FARM MICROBIOLOGY 2008
PART 1: INTRODUCTION TO MICROBIOLOGY

I. The Scope of Microbiology. Microbiology is both a basic and an applied biological science. Most areas of applied microbiology can be related to farming and agriculture.

A. Definitions.

1. Microbiology. Microbiology is simply the study of microorganisms and their activities which impact just about everything on earth – humans, plants, animals, the geography and the atmosphere.

2. Microorganism. Microorganisms are unicellular organisms (or at least capable of living as single cells). This is what differentiates them from multicellular organisms like plants and animals. Also, because of their one-cell size, it usually requires a microscope to see them – hence the term "microorganism."

B. Branches of applied microbiology. (They can overlap.)

1. Medical and veterinary microbiology. Deals with all aspects of infectious disease in humans and animals.

2. Agricultural microbiology. (That's us!!) Deals with all aspects of farming – microbial reactions in soil, diseases of plants, etc. Includes what goes on with silage (also included in food microbiology).

3. Food, dairy and silage microbiology. "Food" microbiology in general concerns a wide variety of products edible to humans and animals such as sausage and other meat; sauerkraut, silage, pickles and other plant products; and dairy products. Food microbiologists seek to check the magnitude of food spoilage and seek ways to prevent it. Also how to utilize microorganisms in the production of food (such as sauerkraut, yogurt and silage). Also how pathogens can be transmitted in food and ways that such can be prevented. Dairy microbiology and silage microbiology are generally treated as specialized areas; the latter is usually not conventionally considered "food microbiology" even though the overall fermentation process is much like what goes on in sauerkraut and pickles!

4. Microbiology of water and waste treatment. Studying quality of water available to us and also the level of contamination of waste water such that the numbers of microorganisms can be reduced or eliminated as necessary (1) before waste water is ultimately dumped into the environment and (2) before it reaches us as drinking water.

5. Industrial microbiology and biotechnology. Examples: Using microorganisms to produce ethanol from grain to be used as a gasoline additive. Biotechnology can manipulate the genes of microorganisms to increase their efficiency.

II. Distribution of Microorganisms and Their Associations with Humans.

A. Distribution. Found in water, soil, air, on the surfaces of humans, plants and animals and on anything that comes into contact with water, soil, air, plants or animals. Basically, microorganisms are found everywhere on Earth where life exists, and wherever you see higher forms (plants and animals) living, you know that there are microorganisms living. Microorganisms are generally found as “mixed cultures.” As a rule, microorganisms will not be found in the blood of healthy humans. Usually, they have a substantial effect on their environment.

B. Harmful effects – examples. Bacteria have a part in each of these.

1. Disease. Microorganisms are the cause of infectious disease including anthrax, malaria, tuberculosis, tetanus, pneumonia, diarrhea, measles, colds and fever blisters.

2. Food spoilage. Most food spoilage, whether it is in a granary or a grocery store, is due to microbial growth.

a. Food as a medium for microorganisms – moisture & nutrients. Bacteria live in the moisture. If one dries out the food or adds a large amount of solute (salt, sugar, etc.) that gets dissolved in the water, one begins to eliminate growth of bacteria in the food and subsequent spoilage. As for nutrients, various components of foods can be attacked by microorganisms – especially proteins, sugars and fats.

b. Milk. When milk spoils due to fermenting organisms, one gets acidic milk which leads to lumpy milk (protein coagulated by the acid).

c. Clear liquids. With clear liquids, one sees cloudiness when bacteria achieve a high concentration.

3. Deterioration of structures, dwellings, textiles, manufactured items, etc. As for food, moisture is required for microbial growth. If enough nutrients get dissolved in the moisture, then microbial growth is probable. High humidity can provide enough moisture for microbial growth (especially molds) on solid surfaces such as shower curtains.

C. Beneficial effects – examples. Bacteria have a part in each of these as well.

1. Production of essential nutrients. Microorganisms that live in the intestinal tract produce digestive enzymes or vitamins or other nutrients useful in the nutrition of the animal.

