplasms by providing a diversity of micro-environments at different stages of recovery.

Differences from organismal evolution

The evolution of neoplasms differs in important ways from the ecology and evolution of organismal populations. Many of the formulae and phenomena analysed in evolutionary theory concern sexually reproducing species. Neoplastic cells are like asexual, single-celled organisms with limited horizontal transfer of genes within the neoplasm[130] and few life-history changes once differentiation has been abrogated. Asexual reproduction of neoplastic cells means there is no meiotic recombination, no Hardy–Weinberg equilibrium of genotypes in the population and no sexual selection. Different cell types in the body are unlike species in that a stem cell can differentiate into any cell type, and non-stem cells might be able to trans-differentiate into different types[131]. The relatively short time frame, and the large-scale genomic alterations in neoplastic progression suggests that neoplastic cells will be unable to evolve complex adaptations to their environment. Most neoplastic mutations seem to remove pathways that suppress proliferation or trigger apoptosis[26,31], or co-opt pathways normally used in development and wound healing.

Parallels to organismal ecology also have their limitations. With a few important exceptions[117], neoplasms do not contain many species or food webs. There is little diversity of resources in a neoplasm, so there are probably limited opportunities for specialization to different niches, except to the extent that there are different microenvironments in an organ.

Many of the differences between neoplasms and populations of sexual organisms simplify the study of evolution in neoplasms. Experimental evolution studies in bacterial systems have helped elucidate the roles of selection and drift in populations, the development of mutator phenotypes and the dynamics of adaptation[132]. Cancer systems share a similar empirical tractability. Asexual reproduction is easier to analyse than sexual reproduction. More importantly, we have access to the ancestral genotype in the normal tissues of the body, which enables us to study how the neoplasm has changed. Evolution in a neoplasm occurs on a timescale of years, not millennia. Life on Earth has provided us with a single example of how evolution can occur, making it difficult to distinguish regularities from historical accidents. By contrast, every new neoplasm is an example of how neoplastic evolution can proceed, modified by the genotype and exposures of a particular individual. Therefore, we might be able to map out the regularities of the fitness landscape that constrains neoplastic evolution[106,133] (FIG. 4). In fact, efforts to develop models of the order of lesions in neoplastic progression are, in effect, the cartography of neoplastic fitness landscapes.

Conclusions and future directions

To understand cancer, we need to understand and measure the population dynamics and evolutionary parameters of neoplasms. These measurements provide biomarkers that can be used for risk stratification, intermediate endpoints and targets for new drugs. One study in HIV has shown that anti-viral therapy reduced the rate of HIV evolution by two orders of magnitude[134]. Can this be shown in cancer? We need to understand the fitness landscapes of neoplasms to better predict how a particular neoplasm will evolve. We will also need to interfere with clonal evolution — change the fitness landscape and push the population of neoplastic cells down alternative paths to prevent and treat cancer[10]. To understand the evolutionary consequences of our therapeutic strategies, we need to assay the genetics of neoplasms both before and after interventions as part of clinical trials. The development of inexpensive,

Fitness landscape
A multi-dimensional space in which every point represents the genotype or phenotype of a cell and its fitness value. Points are connected if a mutational event can transform one genotype (or phenotype) into the other.

high-throughput, single-cell genomic assays will be important to all these endeavors.

Because neoplastic evolution tends to produce therapeutically resistant clones, one of the most powerful strategies is to prevent the initiation of a neoplasm in the first place, as might be achieved with the use of human papillomavirus vaccines to prevent cervical cancer[135]. If initiation cannot be prevented, the early detection of a neoplasm, before it develops a high degree of genetic heterogeneity, will probably lead to increased cure rates[75].

The presence of clonal competition is an unavoidable fact of cancer biology. No matter how we intervene in a neoplasm, some cells will grow back to fill that space. Whether those cells are normal or neoplastic will depend on the therapy and the mosaic of neoplastic cells that was present before therapy. We have highlighted that the fundamental problems of neoplastic progression and cancer therapy are also problems of evolutionary biology, and so it will be important to integrate evolutionary biologists into our cancer research teams. In addition, evolutionary biology should be a required part of the training of cancer biologists and oncologists. The application of evolutionary biology and ecology to cancer is already helping us to better understand, predict and control this disease.

