

MICR2208 Introductory Microbiology

Mid-Semester Examination Notes

PART 1 | HISTORY AND SCOPE OF MICROBIOLOGY AND TAXONOMY

Lecture 1 History and Scope of Microbiology; Lecture 2 Introduction to Taxonomy of Microorganisms

Pre-Microbiology Era (pre-1660)

Aristotle (384-322) and others believed that living organisms could develop from non-living materials, and was called **spontaneous generation**. It was believed that illnesses were caused by supernatural forces, miasmas, and imbalances of the four humours, which included blood, phlegm, cholera, and melancholy. This, however, resulted in the complete inability to control infectious diseases, such as the **Black death** (1347-1352, 23 million died in Europe), **Smallpox** (1800s, 300 million died worldwide), and **Cholera** (1848-1849, 52,000 died in London). During this era, there were sudden deaths in animals in animal husbandry, crop failure in agriculture, production of fermented foods (e.g. cheese, yogurt, beer and wine), and spoilage of foods were common (all fresh foods were eaten immediately and short term storage for dried and pickled goods).

Development of Microscopy

1590	Hans & Zacharias Janssen	Mounted two lenses in a tube to produce the first compound microscope (3- to 10-fold magnification)
1660	Robert Hooke	Published "Micrographia" and drawings and detailed observations of biological material
1676	Anton van Leeuwenhoek	First person to observe microorganisms (70- to 300-fold magnification)
1883	Carl Zeiss & Ernst Abbe	Improved microscopes (such as immersion lenses and apochromatic lenses which reduce chromatic aberration)
1931	Ernst Ruska	Constructed the first electron microscope (100,000-fold magnification)

Spontaneous Generation Controversy

1688	Francesco Redi	Refuted the idea of spontaneous generation by showing that rotting meat carefully kept from flies will not spontaneously produce maggots
1748	John Needham	If flasks of meat broth were boiled long enough (1-2 hours) and then closed, microorganisms did not grow
1861	Louis Pasteur	Proved that microorganisms do not arise by spontaneous generation

Concepts include **sterilisation** – a method that kills all living organisms, and **aseptic technique** – a series of practices to reduce contamination of a sterile site by microbes.

Pasteur's Experiment

(1) A nonsterile liquid (e.g. broth) is poured into a flask; (2) the neck of the flask is drawn out in a flame; (3) the liquid is sterilised and air is forced out the open end; (4) the liquid is cooled slowly and dust and microorganisms will appear trapped in the bend; (5) the liquid remains sterile indefinitely and an aseptic environment is produced.

Proof That Microbes Cause Disease

1835	Agostino Bassi de Lodi	Showed that disease affecting silkworms was caused by a fungus – the first microorganism to be recognised as a contagious agent of animal disease
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1847	Ignaz Semmelweiss	Demonstrated that simply washing hands between delivering babies significantly reduced the spread of childbed fever
1857	Louis Pasteur	Proposed the “germ theory” of disease

Germ Theory of Disease

1876	Robert Koch	First to cultivate anthrax bacteria outside the body using blood serum at body temperature
1877	Julius Richard Petri	Designed a flat circular dish with lid
–	Walter Hesse	Used agar as a solidifying agent (melts at 100°C, solid at 37°C)

Concepts include **pure culture** – the isolation of a single microorganism.

Koch’s Postulates

“**Koch’s postulates**”, published in 1884, was the critical test for the involvement of a microorganism in a disease. Koch’s four postulates were: **(1)** the bacteria must be present in every case of the disease, **(2)** the bacteria must be isolated from the host with the disease and grown in pure culture, **(3)** the specific disease must be reproduced when a pure culture of the bacteria is inoculated into a healthy susceptible host, and **(4)** the bacteria must be recoverable from the experimentally infected host.

However, there are exceptions such as **asymptomatic carriers** (infected by a pathogen but display no signs or symptoms that can still be transmitted to others). This is a problem in Koch’s third postulate.

Environmental Microbiology

–	Martinus Beijerinck	Developed enrichment culture technique, a way to isolate microbes with certain growth preferences
1890	Sergei Winogradsky	Isolated nitrifying bacteria in soil that use atmospheric nitrogen for growth

Concepts include **selective culture** – microbes can be cultured selectively by providing nutrients required by some but not all microorganisms in a sample, and **autotrophic growth** – use chemicals as the sole source of energy and carbon.

Plants and Microbes

1845	M. J. Berkley	Potato blight in Ireland (1845-1848) due to fungal disease
1853	Heinrich de Barry	Rust and smut fungi caused disease in cereals
1889	Beijerinck	Nitrogen fixation in root nodules performed by the bacterium, <i>Rhizobium</i> sp.
1892	Dimitri Ivanowski	Published the first evidence of tobacco mosaic virus

Concepts include **symbiosis** – each partner gains from the relationship.

