Current Biology, Vol. 13, R261–R263, April 1, 2003, ©2003 Elsevier Science Ltd. All rights reserved. DOI 10.1016/S0960-9822(03)00195-7

Somatic Mutation: Early Cancer Steps Depend on Tissue Architecture

Dispatch

Steven A. Frank

Cells progress from a normal to a cancerous state by accumulating mutations. Recent mathematical studies show how the size of the cellular population in a local patch of tissue influences the spread of cancer-promoting mutations.

Cancer results from mutations that alter the normal regulation of cellular birth and death. Thus an important aspect of cancer progression concerns the way in which mutations accumulate in cellular lineages, an issue discussed in an influential paper back in 1975 by Cairns [1]. He noted that tissues such as the skin and colon are divided into many small compartments, such that there is little mixing of cellular populations between adjacent compartments. Stem cells at the base of each compartment renew the tissue, with each stem cell division producing one stem cell and one transit cell. The stem cell remains at the base of the compartment and continues to renew the tissue. The transit cell divides a limited number of times, producing cells that move up from the basal layer and eventually slough off from the surface.

Mutations that occur in the stem lineage remain in the compartment and increase the risk of future cancer. By contrast, mutations in the transit lineage get 'washed out' as they move up the compartment and slough off at the surface. Thus, the tissue architecture clears transit lineage mutations but retains stem lineage mutations. Compartmentalizing tissue also provides defense against aggressive cell lines by physically partitioning the tissue into small units. In order to be successful, a cancer line must obtain mutations that allow invasion of neighboring compartments and promote the spread of the cancer.

Cairns' paper has been widely cited over the past 28 years. But few have expanded these fundamental insights about how tissue architecture and the pattern of cellular lineages affect the rate at which cancerous mutations can accumulate. These quantitative issues call for mathematical analysis [2]. Recently, Martin Nowak and his colleagues have initiated a series of mathematical studies on the accumulation of somatic mutations and progression to cancer [3–5]. A new paper from this group [6] published in this issue of *Current Biology* considers how the size of the cellular population in a compartment affects the success of different kinds of mutations.

Cancer-promoting mutations often fall into one of two classes [7,8] (Figure 1). Gatekeeper mutations abrogate checks and balances on cell division and cell death, often leading to increased rates of cellular proliferation. Caretaker mutations enhance genetic instability by increasing the rate of mutation or chromosomal rearrangement. Michor *et al.* [6] note that gatekeeper mutations have a selective advantage because the cells carrying these mutations replicate faster than their neighbors. By contrast, caretaker mutations are probably disadvantageous when they first arise because most mutations generated by genetic instability will decrease the success of the cell. Genetic instability may eventually lead to a breakdown in gatekeeper functions, but the issue here concerns the success of the initial caretaker mutations.

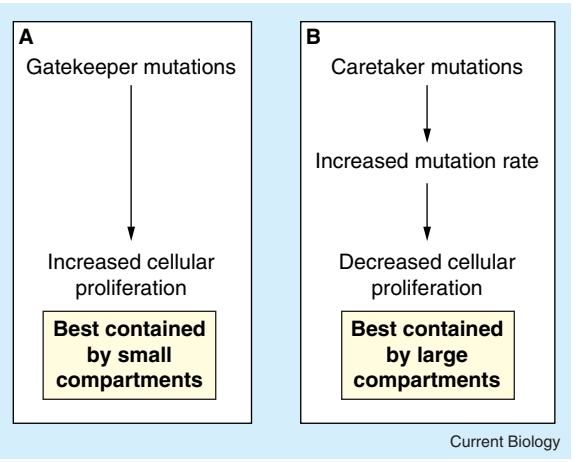

Figure 1. Two major classes of mutation in cancer progression.