1. Nowell, P. C. The clonal evolution of tumor cell populations. *Science* **194**, 23–28 (1976).
The seminal description of cancer as an evolutionary process. Predicts sequences of clonal expansions, individual variation in response to interventions and therapeutic resistance.
2. Crespi, B. & Summers, K. Evolutionary Biology of Cancer. *Trends Ecol. Evol.* **20**, 545–552 (2005).
3. Heppner, G. & Miller, F. The cellular basis of tumor progression. *Int. Rev. Cytol.* **177**, 1–56 (1998).
4. Cairns, J. Mutation selection and the natural history of cancer. *Nature* **255**, 197–200 (1975).
Highlights the interaction between tissue architecture and clonal evolution. Also predicts the retention of a non-recombining, 'immortal' strand of DNA in stem cells.
5. Tsao, J. L. *et al.* Genetic reconstruction of individual colorectal tumor histories. *Proc. Natl Acad. Sci. USA* **97**, 1236–1241 (2000).
Uses phylogenetic methods to trace the common ancestor of microsatellite-unstable clones in colorectal cancer back to a date before an adenoma was detected.
6. Tsao, J. L. *et al.* Colorectal adenoma and cancer divergence. Evidence of multilineage progression. *Am. J. Pathol.* **154**, 1815–1824 (1999).
7. Michor, F., Iwasa, Y. & Nowak, M. A. Dynamics of cancer progression. *Nature Rev. Cancer* **4**, 197–205 (2004).
8. Maley, C. C. *et al.* Genetic clonal diversity predicts progression to esophageal adenocarcinoma. *Nature Genet.* **38**, 468–473 (2006).
Shows that genetic diversity measures from ecology and evolution predict progression to malignancy.
9. Maley, C. C. & Reid, B. J. Natural selection in neoplastic progression of Barrett's esophagus. *Semin. Cancer Biol.* **15**, 474–483 (2005).
10. Maley, C. C., Reid, B. J. & Forrest, S. Cancer prevention strategies that address the evolutionary dynamics of neoplastic cells: simulating benign cell boosters and selection for chemosensitivity. *Cancer Epidemiol. Biomarkers Prev.* **13**, 1375–1384 (2004).
Uses computational models to develop prevention and therapeutic strategies for avoiding the evolution of resistance.
11. Tomlinson, I. P. M. Game-theory models of interactions between tumour cells. *Euro. J. Cancer* **33**, 1495–1500 (1997).
12. Gatenby, R. A. & Vincent, T. L. Application of quantitative models from population biology and evolutionary game theory to tumor therapeutic strategies. *Mol. Cancer Ther.* **2**, 919–927 (2003).
Describes the use of evolutionary and ecological models to develop new approaches to therapy.
13. Gonzalez-Garcia, I., Sole, R. V. & Costa, J. Metapopulation dynamics and spatial heterogeneity in cancer. *Proc. Natl Acad. Sci. USA* **99**, 13085–13089 (2002).
Shows genetic heterogeneity within a neoplasm, and how clones can be intertwined in complex patterns.
14. Brash, D. E., Zhang, W., Grossman, D. & Takeuchi, S. Colonization of adjacent stem cell compartments by mutant keratinocytes. *Semin. Cancer Biol.* **15**, 97–102 (2005).
15. Braakhuis, B. J., Leemans, C. R. & Brakenhoff, R. H. Expanding fields of genetically altered cells in head and neck squamous carcinogenesis. *Semin. Cancer Biol.* **15**, 113–120 (2005).
