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The Chain of Infection and Main Groups of Micro-organisms Causing Infection

The Scope of Microbiology

Microbiology is the scientific study of organisms too small to be seen with the naked eye. They are ubiquitous and many perform essential ecological functions, for example breaking down the molecules of dead animals and plants which then re-enter the ecosystem. Some micro-organisms tolerate extreme conditions where other organisms would not survive (e.g. high temperatures) while others cause disease and are medically important.

Chain of Infection

The Chain of Infection is an epidemiological model applicable to all *pathogens* (micro-organisms able to cause disease). It comprises a series of events that must occur before pathogenic micro-organisms can spread and describes interactions between the pathogen, its host and the environment. Links in the chain are shown in Figure 1.1. Breaking a link in the chain can prevent infection.

Reservoir

The reservoir is where the pathogen lives and multiplies. Possible reservoirs include people (e.g. patients, nursing home residents, health workers and those visiting healthcare premises). They may show signs and symptoms of infection or be asymptomatic because they are mildly infected, incubating the infection or recovering from it. Two people affected by the same organism may present differently and many organisms are carried asymptomatically. For the classic

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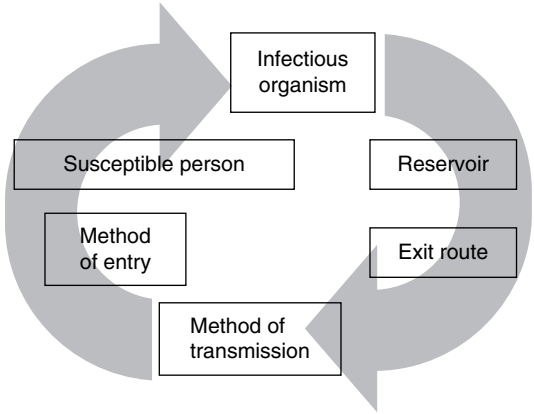


Figure 1.1 The chain of infection.

communicable diseases of childhood (e.g. measles, mumps, rubella) and influenza, other people are the reservoir. Inanimate surfaces and objects (e.g. clinical equipment, clothing) can operate as reservoirs and are sometimes described as *fomites* (Box 1.1).

The environment operates as a reservoir for infections in premises where healthcare is delivered (Box 1.2). It is also the source of infections acquired in the community (e.g. tetanus, legionnaire's disease). Animals are reservoirs for a number of infections including rabies, Ebola disease, Lyme disease and exotic emerging infections such as monkeypox (MPX virus) and zika virus.

Box 1.1 Fomites

Fomites are defined as inanimate objects that can operate as vehicles for the transmission of an infectious agent. In healthcare settings, they include patient care items (e.g. bedclothes, bedpans, urinals) and environmental surfaces. They often give rise to outbreaks of infection because disinfection has not been undertaken or undertaken poorly.

Source: Adapted from Kanamori et al. (2017).

Box 1.2 Environmental Reservoirs for Healthcare-Associated Infection

In premises where healthcare is delivered, drains and sinks can become heavily contaminated with micro-organisms and have been identified as reservoirs when outbreaks occur. Problems are compounded when sinks in clinical areas are used for non-clinical purposes (e.g. to empty washbowls). Innovations to reduce risks include sinks which self-disinfect with chemicals or heat and 'waterless wards' where conventional sinks and plumbing are removed from patient care areas.

Portal of Exit

The portal of exit is the path taken by pathogens to escape from the reservoir. Respiratory pathogens (e.g. colds, influenza) are released in coughs, sneezes and spluttered speech. Enteric pathogens causing food-poisoning escape in vomit and faeces. Skin scales and dust can provide a portal of exit for some bacteria, including those frequently causing healthcare-associated infection (e.g. *Staphylococcus aureus*).

Mode of Transmission

The mode of transmission describes how the pathogen spreads (Table 1.1).

Transmission can occur by direct contact between surfaces such as hands and fomites and via contaminated food and water (e.g. cholera, typhoid). Houseflies (*Musca domestica*) breed in faecal material. Their feet become contaminated with micro-organisms which can be transferred to open wounds. This type of zoonosis is called *mechanical transmission*. It has been documented as a means of spread for *Clostridioides difficile*, methicillin-resistant *S. aureus* (MRSA), *Escherichia coli* and *Salmonella* spp. (Davies et al. 2016). Biological zoonotic transmission occurs when the pathogen lives and multiplies inside a vector. The infectious agent causing malaria (*Plasmodium* spp.) lives inside female mosquitoes (*Anopheles* spp.). Infection is transmitted when the mosquito bites a human host.

The air-borne route is an important mode of transmission for respiratory pathogens and the classic communicable diseases. The 2020 COVID-19 pandemic stimulated renewed interest in air-borne transmission. Conventionally, it was thought to occur via two distinct routes: droplet and air-borne transmission (Table 1.2).

Droplet transmission was proposed according to the findings of research dating from the 1930s that described the theoretical behaviour of particles according to their size (Wells 1934). From this work it was concluded that respiratory secretions are spread in two distinct ways according to their dimensions. According to this school of thought, droplets are thought unlikely to remain air-borne for long

Table 1.1 Modes of microbial transmission.

Direct contact
Indirect spread via fomites
Air-borne spread
Contaminated food and water
Inoculation via skin or mucous membranes
Vertical transmission from mother to foetus
Zoonotic spread from animals

Table 1.2 Respiratory transmission.

Term	Definition	Implications for practice
Droplet	Transmission by large droplets, diameter >5 μm transported via turbulent air flow generated by violent expiratory events (e.g. coughing or sneezing)	Most likely at close range with pathogens deposited on the conjunctivae or mucous membranes of new hosts
Air-borne/ aerosol/droplet nuclei	Transmission via inhalation of small respiratory droplets, typically <5 μm	Remain air-borne long enough to transmit the pathogen over distance and does not depend on face-to-face transmission. Pathogens are deposited deep in the respiratory tract as far as the alveoli

periods. Instead, they fall through gravity because of their relatively large size and their period of infectivity is correspondingly brief.

Aerosols were thought to remain suspended for much longer because of their minute size, depending on environmental conditions (e.g. humidity, turbulence, ventilation) (Tang et al. 2021). It has now been suggested that the distinction between air-borne, aerosol/droplet nuclei and large droplet transmission should be replaced by a unique non-contact air-borne transmission mode (Drossinos et al. 2021).

For many pathogens, there is more than one mode of transmission. Norovirus is spread by droplets released when vomiting occurs, by direct and indirect contact and by eating contaminated shellfish (Hassard et al. 2017). Many viruses responsible for respiratory and gastrointestinal infections can probably also be spread by fomites contaminated with body fluids (Boone and Gerba 2007).

Portal of Entry

The portal of entry is the route taken by pathogens to gain access to the tissues of the new host. The micro-organisms responsible for influenza and the classic communicable diseases are inhaled. Enteric pathogens gain access by ingestion. The urogenital tract is the portal of entry for urinary and sexually transmitted pathogens. Inoculation via skin and mucous membranes is the mode of entry for pathogens causing surgical site infection. Needlestick injury allows access for pathogens causing blood-borne infection: the viruses responsible for human immunodeficiency disease (HIV) and hepatitis B and C. Vertical transmission from mother to foetus occurs by two possible mechanisms: from the maternal to foetal circulation via the placenta (e.g. congenital syphilis) and via the contaminated birth canal (e.g. congenital gonorrhoea). Some pathogens are transmitted vertically via both routes (Box 1.3).