(A) Gatekeeper mutations increase the rate of cellular proliferation. This may occur following mutations of oncogenes that directly accelerate progress through the cell cycle. For example, Myc is an oncogenic transcription factor that influences the gene expression of many other genes [11]. A recent study suggests that Myc-activated genes gear cell physiology to the rapid utilization of carbon sources and the expansion of cellular mass [12]. Tumor suppressor genes are also gatekeepers that increase proliferation by abrogating blocks to progression through the cell cycle. The retinoblastoma gene encodes a tumor suppressor that acts as a transcription factor to control many genes involved in cell proliferation [13]. The retinoblastoma transcription factor can, for example, silence target genes of its binding partner E2F that play a key role in the DNA synthesis (S) phase of the cell cycle. Michor *et al.*'s study [6] shows that small cellular compartments best control gatekeeper mutations. (B) Caretaker mutations cause genetic instability, increasing the rate of cellular mutations. p53 is a caretaker and also the most commonly mutated gene in human cancers [14]. p53 appears to be a transcription factor that regulates a variety of other genes involved in cellular growth [15,16]. Among its functions, p53 blocks progression through the cell cycle in response to DNA damage. Loss of p53 therefore increases the accumulation of mutations. It could be that bypassing DNA repair both increases mutation and increases cellular proliferation, but Michor *et al.* [6] reasonably assume that genetic instability usually causes enough damage to reduce the average rate of cellular proliferation. This group goes on to show that large cellular compartments best control caretaker mutations.

Department of Ecology and Evolutionary Biology, University of California, Irvine, CA 92697-2525, USA.
E-mail: safrank@uci.edu

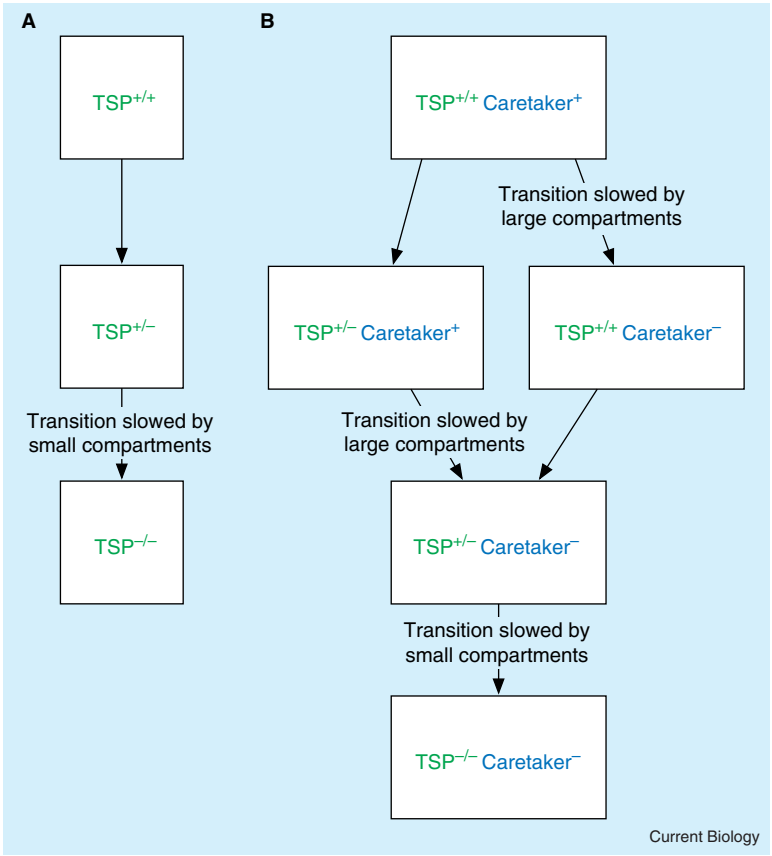

Figure 2. Two pathways to initiate cancer progression.

