## LEARNING OUTCOMES

- 1. Explain the importance of optimal environmental conditions for microbial growth.
- 2. Describe methods used to transport or cultivate microbes with specific growth requirements.

## INTRODUCTION

The growth of microorganisms depends on available nutrients as well as a favorable environment. Microbes flourish where temperature, moisture, pH, and other physical conditions are optimal. These requirements significantly vary among organisms. Bacteria that grow in hot springs have different needs than those found in polar regions. Likewise, organisms such as the archaebacteria, which often require extreme environments for growth, are unlikely pathogens of humans and animals.

When cultivating organisms in a clinical setting, microbiologists must consider the type of growth medium, incubation temperature, amount of oxygen, and available water that are necessary to support growth of the suspected pathogen. For example, oxygen-free glove box chambers are designed to cultivate anaerobic organisms that might otherwise be killed if they were handled out on the bench. Commercial transport systems provide a suitable environment for most organisms that are present in a patient sample to survive between collection and delivery to the microbiology laboratory (Figure 6.1).

In this module, you will cultivate bacteria based on their oxygen requirements and nutritional need. Table 6.1 summarizes the various types of media used in these exercises.



Figure 6.1: Microbiologists use anaerobic glove boxes to work with bacteria that require oxygen-free conditions (left); a culture transport system (right).

## Table 6.1: Common Microbiological Media

Medium	Purpose	Principle	Results
BAP Blood Agar Plate	Nonselective; often used to differentiate streptococci by hemolysis	TSA with 5% sheep red blood cells; bacterial hemolysins act on red blood cells in the agar	Beta (β) = complete hemolysis, clear zone Alpha (α) = partial hemolysis, brown zone Gamma (γ) = no hemolysis, no zone
MSA Mannitol Salt Agar	Selective for Staphylococcus; differentiates between S. aureus and other Staphylococcus sp.	Salt inhibits most non- staphylococcal; <i>S. aureus</i> ferments mannitol to turn agar yellow	S. aureus = growth; fermentation Staphylococcus sp. = growth; no fermentation Gram (-) = no growth
EMB Eosin Methylene Blue Agar	Selective for Gram-negative bacteria, particularly lactose fermenting coliforms	Eosin and methylene blue dyes inhibit Gram-positives; lactose fermentation forms dark colonies	Gram (+) = no growth Gram (-) LF = purple/green sheen Gram (-) NLF = pink
MAC MacConkey Agar	Similar to EMB	Same principle as EMB except crystal violet replaces eosin and methylene blue dyes	Gram (+) = no growth Gram (-) LF = dark pink Gram (-) NLF = colorless
CET Cetrimide Agar	Highly selective for <i>Pseudomonas</i> species	Cetrimide inhibits most bacteria; enhances growth and pigment production by <i>P. aeruginosa</i>	Pseudomonas = growth with pyocyanin; fluorescent under UV Non-pseudomonads = no growth
FTM Fluid Thioglycolate Medium	Enriched, reduced broth to cultivate anaerobes and determining aerotolerance	Oxic and anoxic zones; contains a resazurin indicator which turns pink where oxygen is present	Obligate anaerobes = below resazurin Obligate aerobes = growth in resazurin Facultative anaerobes = heavier in resazurin Aerotolerant = even growth throughout