Box 1.3 Vertical Transmission: Group B Streptococcal Infections in Neonates

One in four women carry Group B *Streptococcus* vaginally without symptoms. It can infect the amniotic fluid before delivery or be acquired during passage down the birth canal, causing neonatal meningitis, pneumonia and septicaemia. Very low-weight babies are at greatest risk of developing severe infection and mortality can be as high as 30%. Infection is either early onset (during the first week of life) or late onset occurring when the infant is between a week and six months old (Heath and Jardine 2014).

Women who are carriers can be identified by screening. Vaginal or rectal swabs are taken at 35–37 weeks gestation. Women who test positive should receive antibiotics during labour or earlier if the membranes rupture before labour commences to avoid the risk of neonatal infection.

Intact skin and mucous membranes are usually good barriers against many potential pathogens but are overcome by the invasive procedures commonly undertaken during healthcare (Stamm 1978).

Interaction Between Host and Pathogen

The immune status of the host, size of the infective dose and virulence of the pathogen are key determinants of infection. This is a complex area currently receiving scientific scrutiny. A key question is why when two people are exposed to the same pathogen, one succumbs but often the other does not.

Some individuals are more susceptible than others. Genetic variation can play an important part. People with sickle cell disease and thalassaemia have inherited innate immunity to malaria. Susceptibility to the same pathogen can also vary between individuals. The likelihood of contracting tuberculosis and HIV appears to be influenced by genetic factors. Immunity can be acquired throughout the lifespan through exposure to infection or vaccination. Some infections are species specific. Few domestic animals are susceptible to viruses able to cause the common cold, for example. Norovirus is a human pathogen but humans are not susceptible to the related feline calicivirus.

Infectious Dose

The infectious dose is the number of organisms required for infection to occur. It varies widely and is much lower for some pathogens than for others (Schmid-Hempel and Frank 2007). It has been estimated that in the case of SARS-Cov-2, the infectious dose could be as few as 100 virus particles (Karimzadeh et al. 2021). In general, the larger the infective dose, the more likely it is that the pathogen will be able to overwhelm the host immune system. Exposure to a patient with tuberculosis

who is exhibiting a productive cough generating prodigious amounts of sputum is much more likely to result in infection than exposure to an asymptomatic patient. The frequency of exposure is also important. People who have multiple sexual partners are more likely to develop sexually transmitted infection and their risk of contracting more than one type of sexually transmitted disease is greater.

Some individuals and situations operate as super-spreaders (Box 1.4). Risky behaviours very likely to result in the transmission of respiratory infection between members of the public have also been reported, particularly in relation to COVID-19; risk is likely to be heightened through exposure to people who are inebriated or emotional.

Super-spreading occurs when a single individual infects a disproportionately large number of contacts (Wong et al. 2015). The outbreaks of Ebola disease 2014–2015 in Guinea and Sierra Leone, the outbreak of Middle Eastern respiratory syndrome (MERS) in 2015, severe acute respiratory syndrome (SARS) in 2003 and more recently the 2020–2021 COVID-19 pandemic have increased interest in super-spreading.

Virulence

Virulence is the ability of a pathogen to cause disease. Severity of the resulting infection is greater if the pathogen is highly virulent and the potential host is exposed to a large number of organisms (infective dose). Virulence factors are the properties of a pathogen that increase its ability to invade potential hosts, colonise them and evade the host defences. Morphology influences virulence.

Pili are minute hair-like processes that enable bacteria to attach to the surface of the host cell. Piliated strains of *Neisseria gonorrhoeae* enable the bacteria to attach to the cervical epithelium. Non-piliated strains are less likely to cause infection.

Box 1.4 Factors Likely to Contribute to Super-Spreading

- Individual behaviour, e.g. particular tendency to generate spluttered speech
- Number of susceptible victims present
- Air flow (respiratory infections)
- High population density (opportunity for spread by direct and indirect contact is increased)
- Specific environmental conditions – the outbreak of Ebola disease in 2014–2015 was associated with traditional burial customs that placed family members of the deceased at risk through contact with body fluids
- Examples of situations likely to promote super-spreading are:
  - hospitals
  - prolonged contact with members of the same household
  - mass travel (aeroplane)

A mucus capsule surrounding the bacterial cell wall helps prevent desiccation for some bacterial strains. Some of the earliest epidemiological investigations exploring the spread of healthcare-associated infection established that strains of *Klebsiella* with mucus capsules were more likely to cause outbreaks than non-encapsulated strains because they could survive desiccation, remained viable for longer on hands and fomites and were more easily transmitted (Casewell and Phillips 1978).

Some bacteria release enzymes that increase their *pathogenicity* (ability to cause disease). Haemolysins are enzymes that destroy red blood cells. *S. aureus*, streptococci and enterococci release haemolysins. Cytotoxins released by *Salmonella* kill cells by destroying proteins. *S. aureus* releases collagenases. These enzymes destroy the protein collagen present in connective tissue. The damaged collagen forms a protective wall around the bacteria, resulting in the formation of an *abscess*. Abscesses contain dead *neutrophils* (pus), cellular debris and live bacteria protected from *phagocytosis* by neutrophils in the tissue fluid of the host.

The structure of bacterial cells can also increase virulence. For example, *Mycobacterium tuberculosis* has a tough, waxy coat that protects it against host defences, including phagocytosis. Many bacteria have numerous virulence factors. Group B *Streptococcus*, which can cause serious infection in pregnant women, neonates and older people, is protected from phagocytosis by its outer polysaccharide coat, pili which enable it to attach itself to host cells and ability to release an enzyme (C5a-ase) that allows it to evade the host immune defences by preventing the migration of neutrophils to the infected site.

Main Groups of Micro-organisms

The main groups of micro-organisms are shown in Table 1.3.

Table 1.3 Main groups of micro-organisms able to cause disease.

Bacteria
Viruses
Fungi
Protozoa
Mycoplasmas
Rickettsiae
Chlamydiae
(Parasitic worms – infestations)

Bacteria

Bacteria are the oldest known group of cellular micro-organisms, thought to have appeared four billion years ago. They are found everywhere but most are *saprophytes*. They live on dead organic matter and play a vital role in degrading complex organic molecules and recycling them into simpler ones used by other organisms to support metabolism. Many species of bacteria are commercially important, for example making wine. The characteristics of bacteria are the same as those of all living organisms (Table 1.4).

Approximately 50 species of bacteria cause disease in animals, plants and humans but virulence is highly variable. For example, *Yersinia pestis*, the pathogen responsible for plague, always causes very serious infection. In contrast, some bacteria called *opportunists* do not attack healthy tissues and are likely to cause serious infection only in people who are already sick and whose immune systems are compromised. Bacteria responsible for many healthcare-associated infections (e.g. *Klebsiella* spp., *Escherichia coli*, *Pseudomonas* spp.) are opportunistic.

The normal *commensal flora* is made up of bacteria and other micro-organisms living harmlessly in or on the body, for example on the skin or in the gastrointestinal tract. They receive nourishment and shelter from the host and in return protect the host from other invading pathogens in a reciprocal arrangement. Commensal micro-organisms can cause infection if they are transferred to a different anatomical location (Table 1.5).

Endogenous (self) infection results in the transfer of micro-organisms from one anatomical site to another on the same person. Transfer of *E. coli* from the perineum to the urinary tract of the same individual is an example. *Exogenous (cross) infection occurs* when micro-organisms are transferred between different people, for example on the hands of health workers or via fomites. Infection prevention and control strategies are aimed at containing both endogenous and exogenous transmission.

Table 1.4 Characteristics of living organisms.

