

$$\mu_{\text{net}} = \mu_g - k_d \quad (6.2b)$$

where X is cell mass concentration (g/l), t is time (h), and μ_{net} is net specific growth rate (h^{-1}). The net specific growth is the difference between a gross specific growth rate, μ_g (h^{-1}), and the rate of loss of cell mass due to cell death or endogenous metabolism, $k_d(\text{h}^{-1})$.

Microbial growth can also be described in terms of cell number concentration, N , as well as X . In that case

$$\mu_R \equiv \frac{1}{N} \frac{dN}{dt} \quad (6.3)$$

where μ_R is the net specific replication rate (h^{-1}). If we ignore cell death, k_d , then we use the symbol μ'_R ; and in cases where cell death is unimportant, μ_R will equal μ'_R .

In this chapter we will discuss how the specific growth rate changes with its environment. First, we will consider growth in batch culture, where growth conditions are constantly changing.

6.2. BATCH GROWTH

Batch growth refers to culturing cells in a vessel with an initial charge of medium that is not altered by further nutrient addition or removal. This form of cultivation is simple and widely used both in the laboratory and industrially.

6.2.1. Quantifying Cell Concentration

The quantification of cell concentration in a culture medium is essential for the determination of the kinetics and stoichiometry of microbial growth. The methods used in the quantification of cell concentration can be classified in two categories: direct and indirect. In many cases, the direct methods are not feasible due to the presence of suspended solids or other interfering compounds in the medium. Either cell number or cell mass can be quantified depending on the type of information needed and the properties of the system. Cell mass concentration is often preferred to the measurement of cell number density when only one is measured, but the combination of the two measurements is often desirable.

6.2.1.1. Determining cell number density. A Petroff–Hausser slide or a *hemocytometer* is often used for direct cell counting. In this method, a calibrated grid is placed over the culture chamber, and the number of cells per grid square is counted using a microscope. To be statistically reliable, at least 20 grid squares must be counted and averaged. The culture medium should be clear and free of particles that could hide cells or be confused with cells. Stains can be used to distinguish between dead and live cells. This method is suitable for nonaggregated cultures. It is difficult to count molds under the microscope because of their mycelial nature.

Plates containing appropriate growth medium gelled with agar (Petri dishes) are used for counting viable cells. (The word *viable* used in this context means capable of reproduction.) Culture samples are diluted and spread on the agar surface and the plates are incubated. Colonies are counted on the agar surface following the incubation period. The results are expressed in terms of colony-forming units (CFU). If cells form aggregates, then a single