2. Production of O_2 in the atmosphere. At least 50% of the O_2 on Earth is produced by algae and cyanobacteria – microorganisms that possess the same photosynthetic process found in regular plants.

3. Primary source of organic material. At least half of the fixed (organic) carbon on Earth is produced by autotrophic microorganisms. Autotrophs (by definition) take up CO_2 and convert it to organic matter; this is called primary production. Later we will get into “**cryptobiotic soil**” in which certain organisms start the process of “turning sand into topsoil” by utilizing nitrogen, oxygen and carbon dioxide from the air and using sunlight as a source of energy.

4. Replenishment of soil nitrogen. Many bacteria fix nitrogen, a process which removes some N_2 from the atmosphere and converts it to ammonia. Thus it is a nitrogen source for these bacteria and subsequently for use by plants and animals.

5. Biodegradation: Breakdown of complex organic materials. It is said that there is no natural organic compound that cannot be degraded by the combined activities of microorganisms. Not just natural compounds but also a lot of man-made compounds such as TNT.

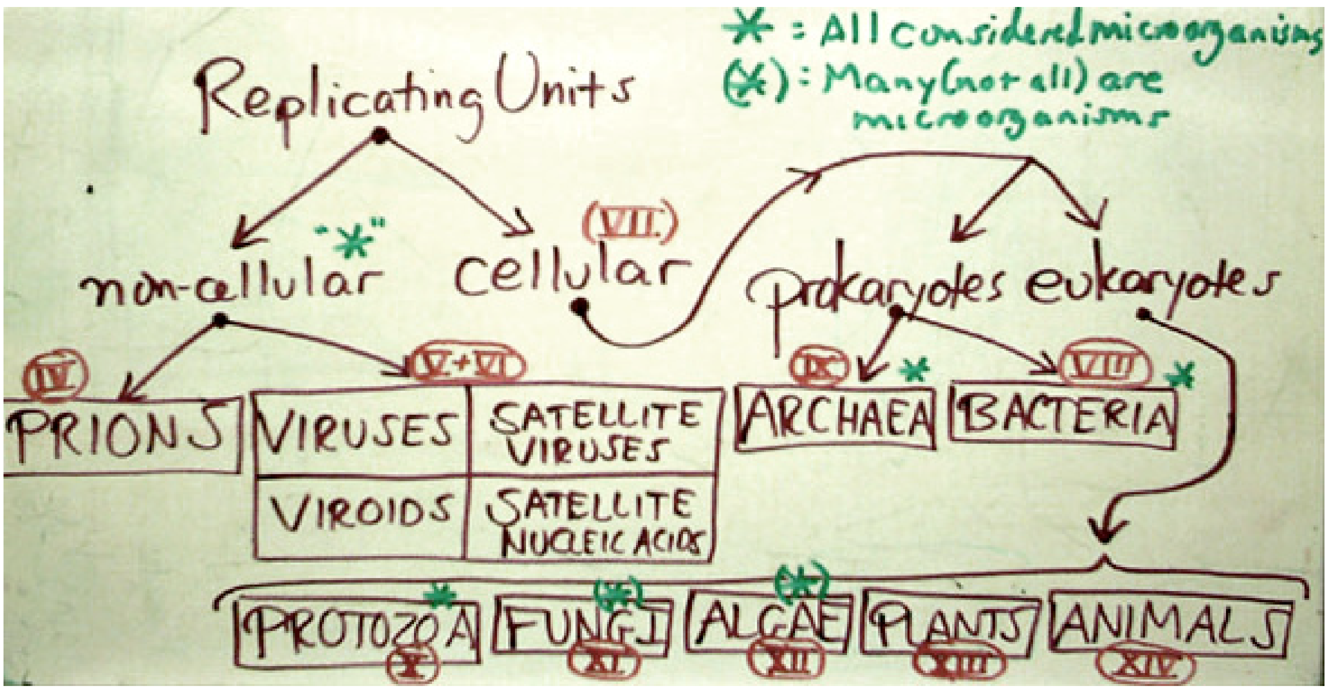
6. Source of antibiotics and drugs. Penicillin, tetracycline and most other antibiotics are produced by bacteria and molds that live in the soil.