Cellular structure apparent
Respire and generate energy
Grow and adapt
Metabolise (consume food and convert it into energy)
Maintain homeostasis (keep a stable internal environment despite external environmental change)
Respond to environmental change
Move
Reproduce and transfer genetic material to offspring

Table 1.5 Infections caused by the normal commensal flora: examples.

Escherichia coli: urinary tract infection
Staphylococcus aureus: surgical site infection
Clostridiodes difficile: overgrowth in the large intestine after treatment with broad-spectrum antibiotics
Enterococcus faecalis: urinary tract, wound or bloodstream infection
Streptococcus pneumoniae: pneumonia, otitis media, meningitis, bloodstream infection
Bacteroides fragilis: abscesses, peritonitis

Infection and Colonisation

Understanding the difference between *infection* and *colonisation* is key to interpreting information on microbiology reports. Infection occurs when a pathogen gains access to the host tissues and elicits a host response, giving rise to the signs and symptoms of infection, such as *pyrexia*, *inflammation* and pus in a wound or the appearance of neutrophils ('pus cells') in urine. Colonisation occurs when pathogens are present but there is no host response and no signs or symptoms of infection.

Infection is very likely to be present if there are large numbers of the same type of pathogen accompanied by large numbers of neutrophils, pain, tenderness and pyrexia. A microbiology report indicating scant growth or 'mixed growth' of a number of different types of micro-organisms with few neutrophils suggests colonisation. Colonisation is clinically significant because the colonised site operates as a reservoir and the micro-organisms can be transferred to a susceptible site such as a wound via hands or fomites. Colonisation often occurs before infection develops.

Bacterial Growth Requirements

Bacteria share the same characteristics of life as higher organisms (see Table 1.5). All bacteria require water but otherwise their growth requirements vary enormously, reflecting the wide variety of environments they inhabit. Gram-negative bacteria have very simple growth requirements, needing only water, simple nutrients and warmth to multiply. Most human pathogens grow and reproduce optimally at 37 °C and thrive in the warm, damp, densely populated hospital environment, explaining why risk of infection, especially from opportunists, is high. A key measure of growth is the doubling time which varies according to environmental conditions. For *E. coli*, doubling time is about 20 minutes in a nutrient-rich environment but 15 hours under less favourable circumstances (Gibson et al. 2018).

Keeping the clinical environment and equipment dry and clean helps eliminate reservoirs and reduces the risk of cross-infection caused by Gram-negative

Box 1.5 Risks of Cross-Infection caused by Gram-Negative Opportunists

Gram-negative bacteria have been isolated from damp cleaning equipment in wards and other items (e.g. cleaning cloths, mop heads, wash bowls, infant feeding bottles) where they multiply in large numbers, forming reservoirs that have been associated with outbreaks, and from equipment left immersed in disinfectant solution. Cross-infection should be avoided by using disposables and by drying other equipment after use and storing items dry.

Source: Adapted from Ayliffe et al. (1970).

opportunists (Box 1.5). Gram-positive bacteria are generally able to tolerate dry conditions better than Gram-negative bacteria. They can survive in dust, forming reservoirs.

Some species of bacteria, described as fastidious, have very exacting growth requirements. They need specific nutrients and some do not survive for long outside the host or are unable to survive at all (e.g. *Treponema pallidum*).

All bacteria need to respire and generate energy to metabolise. *Aerobic bacteria* utilise oxygen and thrive in wounds close to the skin. Aerobic bacteria are described as *obligate aerobes* if they depend entirely on oxygen to support metabolism (e.g. *M. tuberculosis*). *Obligate anaerobes* are killed in the presence of atmospheric oxygen and use a different source of energy to respire (e.g. *Bacteroides fragilis*, *Clostridium pefringens*, *C. tetani*). They are found in deeper wounds and within abscesses. *Facultative aerobes* use oxygen to support metabolism if it is present but can switch to anaerobic respiration if it is not (e.g. *S. aureus*, *Staphylococcus epidermidis*, *E. coli*). Most human pathogens are aerobic.

Bacteria vary according to the pH optimal for their survival and this influences distribution of the normal flora. An acidic environment favours *S. aureus* which multiples well on skin which is acidic (pH 4). Lactobacilli thrive in the acidic vaginal environment throughout the female reproductive years. Decrease in oestrogen at the menopause reduces vaginal acidity and the lactobacilli tend to be replaced by other micro-organisms. Infection and vaginal discharge may result. The very low pH of gastric secretions (pH 2) destroys many of the bacteria ingested in food but enteric pathogens such as *Salmonella typhi* (causing typhoid) grow and multiply in the alkaline environment of the small intestine (pH 8).

Bacterial Structure

Bacteria are *unicellular* organisms. This means that one cell on its own operates as an independent, viable organism. There is no division of labour between bacterial cells: one cell performs all the functions of living independently of any others.

Before examination in the microbiology laboratory, all cells and tissues must be 'fixed' (killed) and stained with laboratory dyes to make them visible. Neither the light microscope or the electron microscope (which makes it possible to examine specimens at much higher magnification) depicts organisms in their living state. Electron microscopy makes it possible to study the cellular *ultrastructure* (fine, detailed structure) of bacteria and other cells and tissues.

All bacteria are *prokaryotic*. This means that their genetic material (DNA) lies directly in the cell cytoplasm instead of being separated from it by a nuclear membrane. Bacteria contain a single chromosome carrying their genes. Higher organisms are described as *eukaryotic*. Their chromosomes are separated from the cytoplasm by a nuclear membrane and they contain organelles responsible for cellular respiration (mitochondria) and synthesising protein (ribosomes) which are membrane bound. Prokaryotic cells are simpler in structure than the cells higher organisms. Instead of being equipped with discrete organelles, they contain intracellular membranes called *mesosomes* that undertake cellular functions. Figure 1.2 illustrates an 'idealised' bacterial cell demonstrating all the features possible.

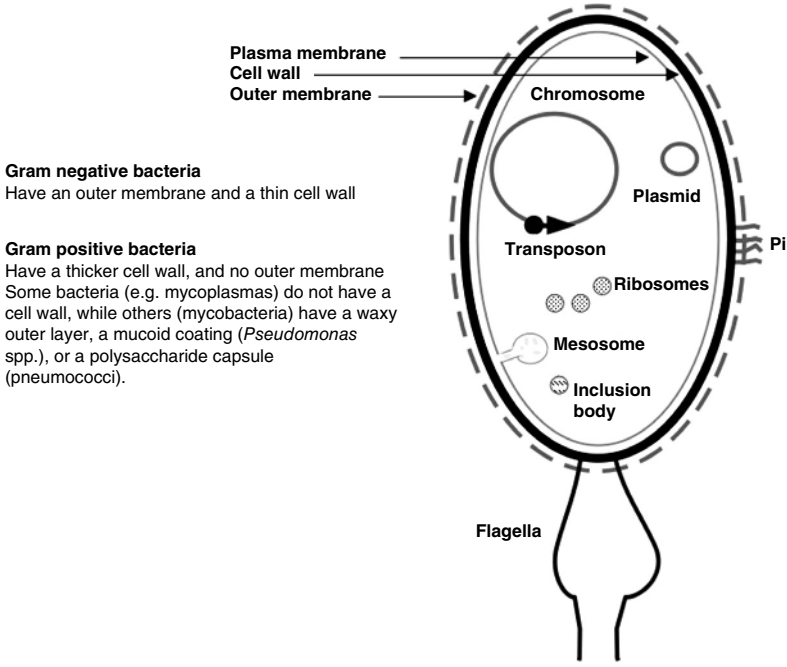


Figure 1.2 The 'idealised' bacterial cell.

