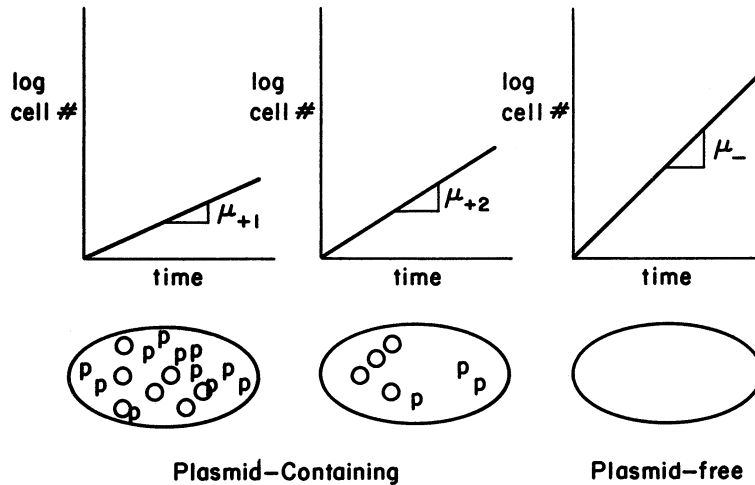


## Genetic Instability



**Figure 14.3.** Cells that contain plasmids (O) actively making protein (p) must direct many of their cellular resources away from growth and hence grow more slowly than plasmid-free cells (that is,  $\mu_{+1} < \mu_{+2} < \mu_-$ ).

plasmid-containing unaltered cells. Genetic instability can occur in any expression system. We illustrate this problem by considering gene expression from plasmids in bacteria.

### 14.4.1. Segregational Loss

Segregational loss occurs when a cell divides such that one of the daughter cells receives no plasmids (see Fig. 14.4). Plasmids can be described as *high-copy-number plasmids* (> 20 copies per cell) and *low-copy-number plasmids* (sometimes as low as one or two copies per cell). Low-copy-number plasmids usually have specific mechanisms to ensure their equal distribution among daughter cells. High-copy-number plasmids are usually distributed randomly (or nearly randomly) among daughter cells following a binomial distribution. For high copy numbers, almost all the daughter cells receive some plasmids, but even if the possibility of forming a plasmid-free cell is low (one per million cell divisions), a large reactor contains so many cells that some plasmid-free cells will be present (e.g., 1000 l with  $10^9$  cells/ml yields  $10^{15}$  cells and  $10^9$  plasmid-free cells being formed every cell generation).

The segregational loss of plasmid can be influenced by many environmental factors, such as dissolved oxygen, temperature, medium composition, and dilution rate in a chemostat. Many plasmids will also form *multimers*, which are multiple copies of the same plasmid attached to each other to form a single unit. The process of multimerization involves using host cell recombination systems. A *dimer* is a replicative unit in which two separate plasmids have been joined, and a *tetramer* is a single unit consisting of four separate monomers fused together.