How Has Microbiology Changed the Way We Live

1860	–	Separation of drinking water and sewerage
1866	–	Introduction of the flushing toilet
1798	Edward Jenner	Performs the first vaccination against smallpox
1867	Joseph Lister	Introduces aseptic techniques to surgery
1910	Paul Ehrlich	Chemotherapy for syphilis
1929	Alexander Fleming	First antibiotic, penicillin

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PART 2 | PROKARYOTIC STRUCTURE AND FUNCTION

Lecture 3 Prokaryotic Structure and Function 1; Lecture 4 Prokaryotic Structure and Function 2; Chapter 3 Bacterial Cell Structure; Chapter 4 Archaeal Cell Structure

PART 2.1 | DIVERSITY OF PROKARYOTES & BINARY FISSION

PART 2.1.1 | Size, Shape and Arrangement

Prokaryotes are generally smaller than eukaryotes, however, their size varies considerably. For example, *Escherichia coli* is an excellent representative of the average size of bacteria (1.1 to 1.5 μm wide by 2.0 to 6.0 μm long). Mycoplasma can be as small as 0.3 μm in diameter whereas some spirochetes can be 500 μm . Thus, a few bacteria are much larger than the average eukaryotic cell (typical plant and animal cells are around 10 to 50 μm in diameter).

Bacterial cells vary considerably in morphology, however there are five basic shapes. Cocci are roughly spherical cells that can exist singly or can be associated in characteristic arrangements that can be useful in their identification. Diplococci arise when cocci divide and remain together to form pairs (e.g. *Neisseria*). Long chains of cocci result when cells adhere after repeated divisions in one plane (e.g. *Streptococcus*). Cocci that divide in several planes at random generate irregular, grape-like clusters (e.g. *Staphylococcus*). Cocci that divide in two or more planes perpendicular to one another form packets (e.g. *Sarcina*, packets of eight cocci). Besides cocci, rod shaped cells exist and are called bacilli. Cells can also be spiral (spirillum), helical (spirochete) or square. Alternatively, bacterial cells can be a combination of these shapes and are called pleiomorphic.

PART 2.1.2 | Binary Fission

Most bacterial and archaeal cells reproduce by binary fission. Binary fission is a simple type of cell division: the cell elongates as new material is synthesised, replicates its chromosome, and separates the newly formed DNA molecules so there is one chromosome in each half of the cell. Finally, a septum (cross wall) is formed at midcell, dividing the parent cell into two progeny cells, each having its own chromosome and a complement of other cellular constituents.

When one bacterial cell undergoes repeated rounds of division on a solid surface, it results in a single colony composed of identical cells. This is a very fast process; *Escherichia coli*, for example, undergoes one round of cell division every 20 minutes.

PART 2.2 | BACTERIAL COMPOSITION & STRUCTURE

PART 2.2.1 | Composition

Typically, bacterial cells are composed of 70% water and 30% dry weight. This dry weight is composed of 3% DNA, 12% RNA and 70% protein found in ribosomes and enzymes. The remainder constitutes the cell wall and plasma membrane with 5% being polysaccharide, 6% lipid and 4% phospholipid.

PART 2.2.2 | The Nucleoid & Plasmids

The nucleoid is a region that contains the cell's double stranded deoxyribonucleic acid (dsDNA) chromosome that can either be circular, or linear (sometimes multiple) that is complexed to basic proteins that enable supercoiling (<0.2 μm in diameter). This dsDNA encodes for genes which are essential for life.

Plasmids are small extrachromosomal dsDNA molecules that can exist independently of the chromosome. They are usually circular (but can be linear), are supercoiled by proteins, and can exist singly or as multiple copies. Plasmids have relatively few genes, and do not encode for genes that are essential for life. Instead they encode for genes that confer a selective advantage to the bacterium in certain environments. Plasmids replicate independently of the host chromosome, but use the cell's DNA-synthesising machinery to replicate.

Type	Function	Hosts
Conjugative (e.g. F) Plasmids	Transfer of DNA from one cell to another	<i>Escherichia coli</i> , <i>Salmonella</i> , <i>Citrobacter</i>
R Plasmids	Carry antibiotic-resistance genes	<i>Pseudomonas</i> and many other Gram-negative bacteria
Virulence Plasmids	Carry virulence genes	<i>Agrobacterium tumefaciens</i>
Metabolic Plasmids	Carry genes for enzymes	<i>Pseudomonas</i>

PART 2.2.3 | The Cytoplasm

The cytoplasmic matrix is the material bounded by the plasma membrane. It is composed of 70% water, ribosomes (which are composed of ribonucleic acid [RNA] and two protein subunits: 30S and 50S) that are essential for protein synthesis and a highly-organised cytoskeleton-like system of proteins for cell division.