Bacteria have rigid cell walls giving structural support while protecting the cellular contents. The walls consist of a mesh of polysaccharides and amino acids called peptidoglycans. Historically, bacterial cell walls have been of intense interest because they determine the staining properties of the bacterium (reaction to Gram's dye), many aspects of bacterial behaviour and are the target for antibacterial drugs.

The cell walls of Gram-positive bacteria consist of a very thick peptidoglycan mesh which enables the bacteria to withstand the immune system of the host although they are still susceptible to some enzymes, including lysozyme. Gram-negative bacteria have more complex cell walls consisting of a thin layer of peptidoglycan surrounded by an outer membrane made up of protein, phospholipid and lipopolysaccharide molecules (endotoxins). Endotoxins are highly toxic to eukaryotic organisms and are responsible for many of the signs and symptoms arising when infection occurs. The outer membrane of Gram-negative bacteria enables them to withstand the action of many disinfectants but its relative thinness compared to Gram-positive bacterial cell walls makes them more susceptible to desiccation. The β-lactam ring of the penicillin molecule prevents the synthesis of peptidoglycan, allowing it to destroy the bacterial cell. Eukaryotic organisms do not contain peptidoglycan, explaining why penicillin can be given safely to treat infection. Many other antibiotics also target bacterial cell walls.

The difference between prokaryotic and eukaryotic cells is clinically important because prokaryotic organisms are damaged or destroyed by antimicrobial drugs that do not have the same effect on eukaryotic organisms. Antibiotics operate as 'magic bullets' specifically able to target bacteria without damaging the host in the same way. Some antibiotics prevent the synthesis of bacterial cell walls (e.g. cephalosporins, vancomycin). Others prevent the synthesis of bacterial proteins (e.g. gentamicin, erythromycin) or the formation of bacterial DNA (e.g. trimethoprim, co-trimoxazole).

Some bacteria are equipped with whip-like flagellae. This enables them to be highly motile, e.g. *Salmonella* spp. which cause food poisoning and *Proteus* spp. which are opportunistic and able to cause healthcare-associated infections. Other species are equipped with minute hairs called pili.

Classifying Bacteria

Bacterial Morphology

Bacteria are classified according to their morphological characteristics (shape) (Figure 1.3).

Cocci are round. They form clusters (e.g. *S. aureus*), chains (e.g. *Streptococcus pneumoniae*) or occur in pairs called diplococci (e.g. *N. gonorrhoeae*). *Bacilli* are rod-shaped bacteria occurring singly or in chains (e.g. *E. coli*, *Pseudomonas*, *Klebsiella*, *Proteus*, *Serratia*, *Salmonella*). *Vibrios* are curved (e.g. *Vibrio cholerae*, *Campylobacter*). Spirochaetes are tiny, flexible spirals (e.g. *T. pallidum* [syphilis], *Leptospira* [Weil's disease] and *Borrelia* [Lyme disease].

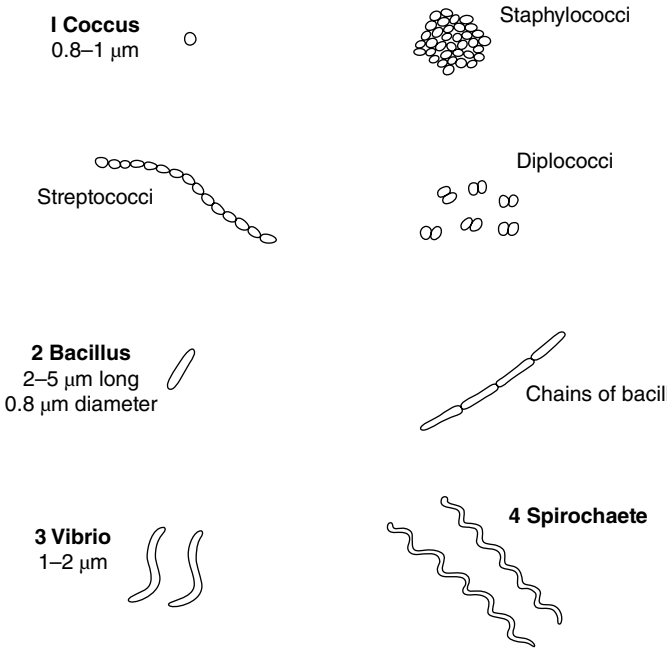


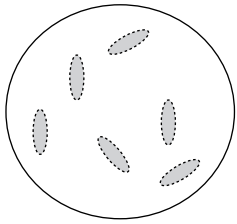
Figure 1.3 Bacterial morphology.

Gram Staining Reaction

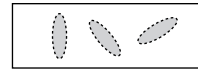
Gram staining is the first step taken to identify bacteria in a laboratory specimen (Figure 1.4). Reaction to Gram's dye is determined by the chemical composition of the bacterial cell wall.

Gram-positive bacteria take up and retain Gram's stain, appearing blue. They are generally resistant to desiccation, tolerate dry conditions and are not flagellated or motile. Some species sporulate. They develop a thick layer of peptidoglycan around themselves, protecting the cellular contents against adverse conditions such as desiccation or lack of nutrients. When conditions become less hostile, the spores hatch to release vegetative bacteria able to grow and reproduce. *Bacillus cereus* is a Gram-positive species well known for causing food poisoning associated with Chinese meals. The spores, which are often present in rice, survive boiling. If the rice is then kept at a warm temperature for several hours, the vegetative bacteria emerge and multiply to form a large infective dose. Gentle reheating (to make 'special fried rice') does not destroy them and if ingested, the result is gastrointestinal upset.

Gram-positive bacteria release chemicals called *exotoxins*. Exotoxins are released outside the bacterial cell into the extracellular fluid of the host where they dissolve and are carried throughout tissues. They either destroy host cells or inhibit specific



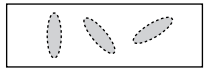
Identify organism of interest



Prepare a smear of the bacteria and then use heat to fix the bacteria to the slide

Flood the slide with 0.5% crystal violet and leave for 30 seconds. This will dye the bacteria purple.
Wash the slide with water

Flood the slide with (1%) Lugol's iodine (also known as Gram's iodine) and leave for 30 seconds.
Wash the slide with water

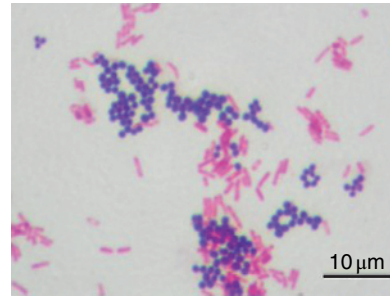


Decolourise with 95–100% ethanol or acetone until colour ceases to run out of the smear
Wash the slide with water

Flood the slide with 0.1% safranin as a counterstain. Wash the slide with water and blot dry
Examine the slide to observe cell morphology and Gram reaction

Gram positive organisms stain blue or purple.

Gram negative organisms stain pink or red.



Staphylococcus aureus a Gram positive bacterium in purple, and *Escherichia coli* a Gram negative bacterium in pink

Figure 1.4 The Gram staining reaction.

cellular functions and can have very serious, often lethal effects. The exotoxin released by *C. tetani* is a potent neurotransmitter responsible for the paralysis caused by tetanus. Its release causes the muscles to go into spasm. Tetanus used to be called 'lockjaw' because of its effects on the facial muscles. If paralysis of the respiratory muscles occurs, the result is respiratory arrest, progressing to cardiac arrest and death. Ingestion of the exotoxins released by *S. aureus* causes food intoxication. The signs and symptoms are rapid-onset diarrhoea and vomiting, often within six hours. The illness is often described as 'mild' but can last several days, resulting in dehydration, especially in older people. Food intoxication cannot be prevented by heating food to a high temperature because exotoxins are heat resistant.