16. Maley, C. C. *et al.* Selectively advantageous mutations and hitchhikers in neoplasms: p16 lesions are selected in Barrett's esophagus. *Cancer Res.* **64**, 3414–3427 (2004).
17. Franklin, W. A. *et al.* Widely dispersed p53 mutation in respiratory epithelium. A novel mechanism for field carcinogenesis. *J. Clin. Invest.* **100**, 2133–2137 (1997).
Shows that neoplastic clones can spread over large surface areas.
18. Castro, M. A., Onsten, T. T., de Almeida, R. M. & Moreira, J. C. Profiling cytogenetic diversity with entropy-based karyotypic analysis. *J. Theor. Biol.* **234**, 487–495 (2005).
19. Keller, L. K. *Levels of Selection in Evolution* (Princeton University Press, Princeton, New Jersey, 1999).
20. Leroi, A. M., Koufopanou, V. & Burt, A. Cancer selection. *Nature Rev. Cancer* **3**, 226–231 (2003).
21. Weinstein, B. S. & Ciszek, D. The reserve-capacity hypothesis: evolutionary origins and modern implications of the trade-off between tumor-suppression and tissue-repair. *Exp. Gerontol.* **37**, 615–627 (2002).
22. Frank, S. A. & Nowak, M. A. Problems of somatic mutation and cancer. *Bioessays* **26**, 291–299 (2004).
23. Campisi, J. Aging, tumor suppression and cancer: high wire-act! *Mech. Ageing Dev.* **126**, 51–58 (2005).
24. Summers, K., da Silva, J. & Farwell, M. Intragenomic conflict and cancer. *Med. Hypotheses* **59**, 170–179 (2002).
25. Roth, M. J. *et al.* Genetic progression and heterogeneity associated with the development of esophageal squamous cell carcinoma. *Cancer Res.* **61**, 4098–4104 (2001).
26. Hanahan, D. & Weinberg, R. A. The hallmarks of cancer. *Cell* **100**, 57–70 (2000).
27. Stoler, D. L. *et al.* The onset and extent of genomic instability in sporadic colorectal tumor progression. *Proc. Natl Acad. Sci. USA* **96**, 15121–15126 (1999).
Measures the frequency of genomic alterations in colorectal cancer at approximately 11,000 per clone.
28. Sole, R. V. & Deisboeck, T. S. An error catastrophe in cancer? *J. Theor. Biol.* **228**, 47–54 (2004).
29. Breivik, J. The evolutionary origin of genetic instability in cancer development. *Semin. Cancer Biol.* **15**, 51–60 (2005).
30. Michor, F., Iwasa, Y., Vogelstein, B., Lengauer, C. & Nowak, M. A. Can chromosomal instability initiate tumorigenesis? *Semin. Cancer Biol.* **15**, 43–49 (2005).
31. Rajagopalan, H., Nowak, M. A., Vogelstein, B. & Lengauer, C. The significance of unstable chromosomes in colorectal cancer. *Nature Rev. Cancer* **3**, 695–701 (2003).
32. Gray, M. W., Burger, G. & Lang, B. F. Mitochondrial evolution. *Science* **283**, 1476–1481 (1999).
33. Baylin, S. B. & Herman, J. G. DNA hypermethylation in tumorigenesis- epigenetics joins genetics. *Trends Genet.* **16**, 168–174 (2000).
34. Feinberg, A. P., Ohlsson, R. & Henikoff, S. The epigenetic progenitor origin of human cancer. *Nature Rev. Genet.* **7**, 21–33 (2006).
35. Weisenberger, D. J. *et al.* CpG island methylator phenotype underlies sporadic microsatellite instability and is tightly associated with BRAF mutation in colorectal cancer. *Nature Genet.* **38**, 787–793 (2006).
36. Horie-Inoue, K. & Inoue, S. Epigenetic and proteolytic inactivation of 14–3-3sigma in breast and prostate cancers. *Semin. Cancer Biol.* **16**, 235–239 (2006).