7. Manufacture of fermented foods and beverages. Includes beer, wine, bread, yogurt, cheese, sauerkraut, silage, sausage, pickles, olives, soda crackers, tea, etc.

8. Other biotechnological products – e.g., vaccines, enzymes, genes, etc.

9. "Future uses." New fuels, foods, adhesive materials, etc.

III. General Outline of the Forms of Life (and Pseudo-Life) Represented in the Rest of The Introductory Lecture:



IV. Prions.

A. Structure and replication. Structure of prion is almost identical to protein in certain specific neurological tissue. No genetic material. Invading prions somehow cause native proteins to change to become like them – thus a "replication" of sorts.

B. Resistance. Can resist being inactivated by boiling, acid (down to pH 3), UV light and formaldehyde. Having no nucleic acids, they are then not affected by nucleases (enzymes which break apart nucleic acids). Ways in which they can be inactivated include autoclaving at an advanced temperature of 132°C for at least 2 hours (usual autoclaving is done at 121°C), boiling in a detergent, treatment with alkalis (pH 10 and above) and phenol.

C. Cause a type of amyloid disease called Transmissible Spongiform Encephalopathy (TSE). TSE's are the only amyloid disease found (so far) in which infectious agents are implicated as the cause.

D. Some TSEs:

1. **Creutzfeldt-Jakob Disease (CJD) in humans.**
2. **Bovine Spongiform Encephalopathy (BSE) – also known as mad cow disease).** BSE can be transmitted to humans, causing "variant Creutzfeldt-Jakob disease" (variant CJD, vCJD).
3. **Chronic Wasting Disease (CWD) in deer and elk.**
4. **Scrapie in sheep.**
5. **Kuru in humans.**
6. **Feline Spongiform Encephalopathy.**
7. **Transmissible Mink Encephalopathy.**

What is an "amyloid disease"?

Amyloid diseases are characterized by normal brain proteins becoming altered such that fibers and sponge-like holes are formed in brain tissue.

1. Infectious amyloid diseases are caused by **prions** and are classified under the name **Transmissible Spongiform Encephalopathy** (**TSE**).
2. Non-infectious amyloid diseases include Alzheimer's Disease, Lou Gehrig's Disease and Huntington's Disease.

V. <u>Viruses</u>.

A. General aspects. Are not cells at all, and therefore are not really organisms, although they are definitely microscopic. They also cause a variety of infectious diseases. The study of viruses is virology and it developed as a branch of microbiology.

B. Structure and replication. Viruses are made up of nucleic acid (DNA or RNA; double-stranded or single-stranded) , protein and often phospholipids. They have some of the characteristics of living organisms, but they only "come to life" (i.e., reproduce more of their kind) while infecting plants, animals, humans and various microorganisms including bacteria.

C. Some diseases:

1. **Foot and Mouth Disease (<u>not</u> "Hoof and Mouth" Disease).** Hoof consists of non-living material which virus cannot infect. Infects cloven-hoofed animals such as cattle, sheep, pigs, goats and deer. Big epidemic in 2001 among cattle in the U.K. with over 2000 cases.

2. **Asian Bird Flu.** Avian Influenza is more proper name. Can be transmitted from human to human. Major problem in Asia. Have been outbreaks in U.K. in 1992 and U.S. in the 1920s.

3. **SARS (Severe Acute Respiratory Syndrome).** First reported in Asia in early 2003; became a global outbreak later that year.