Gram-negative bacteria do not retain Gram's stain and appear red under the microscope. They thrive in damp situations, usually have very simple growth requirements, are often flagellated and motile, do not sporulate and many species are intrinsically resistant to antibiotics. *Endotoxins* forming part of cell wall are released when the bacteria die, causing fever and malaise when they escape into the tissue fluids of the host. The symptoms of typhoid (*S. typhi*) and meningitis (*Neisseria meningitidis*) are caused by endotoxin release.

Mycobacterium tuberculosis does not respond well to Gram's stain and is sometimes described as 'acid fast'.

Bacterial Reproduction and Genetics

The way that bacteria reproduce has important clinical implications. All bacteria are able to reproduce asexually by *binary fission*. The parent cell divides into two genetically identical daughter cells, each receiving half the genetic material of the parent (Figure 1.5a). There is no genetic variation and no scope for adaptation to the environment (evolution). The warmth and dampness prevailing in healthcare premises provide ideal conditions for bacterial growth and multiplication. Division can occur every 30 minutes, generating large reservoirs of bacteria, increasing the risk of transmission. If a laboratory specimen is stored at room temperature, any bacteria that it contains multiply rapidly, leading to heavy levels of contamination. The findings of laboratory tests are then more difficult to interpret.

Some species of bacteria are capable of exchanging genetic material, enabling them to adapt to changing environmental conditions. Those best adapted to the environment survive and multiply at the expense of those less well adapted. This process involves the exchange of a circle of extracellular DNA called a *plasmid*. Plasmids are clinically important as they often carry genes conferring resistance to antimicrobial substances (Bennett 2008). Plasmids can replicate and when the bacterial cell, divides each daughter cell will contain a copy of the plasmid. Plasmid transfer between bacteria is possible via four routes (Box 1.6).

Clearly, the progeny arising through plasmid exchange are not genetically identical. Having a gene that enables the bacterium to resist the effects of

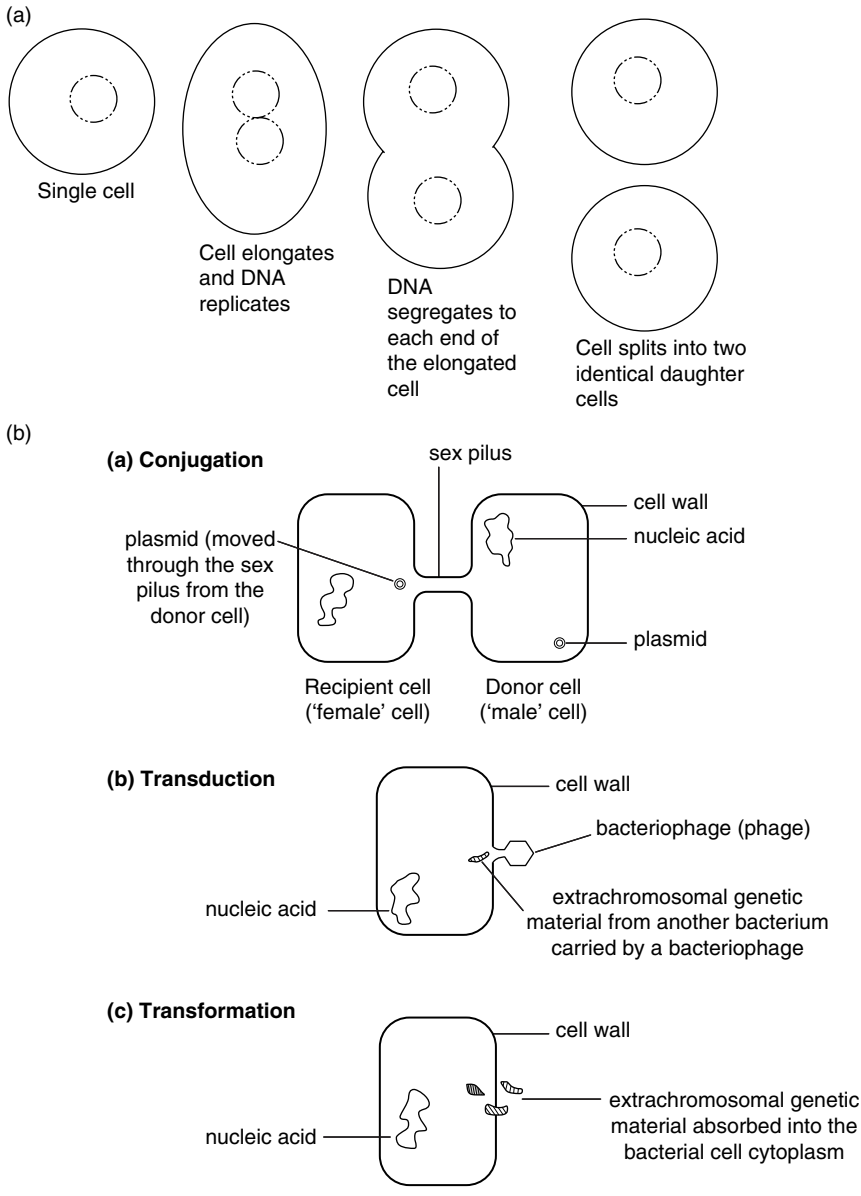


Figure 1.5 (a) Binary fission. (b) Plasmid-mediated bacterial reproduction.

Box 1.6 Mechanisms of Plasmid Transfer

Transformation occurs when some bacteria ('competent bacteria') take up extracellular DNA released into the environment from decomposing or disrupted cells, viral particles or DNA excreted by living cells via their cell walls. This DNA is either carried in the cytoplasm as a plasmid or integrated into the bacterial genome (Thomas and Nielsen 2005).

Conjugation involves the development of a junction between two bacteria, genesis of a pore and a channel called a sex pilus that allows the passage of a plasmid from one cell to the other.

Transduction involves plasmid transfer to a bacterial cell that has become infected with a virus particle called a bacteriophage.

Transposable elements are sequences of DNA able to alter position by moving from a chromosome to a pre-existing plasmid. Once incorporated into the plasmid, they can be transferred to other bacteria through conjugation or transduction. The transposon Tn*1546,* which contains a cluster of genes known as *vanA*, can be transferred between bacteria (Gardete and Tomasz 2014) and has been linked to vancomycin resistance in *S. aureus* (Sievert et al. 2008).