(A) The first tumor suppressor mutation is recessive and has no phenotypic effect. The second mutation increases the rate of cellular proliferation. Rapid cell proliferation is best controlled by small compartments, because chance events in small compartments may cause the disappearance of the second mutation before it can spread. (B) Two independent events begin the process and may occur in either order. On the left, the first tumor suppressor mutation arises, followed by a mutation to a caretaker gene. On the right, a caretaker mutation occurs, followed by the first tumor suppressor mutation. In either case, a caretaker mutation causes greater genetic instability. Genetic instability reduces cellular success, thus large compartments provide the best control, because they reduce chance events that can allow a caretaker mutation to spread in spite of its disadvantage in growth. Once both genetic instability and the heterozygous TSP gene become established, chromosomal aberrations and loss of heterozygosity cause relatively rapid progression to an inactivated TSP locus. An inactivated TSP gene increases cellular proliferation — thus, its spread is best controlled by a small compartment. The opposing pressures on compartment size in the presence of genetic instability suggest that intermediate compartment size is best at controlling this route to cancer progression.

The contrast between advantageous gatekeeper and deleterious caretaker mutations leads to the first question. How does the size of a local cell population influence the probability that gatekeeper and caretaker mutations can spread and thus begin the progression toward cancer? The main insight arises simply from asking the question in this way, as this allows the powerful theory of population genetics to be brought to bear. In a large population, caretaker mutations with increased rates of proliferation almost always succeed, whereas gatekeeper mutations with decreased rates of proliferation almost never succeed. Put another way, natural selection among cell lineages deterministically takes its course in a large population. In small populations, chance events can influence which cell lineages succeed or fail. Thus, small populations increase the probability that deleterious caretaker mutations spread and decrease the probability that advantageous (to the cell) gatekeeper mutations spread.

In terms of cancer risk, large populations lead to cancer progression via initial gatekeeper mutations and rapid cellular proliferation. By contrast, small populations may often begin cancer progression with caretaker mutations and genetic instability. This leads to the second question addressed by Michor *et al.* — what population size minimizes the risk of cancer? This group developed a sophisticated mathematical theory to study rates of cancer progression in different cellular population sizes. They considered the path to cancer via knockout of a tumor suppressor (TSP) gene, a type of gatekeeper that when inactivated causes the cell to proliferate more rapidly [9]. TSP knockout can happen by two independent mutations to the locus, at the usual mutation rate for normal cells (Figure 2A). Alternatively, the cell can acquire one standard mutation to TSP and a mutation to a caretaker gene that raises the rate of chromosomal abnormalities (Figure 2B). A common feature of chromosomal instability is loss of one chromosome of a pair followed by duplication of the remaining chromosome. This loss-duplication can cause a TSP locus that is heterozygous for a knockout mutation to become homozygous for the knockout [10]. Cells with chromosomal instability undergo loss of heterozygosity at a relatively high rate compared with normal somatic mutations. So the rate-limiting pathway for knockout of TSP would be one mutation causing chromosomal instability and one mutation causing heterozygosity to TSP, followed by rapid loss of heterozygosity [3].

I have mentioned many parameters: mutation rates to TSP and caretaker genes, rate of loss of heterozygosity, the growth advantage for TSP knockout cells, the growth disadvantage for cells with caretaker mutations, and the size of the local population of cells. From these parameters one needs to analyze the rates at which new mutations of different kinds occur and become fixed in the local population of cells. Remarkably, Michor *et al.* [6] were able to solve these complicated rate problems with explicit equations. The solutions for the rate equations allowed this group to show how cellular population size affects cancer progression. Small populations are

best for controlling knockouts of TSP but are susceptible to the spread of genetic instability mutations. Thus, for most parameters, intermediate population sizes are favored to balance stochastic increase of deleterious caretaker mutations and deterministic increase of advantageous gatekeeper mutations. At the intermediate population size that best controls cancer, Michor *et al.* [6] calculated the relative contribution of the two pathways to cancer. Recall that one pathway follows two mutations to TSP at the normal somatic rate. The other pathway follows one mutation to cause genetic instability and one mutation to TSP, followed by rapid loss of the second TSP allele via chromosomal loss of heterozygosity. At the optimum population size, a substantial fraction of cancer risk comes via the pathway of early genetic instability followed by loss of heterozygosity. This supports other mathematical work by the same group showing that genetic instability can be an important early step in cancer progression [3–5].

