Supporting Text

Clonal Expansion of Hepatocyte Variants in Chronic Hepatitis. This analysis examines the quantitative relationship between the expansion of variant hepatocyte clones during liver turnover, based on a selective advantage of the variant. Two types of selective advantage are considered together: resistance to killing and preferential cell regeneration.

Assume a liver consisting of two cell types, designated $c0$ and $c1$, where $c0$ cells are normal hepatocytes, and $c1$ cells are variants. $c0$ = 1 liver equivalent, and $c1 \ll c0$ at all times. First-order kill rate constants for the two cell types are $d0$ and $d1$. Normal hepatocytes, $c0$, because they are in a vast majority, can be considered to regenerate at the same absolute rate as they are killed, and, with a first-order growth rate constant $k0 = d0$. Variant hepatocytes, c1, may die and regenerate with different probabilities from that of $c0$, so that $k1$ and $d1$ might not be equal to $k0$ or $d0$. Because $c0(t) >> c1(t)$, the extra mass added to the liver by the addition or depletion of $c1$ variants is neglected.

$$\frac{dc0(t)}{dt} = c0(t)(k0 - d0) = 0 \text{, and} \qquad \frac{dc1(t)}{dt} = c1(t) \cdot (k1 - d1).$$

If $a = \frac{k1}{k0}$ and $b = \frac{d1}{d0}$, where $a, b \geq 0$. The expansion of the variant is as follows:

$$\frac{c1(t)}{c0(t)} = e^{(a \cdot k0 - b \cdot d0) \cdot t} \text{, and} \qquad \ln\left(\frac{c1(t)}{c1(0)}\right) = (a \cdot k0 - b \cdot d0) \cdot t \text{, or since } k0 = d0,$$

$$\ln\left(\frac{c1(t)}{c1(0)}\right) = (a - b) \cdot d0 \cdot t \; .$$

Because $d0 \cdot t$ is the total hepatocyte killing, or the total turnover, $T(t)$, the relationship between clonal expansion of c1 and turnover is as follows:

$$\ln\left(\frac{c1(t)}{c1(0)}\right) = (a - b) \cdot T(t)$$

Fig. 5, which is published as supporting information on the PNAS web site, illustrates this relationship for five values of $a - b$, where $a - b = 0$ represents no selective advantage for the variant.

Clonal Outgrowth due to Random Turnover. We previously published an analysis of the relationship between random turnover of a population of cells and the clonal distribution of cells in that population (1). Briefly, the distribution $F(n,k)$ of clones, where n is the number of members in a clone and k is the number of cycles of random cell killing followed by random cell division, is given by recursive computation of the two equations:

$$f(n,k) = F(n,k-1)/c - n \cdot F(n,k-1)/c + (n+1) \cdot F(n+1,k-1)/c$$

$$F(n,k) = f(n,k) - n \cdot f(n,k)/(c-1) + (n-1) \cdot f(n-1,k)/(c-1)$$

where $F(n=1,0) = c$, $F(n>1,0) = 0$, $f(0,k) = 0$, and $n = 0 \rightarrow c$. To calculate how many cycles of killing would be required to expand one cell out of 5×10^5 cells to a size of 1,000 or more cells, the equations were computed until

$$\sum_{n=1{,}000}^{n=c} F(n,k) = \frac{c}{5 \cdot 10^5} ,$$

where c = 10,000. The expression represents the number of clones of size >1000 cells when c cells undergo k cycles of killing and regeneration. When c = 10,000 the number of clones is 0.02. On average, one clone will expand to a size of 1,000 cells or more when 10,000/c, or 5×10^5 cells, undergo k/c turnovers. The number of turnovers that satisfied this condition was 123.54.

1. Summers, J., Jilbert, A. R., Yang, W., Aldrich, C. E., Saputelli, J., Litwin, S., Toll, E. & Mason, W. S. (2003) *Proc. Natl. Acad. Sci. USA* **100,** 11652−11659.