PART 2.2.4 | Inclusion Bodies

Inclusion bodies reside within the bacterial cytoplasm, are optional, and can either be organic or inorganic. Organic inclusion bodies include: (1) glycogen inclusions, which are α -(1, 4)-glucose polymers with branching side chains that function as an energy reserve (e.g. *Clostridium pasteurianum*), (2) poly- β -hydroxybutyrate (PHB), which are ester linked β -hydroxybutyrate polymers that also function as an energy reserve (e.g. *Ferrobacillus ferrooxidans*), (3) cyanophycin, a polypeptide of arginine and aspartic acid that acts as a nitrogen reserve (e.g. Cyanobacteria), (4) carboxysomes, which are polyhedral paracrystalline arrangements of ribulose-1, 5-bisphosphate carboxylase/oxygenase (RubisCO) that is critical for carbon dioxide fixation, and (5) gas vacuoles, clusters of gas vesicles, that provides buoyancy to some aquatic bacteria.

Inorganic inclusion bodies include sulphur and polyphosphate granules (volutin) (e.g. *Thiomargarita* and *Corynebacterium diphtheriae* respectively). Magnetosomes are found in magnetotactic bacteria and are used to orient themselves in Earth's magnetic field. They are composed of intracellular chains of magnetite (Fe_3O_4) particles enclosed within invaginations of the plasma membrane (e.g. *Magnetospirillum magnetotacticum*).

PART 2.2.5 | Internal Membranes

The internal membranes of photosynthetic cyanobacteria are called thylakoid membranes and are analogous to the thylakoids of chloroplasts. These membranes are lined with phycobilisomes that contain the chlorophyll (pigments) and photosynthetic reaction centres responsible for converting light energy into ATP.

PART 2.3 | BACTERIAL CELL ENVELOPE

PART 2.3.1 | Plasma Membrane

The plasma membrane encompasses the cytoplasm and defines the cell. The plasma membrane of bacteria is composed of amphipathic (ester-linked straight hydrocarbon chain) phospholipids organised as a bilayer that follows the fluid-mosaic model, as well as a variety of proteins. Its primary function is to act as a selectively permeable barrier that determines the transport of molecules into and out of the cell (nutrient transport). The plasma membrane is also the location of many critical metabolic processes, such as respiration.

PART 2.3.2 | Periplasm

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PART 3 | CHARACTERISTICS AND DIVERSITY OF BACTERIA

Lecture 5 Diversity of Gram-Positive Bacteria; Lecture 6 Diversity of Gram-Negative Bacteria; Chapter 21 Deinococcal, Mollicutes, and Nonproteobacterial Gram-Negative Bacteria; Chapter 22 Proteobacteria; Chapter 23 Firmicutes: The Low G + C Gram-Positive Bacteria; Chapter 24 Actinobacteria: The High G + C Gram-Positive Bacteria; Chapter 39 Human Diseases Caused by Bacteria

PART 3.1 | GRAM-POSITIVE BACTERIA

Firmicutes – Low G + C Gram-Positive Bacteria

***Clostridium* spp.**

CHARACTERISTICS: rod, obligate anaerobe, endospore-forming, fermentative, catalase negative

HABITAT: soil, human intestinal tract

<i>Clostridium botulinum</i>	Botulism (botulinum toxin), paralysis, food poisoning
<i>Clostridium tetani</i>	Tetanus (tetanus toxin), paralysis, lockjaw, convulsions
<i>Clostridium perfringens</i>	gas gangrene, food poisoning, diarrhoea

***Bacillus* spp.**

CHARACTERISTICS: rod, aerobic/facultative, endospore-forming, catalase positive

OTHER: some spp. produce antibiotics, e.g. polymyxin

<i>Bacillus subtilis</i>	Non-pathogenic, type species, one of first genomes sequenced
<i>Bacillus cereus</i>	Food poisoning
<i>Bacillus anthracis</i>	Anthrax
<i>Bacillus thuringiensis</i>	Insecticidal parasporal protein crystal

***Listeria* spp.**

CHARACTERISTICS: rod, facultative

<i>Listeria monocytogenes</i>	Listeriosis, neonatal sepsis, stillbirth Risk to pregnant women and immunocompromised individuals
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***Lactobacillus* spp.**

CHARACTERISTICS: rod, facultative, fermentative

HABITAT: human GI & genital tract

***Staphylococcus* spp.**

CHARACTERISTICS: cocci (irregular clusters), facultative, catalase positive

OTHER: associate w/ skin & mucous membranes of vertebrates

<i>Staphylococcus epidermidis</i>	Endocarditis, wound infections, surgical infections Coagulase negative
<i>Staphylococcus aureus</i>	Boils, abscesses, pneumonia, toxic shock syndrome Coagulase positive

***Streptococcus* spp.**

CHARACTERISTICS: cocci (pairs or chains), facultative, catalase negative

OTHER: Lancefield grouping system & type of haemolysis

<i>Streptococcus pyogenes</i>	Sore throat, rheumatic fever, 'flesh-eating' disease (necrotising fasciitis) GAS, β -haemolysis
<i>Streptococcus agalactiae</i>	Sepsis, Meningitis (newborn) GBS, β -haemolysis
<i>Streptococcus pneumoniae</i>	Pneumonia α -haemolysis