

# BIO 226N

## CULTURE MEDIA AND TECHNIQUES

### COMPOSITION OF MEDIA

#### Enriched:

Extract of beef muscle, heart, brain  
Extract of yeast cells

#### Defined - exactly known, pure chemicals:

Carbon and energy source (glucose)  
N -  $\text{NH}_4\text{Cl}$   
S -  $\text{MgSO}_4$   
P -  $\text{Na}_2\text{HPO}_4$   
Common Minerals -  $\text{Mg}^{++}$ ,  $\text{K}^+$ ,  $\text{Na}^+$ ,  $\text{Cl}^-$   
Trace Elements -  $\text{Fe}^{+++}$ ,  $\text{Ca}^{++}$   
Water

### LIQUID OR SOLID (SEMISOLID) MEDIA

Liquid solutions

Semisolid - gelling agent - agar melts at  $100^\circ\text{C}$ ; solidifies again at  $41^\circ\text{C}$ , Petri plate

### STERILIZATION

Dry Heat -  $160\text{-}170^\circ\text{C}$  - 2 hr - glassware  
Autoclave -  $121^\circ\text{C}$  15 lbs pressure/sq. inch, 15 min  
Filtration - sterile filter- 0.45 micron diameter pores  
Flame - incineration of loop  
Poison gas - ethylene oxide/plasticware  
Radiation - gamma, X-ray

### STREAK PLATE FOR PURE CULTURE

### STAINS

Simple stain - fix, add dye (crystal violet, methylene blue)  
Negative stain - background is colored  
Gram stain - crystal violet and iodine and alcohol, counter stain with safranin  
Acid-fast stain - fix, dye with heat, decolorize with acid and alcohol *Mycobacterium*