Measuring Antimicrobial Activity

Antimicrobial activity is measured by determining the smallest amount of agent needed to inhibit the growth of a test organism, a value called the minimum inhibitory concentration (MIC). To determine the MIC for a given agent against a given organism, a series of culture tubes is prepared and inoculated with the same number of microorganisms. Each tube contains medium with an increasing concentration of the agent. After incubation, the tubes are checked for visible growth (turbidity). The MIC is the lowest concentration of agent that completely inhibits the growth of the test organism (Figure 26.10). This is called the tube dilution technique. The MIC is not a constant for a given agent; it varies with the test organism, the inoculum size, the composition of the culture medium, the incubation time, and the conditions of incubation, such as temperature, pH, and aeration. When culture conditions are standardized, however, different antimicrobial agents can be compared to determine which is most effective against a given organism.

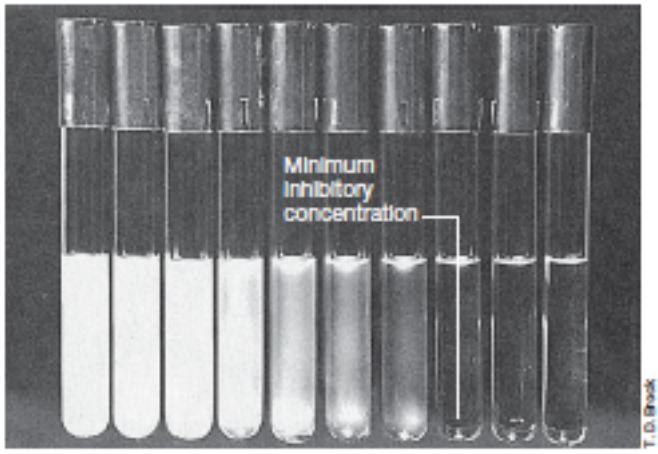

Figure 26.10 Antimicrobial agent susceptibility assay using dilution methods. The assay defines the minimum inhibitory concentration (MIC). A series of increasing concentrations of antimicrobial agent is prepared in the culture medium. Each tube is inoculated with a specific concentration of a test organism, followed by a defined incubation period. Growth, measured as turbidity, occurs in those tubes with antimicrobial agent concentrations below the MIC.

Another common assay for antimicrobial activity is the disc diffusion technique (Figure 26.11). A Petri plate containing an agar medium is inoculated with a culture of the test organism. Known amounts of an antimicrobial agent are added to filter paper discs, which are then placed on the surface of the agar. During incubation, the agent diffuses from the disc into the agar, establishing a gradient; the farther the chemical diffuses away from the filter paper, the lower is the concentration of the agent. At some distance from the disc, the effective MIC is reached. Beyond this point the microorganism grows, but closer to the disc, growth is absent. A zone of inhibition is created with a diameter proportional to the amount of antimicrobial agent added to the disc, the solubility of the agent, the diffusion coefficient, and the overall effectiveness of the agent. The disc diffusion technique and other growth-dependent methods are routinely used to test pathogens for antibiotic susceptibility.

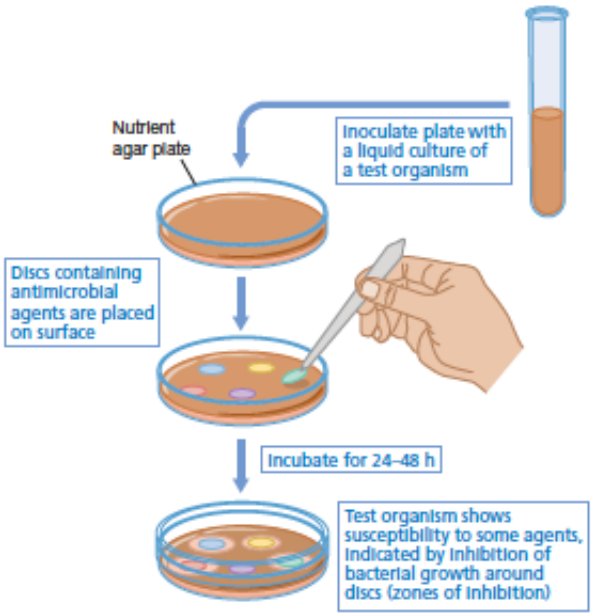

Figure 26.11 Antimicrobial agent susceptibility assay using diffusion methods. The antimicrobial agent diffuses from paper disks into the surrounding agar, inhibiting growth of susceptible microorganisms.