4. West Nile Virus. Major mosquito-borne disease with human cases in nearly all states of the U.S. Seasonal occurance in North America with flare-ups in summer that continue into fall._

5. Monkeypox. Small flare-up in 2003 in humans originating from pet prairie dogs.

VI. <u>Viroids and Satellites</u>

A. Viroid. Even "lower" than virus. Just a molecule of RNA – a single-stranded loop. Can infect potatoes and cucumbers and certain other plants.

B. Satellite Virus. Like a virus in that it is nucleic acid encapsulated with a protein coat. It is termed "satellite" in that it depends on a "helper virus" to multiply. Both the helper virus and the satellite infect a host cell. (Sometimes the helper virus may be unable to reproduce unless its satellite is present.)

C. Satellite Nucleic Acid. Basically just "naked" nucleic acid, and it can "hide" in the protein coat of its helper virus.

VII. Introduction to Cellular Organisms: Prokaryotes and Eukaryotes.

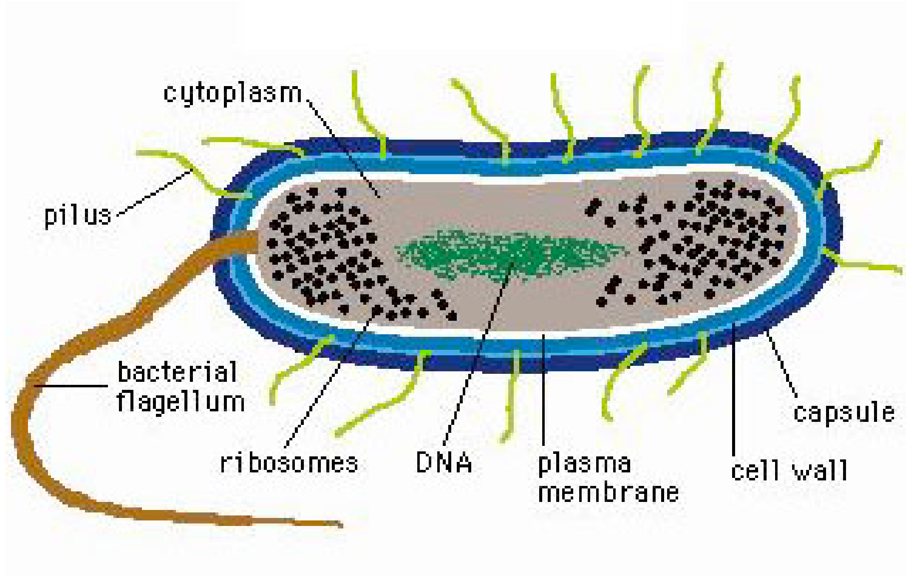
A. Prokaryotic cells. Prokaryotic means "primitive nucleus" which refers to the fact that the cell chromosome (genetic material) is not surrounded by a nuclear membrane. These organisms tend to be unicellular; all that a prokaryotic organism is and does is tied up in that one cell.

B. Eukaryotic cells. Eukaryotic means "true nucleus". The cell nucleus consists of chromosomes (DNA) enclosed by a nuclear membrane. Eukaryotes tend to be multicellular and differentiated except for protozoa, yeasts and many algae which are unicellular._