37. Albertini, R. J., Nicklas, J. A., O'Neill, J. P. & Robison, S. H. *In vivo* somatic mutations in humans: measurement and analysis. *Annu. Rev. Genet.* **24**, 305–326 (1990).
38. Araten, D. J. *et al.* A quantitative measurement of the human somatic mutation rate. *Cancer Res.* **65**, 8111–8117 (2005).
39. Sniegowski, P. D., Gerrish, P. J., Johnson, T. & Shaver, A. The evolution of mutation rates: separating causes from consequences. *Bioessays* **22**, 1057–1066 (2000).
40. Sniegowski, P. D., Gerrish, P. J. & Lenski, R. E. Evolution of high mutation rates in experimental populations of *E. coli*. *Nature* **387**, 703–705 (1997).
41. Dyer, M. A. & Bremner, R. The search for the retinoblastoma cell of origin. *Nature Rev. Cancer* **5**, 91–101 (2005).
42. Renan, M. J. How many mutations are required for tumorigenesis? Implications from human cancer data. *Mol. Carcinogenesis* **7**, 139–146 (1993).
43. Nunney, L. The population genetics of multistage carcinogenesis. *Proc. Biol. Sci.* **270**, 1183–1191 (2003).
44. Loeb, L. A. Mutator phenotype may be required for multistage carcinogenesis. *Cancer Res.* **51**, 3075–3079 (1991).
Shows that the background mutation rate is not adequate to explain carcinogenesis, and proposes that genetic instability might be necessary for cancer to develop.
45. Tomlinson, I. & Bodmer, W. Selection, the mutation rate and cancer: ensuring that the tail does not wag the dog. *Nature Med.* **5**, 11–12 (1999).
46. Moolgavkar, S. H. & Luebeck, E. G. Multistage carcinogenesis and the incidence of human cancer. *Genes Chromosomes Cancer* **38**, 302–306 (2003).
47. Loeb, K. R. & Loeb, L. A. Significance of multiple mutations in cancer. *Carcinogenesis* **21**, 379–385 (2000).
48. Maley, C. C. *et al.* The combination of genetic instability and clonal expansion predicts progression to esophageal adenocarcinoma. *Cancer Res.* **64**, 7629–7633 (2004).
49. Ohta, T. Slightly deleterious mutant substitutions in evolution. *Nature* **246**, 96–98 (1973).
50. Huntly, B. J. & Gilliland, D. G. Leukaemia stem cells and the evolution of cancer-stem-cell research. *Nature Rev. Cancer* **5**, 311–321 (2005).
51. Al-Hajj, M., Wicha, M. S., Benito-Hernandez, A., Morrison, S. J. & Clarke, M. F. Prospective identification of tumorigenic breast cancer cells. *Proc. Natl Acad. Sci. USA* **100**, 3983–3988 (2003).
52. Collins, A. T., Berry, P. A., Hyde, C., Stower, M. J. & Maitland, N. J. Prospective identification of tumorigenic prostate cancer stem cells. *Cancer Res.* **65**, 10946–10951 (2005).
53. Sell, S. Cellular origin of cancer: dedifferentiation or stem cell maturation arrest? *Environ. Health Perspect.* **101**, 15–26 (1993).
54. Potten, C. S., Booth, C. & Pritchard, D. M. The intestinal epithelial stem cell: the mucosal governor. *Int. J. Exp. Pathol.* **78**, 219–243 (1997).
55. Michor, F., Frank, S. A., May, R. M., Iwasa, Y. & Nowak, M. A. Somatic selection for and against cancer. *J. Theor. Biol.* **225**, 377–382 (2003).
56. Frank, S. A. & Nowak, M. A. Cell biology: developmental predisposition to cancer. *Nature* **422**, 494 (2003).
57. Meza, R., Luebeck, E. G. & Moolgavkar, S. H. Gestational mutations and carcinogenesis. *Math. Biosci.* **197**, 188–210 (2005).

