Microbial groups

CE 421/521
Lecture September 14, 2006
Vaccari et al., Chapter 10
Microbes

- Microorganisms – broad category of organisms too small to be seen with the naked eye
- Integral part of every ecosystem
- Roughly $10^6$ to $10^9$ per gram of soil, biofilm or sludge sample
Microbial groups

- **Prokaryotes**
  - *Bacteria* (Including blue green algae)
  - *Archae* – (sometimes archaebacteria)
    - classified during the 1970’s by Carl Woese and George Fox
    - Don’t fit neatly into prokaryotic or eukaryotic class due to their difference in 16S rRNA - separate kingdom?
    - Includes methanogens and halophiles

- **Viruses** – Dimitri Ivanovsky (1893) filtered sap through ceramic filters designed to remove bacteria – still resulted in tobacco mosaic virus

- **Eukaryotes**
Classification of microorganisms

Energy source:
- Chemotrophs – energy from chemical substances
  - Organotrophs – energy from organic compounds
  - Lithotrophs – energy from an inorganic compound
- Phototrophs – energy from sunlight
Classification of microorganisms

Carbon source:
- Heterotrophs – carbon from organic compounds
- Autotrophs – carbon from inorganic compounds

Can have mixed classifications:
- e.g. chemoorganoheterotroph (example E. Coli)
- chemolithoautotroph (example nitrobacter)
Classification of microorganisms

Environmental preferences:
- TEA (anaerobic, aerobic, anoxic)
- Temperature
  - Psychrophiles
  - Mesophiles
  - Thermophiles
- pH
  - Neutrophiles (5-9)
  - Acidophiles (< 5)
  - Alkaliphiles (> 9)

Extremophiles – can grow at extreme temperatures or osmotic pressures (e.g., halophiles)
Microbial Taxonomy

- Morphology: form and visible structure
  - Biochemical activities
  - Phenotype – representing observable characteristics

- Genotype
  - Characterized by DNA or RNA
  - Phylogeny – based on genetic similarities
Difficulty in that genetic exchange occurs between species not necessarily closely related

- **Strain**
  - have a recent parent cell
  - Share genetic properties with minor exceptions

- **Species**
  - Share at least 70% of DNA homology – similarity in DNA sequence
  - Or have rRNA similarity of 97% or greater

- **Genus**
  - Share at least 20% of their DNA homology
  - Or have rRNA similarity of 93-95%
### Nomenclature

1. **E**\___________ (e.g., aquaticus, marina, coli)
2. **H**\______ (e.g., bovus, avium)
3. Environmental **c**\________ (e.g., thermophilus, halophilus)
4. **S**\_______ (e.g., ovalis, longum, spaericus)
5. **C**\_______ (e.g., aureus, niger)
6. **S**\______________ (e.g., denitrificans, avium)
7. **P**\____________ (e.g., methanobacterium, cerevisiae)
8. **D**\____________ (e.g., typhi, botulinum, pneumoniae)
9. **P**\____________ (e.g., winogradskii, burkholderia)
Prokaryotes - shape

- cocci (spherical, e.g., *Streptococcus*)
- bacilli (rod shapes, e.g., *Bacillus subtilis*)
- spirilla (spiral, e.g., *Spirillum volutans*)
- filamentous
Prokaryotes - shape

Unusual

- s________ bacteria - filamentous, surrounded by a sheath
- s________ bacteria - aerobic, gram negative, at end of stalk is a “holdfast” allows it to attach to surfaces
- b________ bacteria, multiply by budding, bud grows flagellum, settles on new surface and buds again
- g________ bacteria, filamentous, gram-negative, “glide” along solids surfaces, *Beggiatoa* and *Thiothrix*: oxidize H$_2$S to S$^0$
Prokaryotes - shape

- **Bdellovibrio** - s________ (0.2-0.3μ) flagellated bacteria that prey on gram-negative bacteria