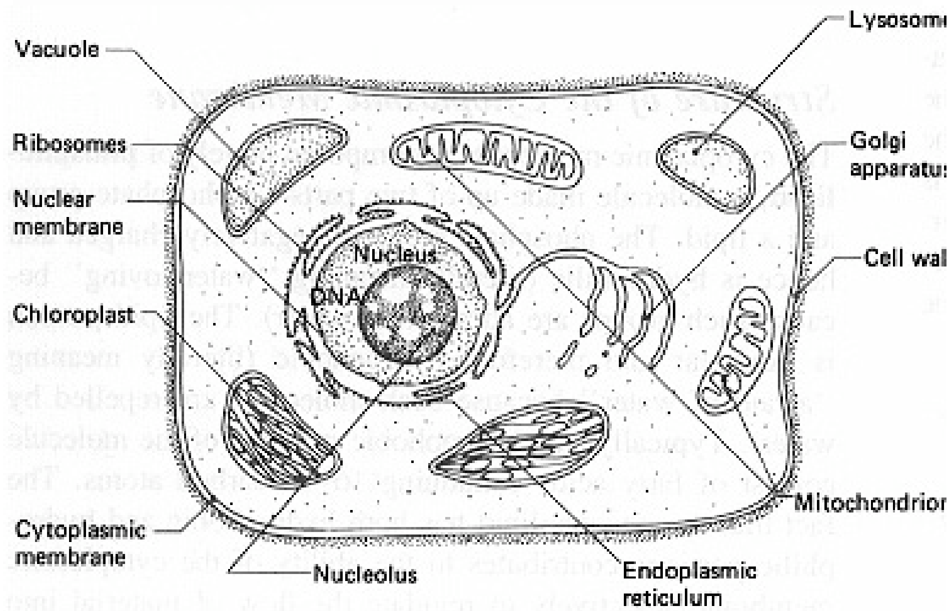
C. Summary Table & Diagrams:

Property	Prokaryotic Cells	Eukaryotic Cells
general kinds of organisms	bacteria, archae	protozoa, fungi, algae, plants, animals
nuclear membrane	absent	present
number of chromosomes	1	>1
size	relatively small	relatively large
unicellular vs. multicellular; degree of differentiation	basically unicellular and undifferentiated	may be unicellular or multicellular; also may be undifferentiated or differentiated (often to great degree)
chloroplasts and mitochondria	absent	present
mitosis and meiosis	absent	present
ribosomes (size)	70S	80S
cytoplasmic streaming	absent	present
membranes	lack sterols; contain saturated or mono-unsaturated lipids	contain sterols and polyunsaturated lipids

typical prokaryotic cell



typical eukaryotic cell



VIII. <u>Bacteria</u> (including cyanobacteria).

A. General aspects. Unicellular organisms without a true nucleus (called prokaryotes), they live everywhere that life exists including in associations with animals and plants. Have variety of beneficial and harmful effects on environment. In fact, list above regarding positive and negative aspects of microorganisms can apply to bacteria alone – apart from other microorganisms.

B. Habitats. Anywhere that you can see that life exists, procaryotes are always there. In addition, procaryotes may be the only form of life in many environments. Aside from habitats, prokaryotes may be found as “significant contaminants” – for example, the habitat for “purple non-sulfur photosynthetic bacteria” is generally thought of as being in shallow, yet anaerobic ponds that can get sunlight reaching to the bottom; these organisms can achieve high populations in an anaerobic habitat where light is available but they can get swept up in currents and into the atmosphere and can be recovered from rain, snow and icicles! (The air and precipitation are not really “habitats” for any organism, but they can be significantly contaminated by them nonetheless.)

C. Unique processes not found in eucaryotes.

1. Special types of photosynthesis. Photosynthetic bacteria other than cyanobacteria produce no O_2, and in many cases organic material (instead of CO_2) is used as the carbon source. Often such organisms assume a variety of red-based colors, such as the purple non-sulfur photosynthetic bacteria mentioned above.

2. Special types of fermentation. End products include lactic acid, butyric acid, acetic acid (used in production of vinegar), alcohols, etc.

3. Use of inorganic compounds as energy source. This is called lithotrophy.

4. Respiration without oxygen. An "oxygen substitute" such as nitrate or sulfate can be used by some organisms. This is anaerobic respiration.

5. Nitrogen-fixation. This is where molecular nitrogen (N_2) from the atmosphere is used as the source of cellular nitrogen compounds.

6. Degradation of many complex organic substances. Many bacteria decompose materials that other microorganisms cannot.

D. A few diseases of recent concern.

1. Anthrax – *Bacillus anthracis*.

2. Bubonic plague – *Yersinia pestis*.

3. Lyme disease – *Borrelia burgdorferi*.

IX. Archaea. A newly-recognized group of prokaryotes formerly included with the bacteria. Some live in extreme environments such as high temperature, low pH or high salt concentrations. Those that produce methane have relevance to farm microbiology. These methane-producers (methanogens) are unique to the Archaea; methanogens are found in no other group of microorganisms.