58. Maynard Smith, J. & Haigh, J. The hitch-hiking effect of a favorable gene. *Genet. Res.* **231**, 1114–1116 (1974).
59. Lewontin, R. C. *The Genetic Basis of Evolutionary Change* (Columbia University Press, New York, 1970).
60. Braakhuis, B. J., Tabor, M. P., Kummer, J. A., Leemans, C. R. & Brakenhoff, R. H. A genetic explanation of Slaughter's concept of field cancerization: evidence and clinical implications. *Cancer Res.* **63**, 1727–1730 (2003).
61. Chao, E. C. & Lipkin, S. M. Molecular models for the tissue specificity of DNA mismatch repair-deficient carcinogenesis. *Nucleic Acids Res.* **34**, 840–852 (2006).
62. Fearon, E. R. & Vogelstein, B. A genetic model for colorectal tumorigenesis. *Cell* **61**, 759–767 (1990).
63. Desper, R. *et al.* Inferring tree models for oncogenesis from comparative genome hybridization data. *J. Comput. Biol.* 37–51 (1999).
64. Smith, G. *et al.* Mutations in APC, Kirsten-ras, and p53 — alternative genetic pathways to colorectal cancer. *Proc. Natl Acad. Sci. USA* **99**, 9433–9438 (2002).
65. Kobayashi, S. *et al.* EGFR mutation and resistance of non-small-cell lung cancer to gefitinib. *N. Engl. J. Med.* **352**, 786–792 (2005).
66. Gorre, M. E. *et al.* Clinical resistance to STI-571 cancer therapy caused by *BCR-ABL* gene mutation or amplification. *Science* **293**, 876–880 (2001).
67. Wang, T. L. *et al.* Digital karyotyping identifies thymidylate synthase amplification as a mechanism of resistance to 5-fluorouracil in metastatic colorectal cancer patients. *Proc. Natl Acad. Sci. USA* **101**, 3089–3094 (2004).
68. Donnenberg, V. S. & Donnenberg, A. D. Multiple drug resistance in cancer revisited: the cancer stem cell hypothesis. *J. Clin. Pharmacol.* **45**, 872–877 (2005).
69. Michor, F. *et al.* Dynamics of chronic myeloid leukaemia. *Nature* **435**, 1267–1270 (2005).
70. Luria, S. E. & Delbruck, M. Mutations of bacteria from virus sensitivity to virus resistance. *Genetics* **28**, 491–511 (1943).
71. Komarova, N. Stochastic modeling of drug resistance in cancer. *J. Theor. Biol.* **239**, 351–366 (2006).
72. Roche-Lestienne, C. & Preudhomme, C. Mutations in the ABL kinase domain pre-exist the onset of imatinib treatment. *Semin. Hematol.* **40**, 80–82 (2003).
73. Iwasa, Y., Nowak, M. A. & Michor, F. Evolution of resistance during clonal expansion. *Genetics* **172**, 2557–2566 (2006).
74. Knudson, A. G. Jr. Mutation and cancer: statistical study of retinoblastoma. *Proc. Natl Acad. Sci. USA* **68**, 820–823 (1971).
Uses a Poisson model to deduce the requirement of two mutations to generate retinoblastoma.
75. Etzioni, R. *et al.* The case for early detection. *Nature Rev. Cancer* **3**, 1–10 (2003).
76. Chabner, B. A. & Roberts, T. G. Jr. Timeline: chemotherapy and the war on cancer. *Nature Rev. Cancer* **5**, 65–72 (2005).
77. Komarova, N. L. & Wodarz, D. Evolutionary dynamics of mutator phenotypes in cancer: implications for chemotherapy. *Cancer Res.* **63**, 6635–6642 (2003).
78. Suiter, A. M., Banziger, O. & Dean, A. M. Fitness consequences of a regulatory polymorphism in a seasonal environment. *Proc. Natl Acad. Sci. USA* **100**, 12782–12786 (2003).
79. Kim, J. J. & Tannock, I. F. Repopulation of cancer cells during therapy: an important cause of treatment failure. *Nature Rev. Cancer* **5**, 516–525 (2005).
80. Kern, W. & Estey, E. H. High-dose cytosine arabinoside in the treatment of acute myeloid leukemia: review of three randomized trials. *Cancer* **107**, 116–124 (2006).
81. Dolken, G. Detection of minimal residual disease. *Adv. Cancer Res.* **82**, 133–185 (2001).
82. Rubin, C. E. *et al.* DNA aneuploidy in colonic biopsies predicts future development of dysplasia in ulcerative colitis. *Gastroenterology* **103**, 1611–1620 (1992).
83. Brentnall, T. A. *et al.* Mutations in the p53 gene: an early marker of neoplastic progression in ulcerative colitis. *Gastroenterology* **107**, 369–378 (1994).
84. van Oijen, M. G. & Slootweg, P. J. Oral field cancerization: carcinogen-induced independent events or micrometastatic deposits? *Cancer Epidemiol. Biomarkers Prev.* **9**, 249–256 (2000).
85. Hunter, K. W. Allelic diversity in the host genetic background may be an important determinant in tumor metastatic dissemination. *Cancer Lett.* **200**, 97–105 (2003).