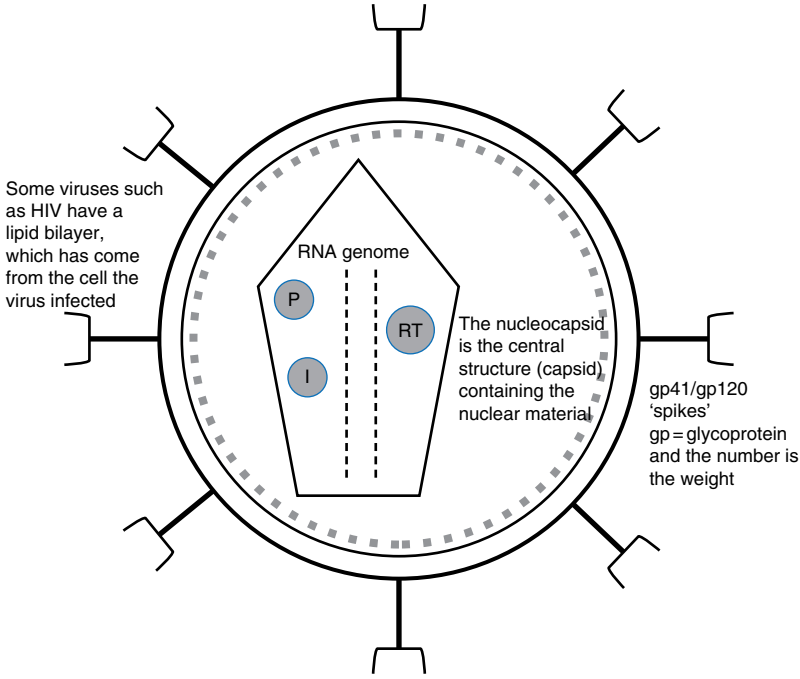
antimicrobials will allow it to thrive and multiply by binary fission in an environment where others would be unable to survive. This is the mechanism driving antimicrobial resistance. Genes conferring resistance to the β-lactamase group of antibiotics (e.g. penicillin, cephalosporins, carbapenems) are carried on plasmids. Many bacteria, mainly Gram-negative species, are able to undergo transformation. Conjugation occurs in Gram-negative and Gram-positive bacteria. *S. aureus* is able to acquire resistance to meticillin via a set of genes called MecA transferred by conjugation. It is also possible for plasmid transfer to take place between Gram-negative and Gram-positive species. Gram-negative resistance to carbapenem is plasmid mediated (Codjoe and Donkor 2017).

Mycoplasmas

Mycoplasmas are a genus of small bacteria that lack cell walls. As a result, they are naturally resistant to antimicrobial drugs that exert their effects by disrupting cell wall synthesis. Several species are human pathogens. *Mycoplasma pneumoniae* causes pneumonia; *M. genitalium* is sexually transmitted.

Viruses

Viruses are very small, typically 10–30 nm and visible only with the electron microscope. Each virus particle consists of a nucleic acid core surrounded by a protein coat called a *capsid* which carries receptors that enable the virus to attach



Viruses are much simpler than bacteria or human cells. This is HIV. It can infect CD4+ cells such as T-helper cells because of the fit between the GP41/GP120 on the outside of the virus and CD4 molecules on the cell.
Because of the unique replication of retroviruses such as HIV which involves making a DNA copy of its RNA genome, it has to carry its own enzyme to do this, known as reverse transcriptase (RT). Other important viral enzymes are integrase (I) and protease (P). Each of these is a target of anti-HIV treatment.

Figure 1.6 Structure of a typical virus.

to the host cell (Figure 1.6). Viral nucleic acid consists of either DNA or RNA but never both. Some viruses are described as 'enveloped' because they have an outer lipid (fat) capsule surrounding the protein coat. 'Naked' viruses lack a lipid capsule. The glycopeptide receptors enable the virus to attach to the host cell. Attachment is always at a specific site on the surface.

Viruses cause disease in humans, animals and plants. One group called *bacteriophages* (phages) infect bacteria. Viruses are obligate parasites. They cannot grow and replicate outside living cells and technically are not alive as they lack all the characteristics of living organisms (see Table 1.4) except the ability to reproduce and pass on genetic information to their progeny. There is ongoing debate between biologists and philosophers regarding whether it is correct to regard viruses as living organisms or whether they occupy a grey zone between the animate and inanimate. It is often assumed that because of their simple structure, they represent the

first, most primitive form of life but it is more likely that viruses are degenerate organisms that have lost their ability to exist independently during evolution.

Life Cycle of Viruses

An example of the life (or replication) cycle of a virus is shown below, in this case HIV. HIV is a retrovirus, meaning that its genetic material is in the form of RNA which has to be turned into DNA to allow the virus to replicate. Viruses attach themselves to the surface of the host cell at specific receptor sites and enter, leaving the protein coat outside (Figure 1.7). Receptor sites are specific for the virus, and

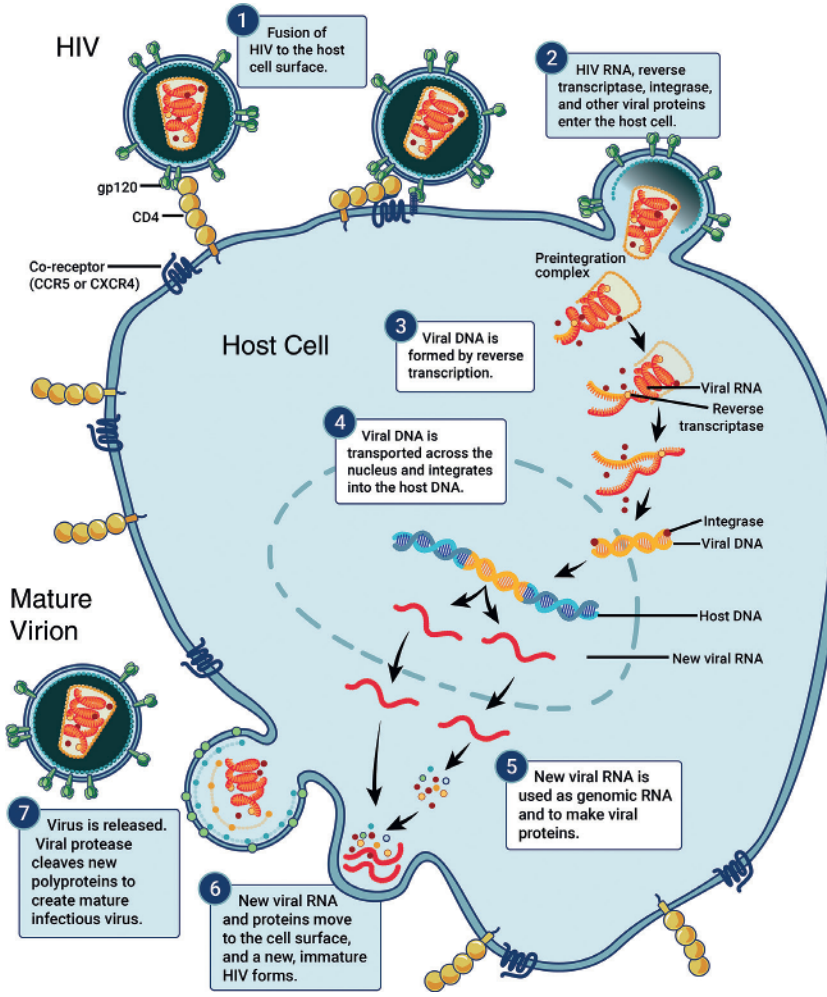


Figure 1.7 Life cycle of a DNA virus.

mean that viruses can only infect cells carrying the receptor that they recognise. They type of cell, tissue or species that a virus can infect is known as its tropism. In the case of HIV, the virus initially attaches itself to CD4 receptors on the surfaces of macrophages lymphocytes and some other cells. Having entered the cell, the viral RNA is converted to DNA by a virus specific enzyme known as reverse transcriptase, and becomes incorporated or integrated into the DNA of the host cell by another viral enzyme known as integrase. It then takes command of its genetic machinery, generating large numbers of new virus proteins. New virus particles are assembled and released into the environment. Most viruses destroy the host cell but as only a few are affected, the host organism recovers. There are exceptions, however. HIV eventually depletes the cells of the host immune system. Viruses able to cause malignancy (e.g. papilloma virus) are also highly destructive.

Treating virus infections with conventional antibiotics is impossible because they lack a cellular structure and are protected inside living cells.