X. Protozoa.

A. General features and nutrition. Non-photosynthetic, unicellular eukaryotic microorganisms that were once classified as animals. (We have some similar cells in our body – specialized body cells such as white blood cells – but we would not think of classifying them as protozoa or even microorganisms, as they are still *Homo sapiens*.) Feed by ingestion of organic nutrients (generally by engulfment). Also can obtain some nutrients in solution which can get through cell membrane. A few cause some important diseases._

B. Distribution and ecology. Wherever there is moisture. Fresh and salt water and soil. Can be free-living or parasitic. Free-living types are decomposers and live by ingesting bacteria and algae. A paramecium can ingest 5 million bacteria in a day. In the sea, they feed on algae. Protozoa (zooplankton) and algae (phytoplankton) are both eaten by higher animals including some whales.

C. Some major diseases.

1. Malaria – *Plasmodium* – infects blood stream. Spread by mosquitoes. Most common serious infectious disease world-wide. "Mal-aria" is Italian for "bad air." Long associated with swamps over the past 2-3 millennia. It is said that a child dies from malaria every thirty seconds.

2. Trypanosomiasis (also known as sleeping sickness) – *Trypanosoma* – affects central nervous system. Carried by the tse-tse fly.

3. Amoebic dysentery – *Entamoeba histolytica*–intestinal parasite. Found in fecally-contaminated water. Enters body by ingestion. Causes diarrhea, abdominal pain, blood in feces.

4. Giardiasis – *Giardia lamblia* – intestinal parasite. Another intestinal parasite found in fecally-contaminated water. Most commonly- identified water-borne illness in the U.S. Mild to severe intestinal disease (vomiting, explosive diarrhea, abdominal cramps, fatigue, weight loss).

5. Cryptosporidiosis – *Cryptosporidum* – intestinal parasite. Intestinal parasite. Can get into water supply and also into milk and fruit juice (such as apple juice). One water-borne outbreak involved over 400,000 people.

XI. Fungi.

A. General features and nutrition. Eukaryotic microorganisms that have rigid cell walls and lack chlorophyll. Feed by absorption of organic nutrients. Absorptive nutrition means that their organic nutrients must be in solution (dissolved) to be taken up by the cells. (Generally the case also for bacteria and some protozoa.) These organisms live mainly in the soil and are responsible for the decomposition (biodegradation) of organic material. Microscopic fungi include molds and yeasts.

B. The microscopic fungi that are called molds.

1. Microscopic appearance – filaments and spores. Molds grow as a cottony mass of filamentous vegetative cells that eventually form fruiting bodies with colored powdery spores. Think about a mold growing on bread, fruit, cheese, the shower curtain, or a bale of hay; they grow the same way in all natural environments. Those growing on solid surfaces get their water from humidity.