86. Bernards, R. & Weinberg, R. A. A progression puzzle. *Nature* **418**, 823 (2002).
87. Bernards, R. & Weinberg, R. A. Bernards and Weinberg reply. *Nature* **419**, 560 (2002).
88. Fidler, I. J. & Kripke, M. L. Metastasis results from preexisting variant cells within a malignant tumor. *Science* **197**, 893–895 (1977).
89. Kirkwood, T. B. Evolution of ageing. *Mech. Ageing Dev.* **123**, 737–745 (2002).
90. Futuyma, D. J. *Evolutionary Biology* (Sinauer Associates Inc., Sunderland, Massachusetts, 1998).
91. Paszek, M. J. *et al.* Tensional homeostasis and the malignant phenotype. *Cancer Cell* **8**, 241–254 (2005).
92. Vaupel, P. & Mayer, A. Hypoxia and anemia: effects on tumor biology and treatment resistance. *Transfus. Clin. Biol.* **12**, 5–10 (2005).
93. Cadet, C., Ferriere, R., Metz, J. A. & van Baalen, M. The evolution of dispersal under demographic stochasticity. *Am. Nat.* **162**, 427–441 (2003).
94. Fidler, I. J. The pathogenesis of cancer metastasis: the 'seed and soil' hypothesis revisited. *Nature Rev. Cancer* **3**, 453–458 (2003).
95. Kolar, C. S. & Lodge, D. M. Progress in invasion biology: predicting invaders. *Trends Ecol. Evol.* **16**, 199–204 (2001).
96. Peterson, A. T. Predicting the geography of species' invasions via ecological niche modeling. *Q. Rev. Biol.* **78**, 419–433 (2003).
97. Shea, K. & Chesson, P. Community ecology theory as a framework for biological invasions. *Trends Ecol. Evol.* **17**, 170–176 (2002).
98. Mable, B. K. Breaking down taxonomic barriers in polyploidy research. *Trends Plant Sci.* **8**, 582–590 (2003).
99. Otto, S. P. & Whitton, J. Polyploid incidence and evolution. *Annu. Rev. Genet.* **34**, 401–437 (2000).
100. Imhof, M. & Schlotterer, C. E. coli microcosms indicate a tight link between predictability of ecosystem dynamics and diversity. *PLoS Genet.* **2**, e103 (2006).
101. Heppner, G., Miller, B., Cooper, D. N. & Miller, F. R. in *Cell Biology of Breast Cancer* (eds McGrath, C. M., Brennan, M. J. & Rich, M. A.) 161–172 (Academic Press, New York, 1980).
102. Guba, M. *et al.* A primary tumor promotes dormancy of solitary tumor cells before inhibiting angiogenesis. *Cancer Res.* **61**, 5575–5579 (2001).
103. Miller, B. E., Miller, F. R., Leith, J. & Heppner, G. H. Growth interaction *in vivo* between tumor subpopulations derived from a single mouse mammary tumor. *Cancer Res.* **40**, 3977–3981 (1980).
Shows that different clones in a neoplasm can affect each other's growth in complex ways.
104. Caignard, A., Martin, M. S., Michel, M. F. & Martin, F. Interaction between two cellular subpopulations of a rat colonic carcinoma when inoculated to the syngeneic host. *Int. J. Cancer* **36**, 273–279 (1985).
105. Bach, L. A., Bentzen, S. M., Alsner, J. & Christiansen, F. B. An evolutionary-game model of tumour-cell interactions: possible relevance to gene therapy. *Eur. J. Cancer* **37**, 2116–2120 (2001).
106. Gatenby, R. A. & Vincent, T. L. An evolutionary model of carcinogenesis. *Cancer Res.* **63**, 6212–6220 (2003).
107. Nagy, J. D. Competition and natural selection in a mathematical model of cancer. *Bull. Math. Biol.* **66**, 663–687 (2004).
108. Seliger, B. Strategies of tumor immune evasion. *BioDrugs* **19**, 347–354 (2005).
109. Uhr, J. W., Scheuermann, R. H., Street, N. E. & Vitetta, E. S. Cancer dormancy: opportunities for new therapeutic approaches. *Nature Med.* **3**, 505–509 (1997).
110. Mitteldorf, J. Chaotic population dynamics and the evolution of aging. *Evol. Ecol. Res.* **8**, 561–574 (2006).
111. Rundhaug, J. E. Matrix metalloproteinases and angiogenesis. *J. Cell. Mol. Med.* **9**, 267–285 (2005).
112. Travisano, M. & Velicer, G. J. Strategies of microbial cheater control. *Trends Microbiol.* **12**, 72–78 (2004).
113. Turner, P. E. Parasitism between co-infecting bacteriophages. *Adv. Ecol. Res.* **37**, 309–332 (2005).
114. Velicer, G. J., Kroos, L. & Lenski, R. E. Developmental cheating in the social bacterium *Myxococcus xanthus*. *Nature* **404**, 598–601 (2000).
115. Jouanneau, J., Moens, G., Bourgeois, Y., Poupon, M. F. & Thiery, J. P. A minority of carcinoma cells producing acidic fibroblast growth factor induces a community effect for tumor progression. *Proc. Natl Acad. Sci. USA* **91**, 286–290 (1994).