Continuously dividing epithelial tissues such as the skin and colon are separated into many small compartments [1]. Each compartment forms an isolated local population in which the dynamics of early cancer progression play out independently. According to Michor *et al.* [6], small epithelial compartments protect against the deterministic spread of gatekeeper knockouts but raise the risk of starting cancer progression from caretaker mutations and genetic instability. This newly developing quantitative theory may also help to explain why different tissues progress toward cancer in different ways [8]. For example, different tissues will no doubt vary in the size of their cellular compartments. Michor *et al.*'s analysis predicts that the small-compartment epithelial tissues more often begin cancer progression via genetic instability than do tissues with larger compartments.

References

1. Cairns, J. (1975). Mutation selection and the natural history of cancer. Nature *255*, 197–200.
2. Gatenby, R.A. and Maini, P.K. (2003). Mathematical oncology: cancer summed up. Nature *421*, 321.
3. Nowak, M.A., Komarova, N.L., Sengupta, A., Jallepalli, P.V., Shih, I.M., Vogelstein, B., and Lengauer, C. (2002). The role of chromosomal instability in tumor initiation. Proc. Natl. Acad. Sci. U.S.A. *99*, 16226–16231.
4. Komarova, N.L., Lengauer, C., Vogelstein, B. and Nowak, M.A. (2003). Dynamics of genetic instability in colorectal cancer. Cancer Biology and Therapy, in press.
5. Komarova, N.L., Sengupta, A. and Nowak, M.A. (2003). Mutation-selection networks of cancer initiation: tumor suppressor genes and chromosomal instability. J. Theor. Biol., in press.
6. Michor, F., Iwasa, Y., Komarova, N.L. and Nowak, M.A. (2003). Local regulation of homeostasis favors chromosomal instability. Curr. Biol. *13*, this issue.
7. Kinzler, K.W. and Vogelstein, B. (1997). Gatekeepers and caretakers. Nature *386*, 761–763.
8. Vogelstein, B. and Kinzler, K.W. (2002). The Genetic Basis of Human Cancer, 2nd ed. (McGraw-Hill, New York).
9. Knudson, A.G. (1993). Antioncogenes and human cancer. Proc. Natl. Acad. Sci. U.S.A. *90*, 10914–10921.
10. Lengauer, C., Kinzler, K.W. and Vogelstein, B. (1998). Genetic instabilities in human cancers. Nature *396*, 643–649.
11. Schuldiner, O., Shor, S. and Benvenisty, N. (2002). A computerized database-scan to identify c-MYC targets. Gene *292*, 91–99.
12. O'Connell, B.C., Cheung, A.F., Simkevich, C.P., Tam, W., Ren, X. Mateya, M.K. and Sedivy, J.M. (2003). A large scale genetic analysis of c-Myc-regulated gene expression patterns. J. Biol. Chem., in press.
13. Newsham, I.F., Hadjistilianou, T. and Cavenee, W.K. (2002). Retinoblastoma. In The Genetic Basis of Human Cancer, B. Vogelstein and K.W. Kinzler, eds. 2nd ed. (McGraw-Hill, New York), pp. 357–386.
14. Malkin, D. (2002). Li-Fraumeni syndrome. In The Genetic Basis of Human Cancer, B. Vogelstein and K.W. Kinzler, eds. 2nd ed. (McGraw-Hill, New York), pp. 387–401.
15. Levine, A.J., Momand, J. and Finlay, C.A. (1991). The p53 tumor suppressor gene. Nature *351*, 453–456.
16. Vogelstein, B. and Kinzler, K.W. (1992). p53 function and dysfunction. Cell *70*, 523–526.