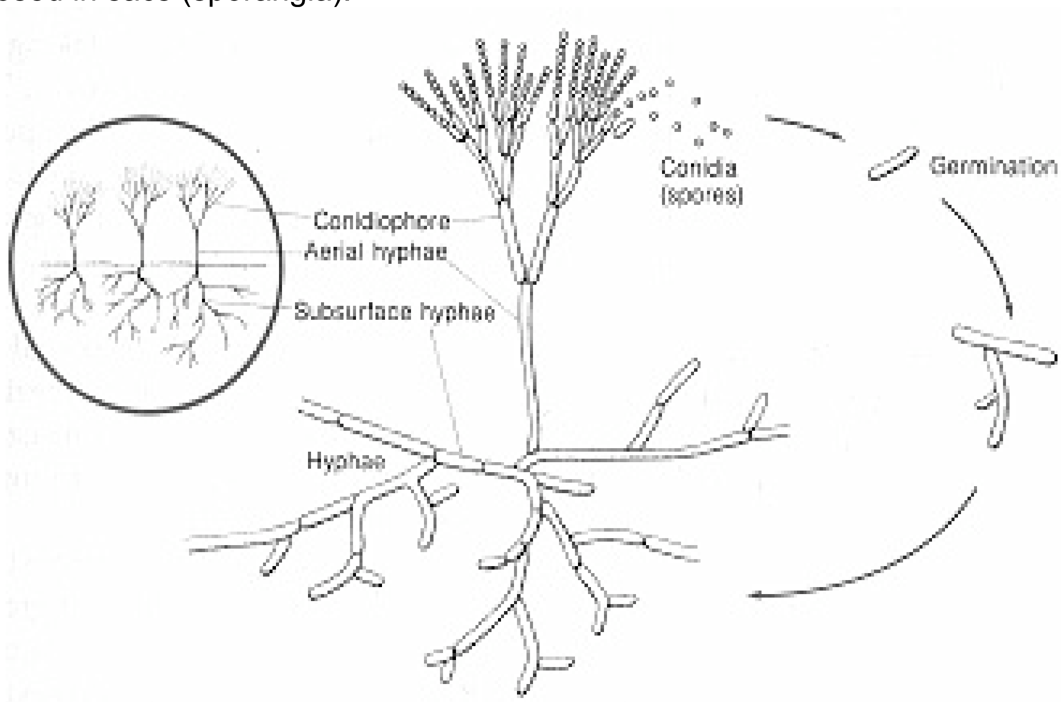
a. Vegetative structure. This is the cottony filamentous structure that is buried in the substrate to absorb nutrients – composed of filamentous cells called "vegetative hyphae." (Hypha=filament.) Erect filaments which bear reproductive spores are called "fertile hyphae." (Note figure below.)

b. Fruiting bodies (asexual reproductive spores). Formed at the tips of the fertile hyphae, the spores can be released and spread in air or water to new suitable habitats._

2. Distribution and ecology. Simply wherever there is enough moisture or where humid enough!

3. Conditions favoring growth. Air, acid or neutral food, low moisture, low temperatures, high sugar. Mainly, molds grow best in dark, moist or humid environments with suitable organic matter to decompose – like your basement, bath tile, refrigerator or silo.

4. Example of a life cycle. An example is shown below. Conidia (or conidiospores) are spores formed in chains. Some molds produce sporangiospores which are enclosed in sacs (sporangia).



asexual life cycle of the mold *Penicillium*

5. Beneficial aspects of molds.

a. Antibiotics. Most notably penicillin which is produced by *Penicillium*. Penicillin was the first commercially-available antibiotic. Tested for the first time on a human in 1941. Much of work on its effectiveness and potential for commercial production was done at UW-Madison in the 1940s.

b. Cheese-ripening. For example, a species of *Penicillium* that is involved in making blue cheese and Camembert cheeses. The blue veins of *Penicillium* in the cheese produce enzymes which alter the texture and flavor as they migrate through the cheese._

c. Other food and industrial applications. Including soy sauce, production of gluconic acid._

d. Biodegradation. Rotting of corn stalks and leaves are familiar examples. In the aerobic soil habitat of the forest floor leaves disappear in a single year through the activities of fungi – makes you wonder why we rake them in the fall._

6. Harmful aspects of molds.

a. Mycotoxins. These are non-protein toxins produced by *Aspergillus* and other molds in that grow in feeds, nuts and grains. Mycotoxins can turn up in the tissues and milk of animals and have been shown to be carcinogenic in animals._

b. Systemic diseases – including blastomycosis and histoplasmosis. Colonize the respiratory tract. Systemic diseases often begin in the lung when spores are inhaled.

c. Superficial diseases – including athlete's foot and ringworm.

d. Spoilage and deterioration. Examples: spoilage of foods and grains; deterioration of structures, clothing, etc.).