116. Axelrod, R., Axelrod, D. E. & Pienta, K. J. Evolution of cooperation among tumor cells. *Proc. Natl Acad. Sci. USA* **103**, 13474–13479 (2006).
117. Mueller, M. M. & Fusenig, N. E. Friends or foes-bipolar effects of the tumour stroma in cancer. *Nature Rev. Cancer* **4**, 839–849 (2004).
118. Shao, Z. M., Nguyen, M. & Barsky, S. H. Human breast carcinoma desmoplasia is PDGF initiated. *Oncogene* **19**, 4337–4345 (2000).
119. Tlsty, T. D. Stromal cells can contribute oncogenic signals. *Semin. Cancer Biol.* **11**, 97–104 (2001).
120. Fukino, K. *et al.* Combined total genome loss of heterozygosity scan of breast cancer stroma and epithelium reveals multiplicity of stromal targets. *Cancer Res.* **64**, 7231–7236 (2004).
Shows that tumour stroma also has genetic lesions, and so must be co-evolving with the epithelium.
121. Paterson, R. F. *et al.* Molecular genetic alterations in the laser-capture-microdissected stroma adjacent to bladder carcinoma. *Cancer* **98**, 1830–1836 (2003).
122. Ishiguro, K., Yoshida, T., Yagishita, H., Numata, Y. & Okayasu, T. Epithelial and stromal genetic instability contributes to genesis of colorectal adenomas. *Gut* **55**, 695–702 (2006).
123. Kenny, P. A. & Bissell, M. J. Tumor reversion: correction of malignant behavior by microenvironmental cues. *Int. J. Cancer* **107**, 688–695 (2003).
124. Mintz, B. & Illmensee, K. Normal genetically mosaic mice produced from malignant teratocarcinoma cells. *Proc. Natl Acad. Sci. USA* **72**, 3585–3589 (1975).
125. Miller, F. R. & Heppner, G. H. Cellular interactions in metastasis. *Cancer Metastasis Rev.* **9**, 21–34 (1990).
126. Kuperwasser, C. *et al.* Reconstruction of functionally normal and malignant human breast tissues in mice. *Proc. Natl Acad. Sci. USA* **101**, 4966–4971 (2004).
127. Bhowmick, N. A. *et al.* TGF-β signaling in fibroblasts modulates the oncogenic potential of adjacent epithelia. *Science* **303**, 848–851 (2004).
128. Orimo, A. *et al.* Stromal fibroblasts present in invasive human breast carcinomas promote tumor growth and angiogenesis through elevated SDF-1/CXCL12 secretion. *Cell* **121**, 335–348 (2005).
129. Roxburgh, S. H., Shea, K. & Wilson, J. B. The intermediate disturbance hypothesis: patch dynamics and mechanisms of species coexistence. *Ecology* **85**, 359–371 (2004).
130. Bjerkvig, R., Tysnes, B. B., Aboody, K. S., Najbauer, J. & Terzis, A. J. Opinion: the origin of the cancer stem cell: current controversies and new insights. *Nature Rev. Cancer* **5**, 899–904 (2005).
131. Boyer, B., Valles, A. M. & Edme, N. Induction and regulation of epithelial-mesenchymal transitions. *Biochem. Pharmacol.* **60**, 1091–1099 (2000).
132. Elena, S. F. & Lenski, R. E. Evolution experiments with microorganisms: the dynamics and genetic bases of adaptation. *Nature Rev. Genet.* **4**, 457–469 (2003).
133. Spencer, S. L., Gerety, R. A., Pienta, K. J. & Forrest, S. Modeling somatic evolution in tumorigenesis. *PLoS Comput. Biol.* **2**, e108 (2006).
134. Drummond, A., Forsberg, R. & Rodrigo, A. G. The inference of stepwise changes in substitution rates using serial sequence samples. *Mol. Biol. Evol.* **18**, 1365–1371 (2001).
135. Harper, D. M. *et al.* Sustained efficacy up to 4.5 years of a bivalent L1 virus-like particle vaccine against human papillomavirus types 16 and 18: follow-up from a randomised control trial. *Lancet* **367**, 1247–1255 (2006).
136. Jakobisiak, M., Lasek, W. & Golab, J. Natural mechanisms protecting against cancer. *Immunol. Lett.* **90**, 103–122 (2003).
137. Sherr, C. J. Tumor surveillance via the ARF-p53 pathway. *Genes Dev.* **12**, 2984–2991 (1998).
138. Michor, F., Nowak, M. A., Frank, S. A. & Iwasa, Y. Stochastic elimination of cancer cells. *Proc. Biol. Sci.* **270**, 2017–2024 (2003).
139. Torres-Montaner, A. & Hughes, D. A hypothetical anti-neoplastic mechanism associated to reserve cells. *J. Theor. Biol.* **231**, 239–248 (2004).
140. Frank, S. A., Iwasa, Y. & Nowak, M. A. Patterns of cell division and the risk of cancer. *Genetics* **163**, 1527–1532 (2003).
141. Komarova, N. L. & Cheng, P. Epithelial tissue architecture protects against cancer. *Math. Biosci.* **200**, 90–117 (2006).
142. Nowak, M. A., Michor, F. & Iwasa, Y. The linear process of somatic evolution. *Proc. Natl Acad. Sci. USA* **100**, 14966–14969 (2003).