Acute and Dormant Virus Infections

Some virus infections are acute, others cause dormant infections. When the infection is acute, new virus particles either bud out of the host cell membrane or cause it to burst (lyse) and release them. The host cell dies and the infection resolves quickly. Minor respiratory infections such as colds, influenza and varicella (chickenpox) are examples of acute virus infections. When the infection is dormant, the virus remains inactive inside the host cell for months or years. Reactivation occurs under specific circumstances, often when the host becomes unwell. The symptoms of viral infection manifest long after the initial infection. Herpes zoster (shingles) is an example of a dormant virus infection which typically occurs in older people, often if they develop another illness.

Classifying Viruses

Numerous different ways of classifying viruses have been devised over the years. Modern systems are based on the shape of the virus particle. Some of the major groups of clinical importance are presented below.

Adenoviruses

Adenoviruses are DNA viruses able to cause a wide range of common illnesses including colds, sore throats, influenza-like infections, conjunctivitis and gastrointestinal infections. There appears to be more than one mode of spread and portal of entry, including direct contact and the air-borne route. Infections occur most often in young children and those who have close contact with them. Infections are more serious in individuals who are immunosuppressed. The period of infectivity is prolonged. Risk factors include crowded conditions, especially indoors.

Herpesviruses

Herpesviruses are DNA viruses responsible for a number of different types of infection. Herpes simplex types 1 and 2 cause cold sores and genital infections. The varicella zoster virus causes chickenpox, which is an acute, usually mild infection affecting children. The virus can remain dormant in the dorsal horn cells of the nerves after the acute infection is over and become reactivated at a later stage, giving rise to herpes zoster (shingles). Vesicular lesions containing virus particles appear on the skin, mirroring the path of the nerve over its surface. The Epstein–Barr virus is responsible for glandular fever (infectious mononucleosis). *Cytomegalovirus* often results in asymptomatic infections but can cause respiratory or gastrointestinal symptoms.

Caliciviruses

Caliciviruses are RNA viruses responsible for gastroenteritis. The most notorious is norovirus which causes 'winter vomiting'. Outbreaks are very common in community and hospital settings and are very difficult to control.

Other clinically important RNA viruses are shown in Table 1.6.

Prions

Prions are protein particles that do not contain genetic material. They are responsible for a number of serious neurological diseases (Ironside et al. 2017). These conditions have a high public profile but are rare, occasionally causing sporadic infections in the community (Haywood 1997). Examples are Creutzfeld–Jakob disease, bovine spongiform encephalopathy, fatal familial insomnia and kuru. Infection is thought to occur following changes in prion structure. Iatrogenic transmission appears possible, for example during transplant surgery, blood transfusion or if surgical instruments become contaminated with neurological tissue, cerebrospinal fluid or blood.

Prions are resistant to disinfection and sterilization. Special approaches must be taken to contain risks in operating theatres.

Table 1.6 RNA viruses.

Piconaviruses, e.g. rhinovirus, coxsackie virus, hepatitis A
Togavirus, e.g. yellow fever, dengue fever, hepatitis C
Rubella
Orthomyxovirus, e.g. influenza
Calicivirus, e.g. norovirus
Coronavirus, e.g. colds, COVID, SARS
Retrovirus, e.g. HIV, leukaemia viruses

Fungal Infections

Fungi are eukaryotic organisms exhibiting all the characteristics of living organisms (see Table 1.2). They form huge quantities of minute spores spread via the air. Fungi are classified separately from other organisms but have most in common with animals. Their cellular organelles are similar to those of animals and they contain the storage carbohydrate glycogen also found in animals. Over 300 000 species exist and there are undoubtedly many more to be discovered and classified. Most are harmless saprophytes living in soil and water and some are commercially important in cheese and bread making. About 600 species have been associated with human disease and some species are opportunistic (e.g. *Candida* spp., *Aspergillus* spp.) (Caceres et al. 2020). Their similarity to animals makes developing antifungals challenging.

Structurally, fungi fall into two groups: simple forms (yeasts), that reproduce asexually by forming buds, and those with a more complex structure. Complex fungi are made up of microscopic, filamentous *hyphae* which absorb nutrients and water. Collectively hyphae are called a *mycelium*. The visible fruiting bodies of mushrooms and toadstools are large collections of hyphae visible to the naked eye. Fungi are highly invasive because the finger-like hyphae enable them to grow between cells and into organs.

Fungal infections are called mycoses. A *mycosis* can be superficial or systemic. Superficial mycoses are restricted to subcutaneous tissues that contain the protein keratin (the skin and its appendages [nails, hair]) and to the mucous membranes. The deeper tissues are not invaded and if a host response occurs, it is only mild. Nevertheless, superficial mycoses are distressing and because they are often visible to other people, can be a source of embarrassment and lead to stigmatisation. Athlete's foot (*Trichophyton interdigitale*) and ringworm (*Tinea capitis*) are examples of superficial mycoses affecting the skin and appendages. *Candida albicans* is a superficial mycosis responsible for 'thrush' affecting the mucous membranes lining the oral cavity and vagina. Superficial mycoses are usually amenable to topical treatment with creams, ointments and pessaries.

Systemic mycoses affect the internal tissues (Box 1.7). The portal of entry is either via the respiratory or gastrointestinal tract or the skin. The host is usually immunocompromised and the outcome of infection is often a severe illness (Hope et al. 2013). *Aspergillosis fumigatus*, *Candida auris* and *Rhizopus* spp. can give rise to systemic mycoses. *Pneumocystis* spp. are opportunistic but can cause severe lung disease in immunocompromised patients. This mycosis first gained attention during the HIV pandemic in the 1980s when many people became severely ill.

The management of systemic fungal infections is challenging because patients are usually already very ill and few effective antifungal agents are available to treat them. Their development is encouraged by warm, humid conditions. At one time, systemic mycoses were uncommon in temperate climates but the *incidence* is now increasing, probably as an effect of global warming. Advancements in medical

Box 1.7 Invasive Fungal Disease

Invasive fungal disease is an important cause of morbidity and mortality amongst critically ill and immunocompromised patients. Construction and renovation activities can cause extensive contamination of the healthcare environment, resulting in clusters of infection. The main culprit is *Aspergillus* spp. Recommendations for decreasing risk include minimising dust likely to be contaminated by spores generated during construction work and preventing dust infiltrating clinical areas by using high-efficiency filtration systems near patients at high risk.

Source: Adpated from Kanamori et al. (2015).

science have increased the number of severely immunocompromised patients and this is also considered an important contributory factor to the increasing incidence of invasive fungal disease (de Cássia Orlandi Sardi et al. 2018).

Protozoa

Protozoa are unicellular microscopic animals. Many species exist but most live harmlessly in water and soil. Under adverse conditions, they form thick-walled cysts which can survive for long periods until a return to more favourable conditions. A few species are pathogenic (Table 1.7).

Malaria is a protozoal infection responsible for a major public health problem globally (Box 1.8).

Chlamydiae

Chlamydiae combine the characteristics of bacteria and viruses but can be treated with antibiotics. All are obligate intracellular organisms. Many species exist and a few are important human pathogens responsible for serious public health problems. *C. trachomatis* is the leading cause of blindness in low-income countries. In higher income countries, it is most commonly spread through vaginal, oral and anal sex. Men develop non-specific urethritis. Women develop *salpingitis* which is a leading cause of subfertility. Neonates can develop pneumonia or serious eye infections during passage down an infected birth canal.

Table 1.7 Pathogenic protozoa.