- Actinomycetes- gram-positive, f__________________, have branching filaments similar to fungi - *Streptomyces* and *Nocardia*

- Cyanobacteria - b____________-g____________ algae, procaryotes, contain chlorophyll a, have characteristic blue-green color, contain gas vacuoles that enable them to float to maximize photosynthesis, responsible for algal blooms, some are toxic
Bdellovibrio

Actinomycetes
Cyanobacteria
Fungi

e__________, produce long filaments called hyphae containing c____________
heterotrophs, use o______________ compounds for carbon and energy
found during n_________________ limitations, low D.O., low pH conditions
important in the cycling of organics – degradation of plant polymers cellulose and lignin
primarily aerobic (except for fermentative yeast)
Fungal cell wall

FIGURE 2.17  Structure of a fungal cell wall.
Algae

most are u____________________, floating, phytoplankton

some are f____________________

most are p____________________

all contain chl____________________ a, some b and c

found in o____________________ ponds, polishing ponds, aerobic lagoons
Algal Cell

**FIGURE 2.18** Structure of a typical algal cell.
Protozoa

Unicellular
Heterotrophs
Classification
- sarcodina (amoebae)
- mastigophora (flagellates)
- ciliophora (ciliates)
- sporozoa
Viruses

- small c________________ particles (not procaryotes or eucaryotes) are they alive?
- replication occurs in h________________

**Structure**
- c___________ of nucleic acid (could be double or single stranded, DNA or RNA) surrounded by protein coat (capsid)
- main shapes
  - h________________
  - p________________
  - c________________
FIGURE 2.4 Simple forms of viruses and their components. The naked icosahedral viruses (A) resemble small crystals; the enveloped icosahedral viruses (B) are made up of icosahedral nucleocapsids surrounded by the envelope; naked helical viruses (C) resemble rods with a fine regular helical pattern in their surface; enveloped helical viruses (D) are helical nucleocapsids surrounded by the envelope; and complex viruses (E) are mixtures of helical and icosahedral and other structural shapes.
Virus Replication

1. Ad______________________ - virus adsorbs to specific receptors, receptors can be polysaccharides, proteins, or lipoproteins
2. En______________________ - various particle or nucleic acid material enters cell
3. Ec______________________ - capsid is stripped away, releasing genetic material
4. Mu______________________ - viral nucleic acids are replicated using machinery of host cell
5. Ma______________________ - protein coat is synthesized and combined with nucleic acid to form nucleocapsid
6. Re_______________________ of mature virions - host cell ruptures release active viruses
Virus Detection and Enumeration

- **animal i___________________**  - newborn mice injected with inoculum and observed for signs of disease
- **t_____________ cultures**  - viruses quantified by measuring effect on host cell lines forming a monolayer on glass or plastic assay bottles, effect is measure by
- **p________________ assay**  - virus is placed on surface of host cell monolayer, virus replication leads to localized area of cell destruction called plaques
- **s__________ dilution endpoint**  - virus suspension is diluted serially and the highest dilution (smallest amount of virus) that causes a cytopathic effect in 50% of samples is reported as the tissue culture infectious dose (TCID50)
- **most p____________ number**  - serial dilutions placed in tubes or microwells with host cells, positive tubes are recorded and MPN value computed from standardized MPN table.
**MPN**

- Uses serial dilutions and statistical probabilities for the most likely number of organisms giving a positive response.

*Figure 11.13*  MPN method schematic. Positive tubes are shown as dark after incubation.
Example: Take 1 mL of sample and add to 1 L of water then perform the following serial dilutions: 10 mL, 1 mL, and 0.1 mL and incubate with substrate. If we get 5 positive tubes in the first dilution, 4 positive tubes in the second dilution, and 1 in the last dilution, what is the MPN of the sample?

Solution: from the following table we can see that the as diluted MPN is 170. Since we had a 1000 fold dilution to start with, the resulting MPN is 170,000 organisms per 100 mL

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