C. The microscopic fungi that are called <u>yeasts</u>.

1. Microscopic appearance – single cells, not filamentous. Generally unicellular and oval-shaped. Reproducing by budding or binary fission. Classic shape is large oval cell with buds forming.

2. Distribution and ecology. Ubiquitous (everywhere!): plants, humans & animals (inside & out), water, soil, etc.

3. Conditions favoring growth. Air, sugar, acid food, liquid, wide temperature range.

4. Beneficial aspects of yeasts.

a. Useful fermentations (bread, beer, wine, industrial alcohol) – generally performed by *Saccharomyces cerevisiae*. The overall process is this: The most commonly-used yeast that ferments sugars to ethanol and CO_2 is *Saccharomyces cerevisiae*. CO_2 is the desired end product of breadmakers that leavens bread, ethanol is the sought-after end product of brewers and winemakers. For a reliable and reproducible process, one needs to start off the fermentation of the raw product with a starter culture, as one cannot depend on a wild fermentation (i.e., fermentation by indigenous organisms) to do the job.

b. Feed yeasts. Here the yeast cells themselves are the nutrient. Such a source of "single-cell protein" is the "torula" yeast. Vitamins are also derived from yeasts.

5. Harmful aspects of yeasts.

a. Food spoilage.

b. Disease – including *Candida*. There is one yeast, *Candida*, that associates with humans in the mouth, intestine and vagina. It grows in most moist areas. It can cause disease under some conditions. In the oral cavity, it causes thrush. It is the most common cause of vaginitis. In dogs, it causes external ear infections.

D. What about fungi that are not considered "microorganisms"? Mushrooms, rusts, smuts, etc. Some are important regarding plant disease.

XII. Algae.

A. General features and nutrition. Chlorophyll-containing eukaryotic organisms that perform plant-type (O_2-producing) photosynthesis. Chlorophyll is in discrete organelles called chloroplasts. Called photoautotrophic in that CO_2 is source of carbon (such an organism is an autotroph) while light is source of energy (such an organism is a phototroph). Classic photosynthetic equation we grew up with is as follows:

$$CO_2 + H_2O \text{ ---------> } CH_2O \text{ (cell material)} + O_2$$

B. Environmental importance.

1. General distribution and ecology. Aquatic and moist environments exposed to sunlight.

2. Primary producer of organic material along with cyanobacteria. Cyanobacteria and algae are generally the first organisms to become established in a barren environment on land. In "cryptobiotic soil" habitats, these organisms – through the process of photosynthesis – utilize moisture, sunlight and atmospheric carbon dioxide to produce organic material and oxygen. Cyanobacteria can utilize atmospheric nitrogen gas which eventually finds its way into amino acids and subsequently proteins of various organisms. Bacteria and fungi also colonize this habitat, and eventually – as more and higher organisms participate in the cycling of elements – organic material builds up, forming a substrate for plants and habitat for animals. (This is an extremely abbreviated, general summary. Google "cryptobiotic soil" for a more complete picture.)

3. Phytoplankton. In the sea, algae and cyanobacteria make up the phytoplankton – free-floating photosynthetic organisms which are a major source of O_2 and also a major source of nutrients for other sea microbes and creatures (including some whales).

C. Microscopic algae. These would be those types of algae that are true microorganisms.

D. What are "blue-green algae"? This is an old name for what are now known as cyanobacteria. Even though they have been classified with algae, their overall similarity to bacteria has been noted since the 19th century.

E. What about algae that are not considered "microorganisms"? Some can attain lengths of 45 feet or more, such as the brown algae of the sea. One type is used as the source of agar – the solidifying agent in bacteriological media.

F. Associations with humans. Generally considered harmless in themselves. Large numbers can be a nuisance in bodies in water when clogging machines or decomposing. Some produce toxins in the water which poison fish (e.g., "red tide"), and some produce toxins which are retained in shellfish and cause a food poisoning (toxins not destroyed by cooking).

XIII. and XIV. Plants and Animals.