Uses a mathematical model to analyse the evolutionary dynamics of cells in structured tissues.
143. Frank, S. A. Genetic predisposition to cancer — insights from population genetics. *Nature Rev. Genet.* **5**, 764–772 (2004).
144. Fleming, M. A., Potter, J. D., Ramirez, C. J., Ostrander, G. K. & Ostrander, E. A. Understanding missense mutations in the *BRCA1* gene: an evolutionary approach. *Proc. Natl Acad. Sci. USA* **100**, 1151–1156 (2003).
145. Slatkin, M. & Rannala, B. Estimating allele age. *Annu. Rev. Genomics Hum. Genet.* **1**, 225–249 (2000).
146. Deng, C. X. BRCA1: cell cycle checkpoint, genetic instability, DNA damage response and cancer evolution. *Nucleic Acids Res.* **34**, 1416–1426 (2006).
147. Jernstrom, H., Johannsson, O., Borg, A., Ivarsson, H. & Olsson, H. BRCA1-positive patients are small for gestational age compared with their unaffected relatives. *Eur. J. Cancer* **34**, 368–371 (1998).
148. Smith, T. M. *et al.* Complete genomic sequence and analysis of 117 kb of human DNA containing the gene *BRCA1*. *Genome Res.* **6**, 1029–1049 (1996).
149. Pavlicek, A. *et al.* Evolution of the tumor suppressor BRCA1 locus in primates: implications for cancer predisposition. *Hum. Mol. Genet.* **13**, 2737–2751 (2004).
150. Summers, K. & Crespi, B. Cadherins in maternal–foetal interactions: red queen with a green beard? *Proc. Biol. Sci.* **272**, 643–649 (2005).
151. Nielsen, R. *et al.* A scan for positively selected genes in the genomes of humans and chimpanzees. *PLoS Biol.* **3**, e170 (2005).
152. Crespi, B. J. & Summers, K. Positive selection in the evolution of cancer. *Biol. Rev. Camb. Philos. Soc.* **81**, 407–424 (2006).
153. Hernandez, L., Kozlov, S., Piras, G. & Stewart, C. L. Paternal and maternal genomes confer opposite effects on proliferation, cell-cycle length, senescence, and tumor formation. *Proc. Natl Acad. Sci. USA* **100**, 13344–13349 (2003).
154. Moran, P. A. P. Random processes in genetics. *Proc. Camb. Phil. Soc.* **54**, 60–71 (1958).
155. Ewens, W. J. *Mathematical Population Genetics* (Springer-Verlag, New York, 2004).
156. Reid, B. J. *et al.* Barrett's esophagus: ordering the events that lead to cancer. *Euro. J. Cancer Prev.* **5** (Suppl. 2), 57–65 (1996).
Shows how spatial information can be used to infer dependencies between genetic alterations in carcinogenesis.
157. Wang, G. Q. *et al.* Histological precursors of oesophageal squamous cell carcinoma: results from a 13 year prospective follow up study in a high risk population. *Gut* **54**, 187–192 (2005).
158. Reid, B. J. *et al.* Predictors of progression in Barrett's esophagus II: baseline 17p (p53) loss of heterozygosity identifies a patient subset at increased risk for neoplastic progression. *Am. J. Gastroenterol.* **96**, 2839–2848 (2001).
159. Dolan, K., Morris, A. I., Gosney, J. R., Field, J. K. & Sutton, R. Loss of heterozygosity on chromosome 17p predicts neoplastic progression in Barrett's esophagus. *J. Gastroenterol. Hepatol.* **18**, 683–689 (2003).
160. Teodori, L. *et al.* DNA/protein flow cytometry as a predictive marker of malignancy in dysplasia-free Barrett's esophagus: thirteen-year follow-up study on a cohort of patients. *Cytometry* **34**, 257–263 (1998).
161. Lee, J. J. *et al.* Predicting cancer development in oral leukoplakia: ten years of translational research. *Clin. Cancer Res.* **6**, 1702–1710 (2000).
162. Rosin, M. P. *et al.* Use of allelic loss to predict malignant risk for low-grade oral epithelial dysplasia. *Clin. Cancer Res.* **6**, 357–362 (2000).
163. Befrits, R., Hammarberg, C., Rubio, C., Jaramillo, E. & Tribukait, B. DNA aneuploidy and histologic dysplasia in long-standing ulcerative colitis. A 10-year follow-up study. *Dis. Colon. Rectum* **37**, 313–319; discussion 319–320 (1994).
164. Lofberg, R., Brostrom, O., Karlen, P., Tribukait, B. & Ost, A. Colonoscopic surveillance in long-standing total ulcerative colitis-a 15-year follow-up study. *Gastroenterology* **4**, 1021–1021 (1990).
165. Wong, B. C. *et al.* Helicobacter pylori eradication to prevent gastric cancer in a high-risk region of China: a randomized controlled trial. *JAMA* **291**, 187–194 (2004).
166. Thompson, I. M. *et al.* The influence of finasteride on the development of prostate cancer. *N. Engl. J. Med.* **349**, 215–224 (2003).
167. Abrams, P. A. On classifying interactions between populations. *Oecologia* **73**, 272–281 (1987).
168. Heppner, G. H., Miller, B. E. & Miller, F. R. Tumor subpopulation interactions in neoplasms. *Biochim. Biophys. Acta* **695**, 215–226 (1983).

Acknowledgements
This work was supported by the US National Institutes of Health, the Commonwealth of Pennsylvania, and the Pew Charitable Trust, and initiated by the Santa Fe Institute. We thank W. Ewens, M. Carroll and J. Radich for helpful comments. We apologize to our peers whose work we were unable to cite owing to space limitations.

Competing interests statement
The authors declare no competing financial interests.

DATABASES
The following terms in this article are linked online to:
Entrez Gene:
http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?db=gene
ABL | BCR | *CDKN2A* | EGFR | HGF | *MLH1* | *MLH3* | *MSH6* | SDF1 | *SFN* | TGFβ1 | *TP53* |
National Cancer Institute:
http://www.cancer.gov
chronic myeloid leukaemia | colorectal cancer

FURTHER INFORMATION
Carlo C. Maley's homepage: http://www.wistar.org/research_facilites/maley/research.htm
Access to this interactive links box is free online.