Plasmodium spp. (malaria): biological transmission by insect vector
Trichomonas vaginalis: sexual transmission
Giardia lamblia: food-borne and water-borne transmission
Entamoeba histolytica (amoebic dysentery): food-borne and water-borne transmission
Toxoplasma gondii: transmission in undercooked meat, cat faeces, vertical transmission in the human host

Box 1.8 Malaria

According to the World Health Organization (WHO), there are 229 million cases of malaria globally. In 2019, 409 000 deaths were reported, of which nearly 70% were in children. Nearly half the global population is at risk and the most affected nation is Africa. Symptoms vary. Mild infections are hard to diagnose. Severe infections cause acute febrile illness with fever, chills and malaise. The parasites (plasmodia) live and multiple inside erythrocytes and symptoms recur when they burst, releasing the plasmodia into the bloodstream. Long-term effects include anaemia, respiratory distress resulting from metabolic disturbance and cerebral malaria. Developing effective vaccines has been challenging because the plasmodia are protected inside the erythrocytes but the discovery of an effective vaccine was announced by Oxford University in 2021. Malaria is also prevented by public health measures to control the vector: insecticides, use of antimalarial prophylaxis and drainage of swamps where the mosquitoes live.

Numerous drugs are used in malarial chemoprophylaxis, but resistance is widely reported so travellers should be advised to seek the latest information from travel centres. It may be necessary to take several drugs in combination to reduce the possibility of the plasmodia developing resistance. Drug regimens differ between regions according to the levels of risk and existing drug resistance.

Parasites

There are two broad group of parasites: endoparasites which live inside the body and ectoparasites which live on the surface.

Endoparasites

Helminthic (Worm) Infestations

Numerous species can infest humans. Some are microscopic. Others are large and clearly visible to the naked eye but the eggs and early larval stages are visible only under the microscope and specimens are sent to the medical microbiology department for diagnosis. Identification is usually by examining specimens of recently passed faeces ('hot' stools) because any microscopic forms they contain are likely to be alive and active, increasing the chances of diagnosis. There are two groups of helminths: roundworms and flatworms (Table 1.8).

Some helminthic infestations are minor and a nuisance. Others constitute a major public health challenge.

Enterobius vermicularis (threadworm) is the most common helminth in the UK. It usually affects children. The worms infest the large bowel and emerge onto the perineum at night where they lay their eggs. Many people are unaware that they have threadworms but perianal and vaginal itching are common. Scratching followed by swallowing eggs contaminating the fingers perpetuates reinfestation.

Table 1.8 Helminths.

Roundworms
Enterobius vermicularis (threadworm)
Ascaris lumbricoides (roundworn)
Necator spp. (hookworm)
Strongyloides spp.
Flatworms
Taenia spp. (tapeworm)

Adults are less likely to be affected than children because the adult gastric secretions are more acidic and destroy the eggs more effectively. The eggs can survive on surfaces for up to two weeks and are readily transferred to bedclothes, flannels, towels, toys and kitchen utensils. Transmission occurs easily between members of the same household and all must receive treatment.

Mebendazole is the most commonly used medication and is available over the counter. It kills the adult worms within a few days but does not destroy the eggs. Stringent hygiene measures are necessary to eradicate the infestation: washing bedlinen, towels and soft toys, thoroughly vacuum cleaning the house, especially bedrooms, and not eating in bedrooms to avoid swallowing eggs shaken off bedclothes. Thorough hand hygiene and scrubbing nails should be encouraged. Nail biting and thumb sucking should be discouraged. Precautions should continue for six weeks as this is the life cycle of *E. vermicularis*.

Other species of helminths are a leading cause of chronic ill health in warm, moist climates where sanitation is poor. The worms inhabit the intestine, the eggs escape in faeces and are transmitted to other people on contaminated fingers and food. Infestation is very common, especially in children. Light infestations may have few ill effects. More severe infestations interfere with the absorption of food, contributing to undernutrition and anaemia. Helminthic infestation is a major and underappreciated cause of poor educational attainment and economic underdevelopment globally but is preventable.

Ectoparasites

Ectoparasites live outside the host, either on or in the skin (Table 1.9).

Outbreaks of scabies are often reported in hospitals and nursing homes. Infestation is caused by mites.

Scabies is caused by the mite *Sarcoptes scabiei* which is transmitted directly and indirectly by skin contact. The mite burrows into the skin and lays eggs, giving rise to intense itching and a papular rash. Symptoms appear in 4–8 weeks. Scabies causes intense itching which may persist for some time after treatment, can lead to skin infections and can make existing skin conditions (e.g. psoriasis) worse.

Table 1.9 Ectoparasites: examples.

Sarcoptes scabiei	Scabies
Cimex lectularius	Bed bug
Pediculus humanus capitis	Head louse
Pediculus humanus humanus	Body louse
Phthirius pubis	Pubic louse

Transmission requires direct, prolonged skin-to-skin contact, contact on clothing, bedclothing or furnishings. Scabies can spread rapidly in crowded conditions, especially as asymptomatic individuals can be a source of infestation. Eradication involves washing all clothing and bedclothing at 50 °C or higher. Items that cannot be washed should be placed in a bag which should be sealed for three days while the mites perish. Treatment is with 5% permethrin cream or 0.5 malathion lotion available over-the-counter.

'Crusted' (Norwegian) scabies results when large numbers of mites are present and is particularly likely to result in outbreaks in hospitals, nursing homes and prisons which may be hard to control (Vorou et al. 2007). Treatment with ivermectin can be effective.

Head Lice

Head lice (*Pediculus humanus capitis*) are very common. Children and sometimes adults are affected. Transmission occurs when two heads touch and the lice are able to jump. They prefer clean hair because it enables them to move around easily and as they like warmth, they tend to congregate close to the scalp. Lice are difficult to dislodge and cannot be removed by ordinary shampooing. Adult females lay large numbers of eggs close to the base of the hair shafts. The usual symptom, itching, may not develop for several weeks. Lice can be removed by wet combing. Chemical insecticides are not always effective and resistance is developing.

Pubic Lice

Pubic lice (*Phthirius pubis*) infest the coarse body hair and are spread by close contact, including between family members as well as during sexual contact. They die rapidly in the environment and are not spread by clothing or other objects. Treatment is with insecticides, either topically (e.g. malathion) or orally (ivermectin).

Suggested Activities

Exercise 1.1 Self-assessment

1 Complete the following sentence: 'A microbial pathogen is defined as . . .'

2 A reservoir of infection is:
- **A** Any source of infection inside the patient zone
- **B** Where the pathogen survives but cannot multiply
- **C** Where the pathogen lives and multiplies
- **D** Any source of infection outside the patient zone.

3 List seven ways in which micro-organisms spread.

4 The infectious dose is:
- **A** The number of organisms required for colonisation to occur
- **B** The number of organisms required for infection to occur
- **C** The number of organisms required for cross-infection to occur
- **D** The number of organisms required for cross-contamination to occur.

5 Under favourable circumstances, most bacteria are able to cause infection. True/False

6 Explain the key difference between endogenous infection and exogenous infection.

7 DNA viruses reproduce by binary fission.
True/False

8 Which of the following cause mycoses?
- **A** *T. pallidum*
- **B** *Pneumocystis* spp.
- **C** *Aspergillus fumigatus*
- **D** All of the above

9 Which of the following are pathogenic protozoa?
- **A** *Plasmodium* spp., *Trichomonas vaginalis*, *Entamoeba histolytica*, *T. pallidum*
- **B** *Plasmodium* spp., *M. genitalium*, *E. histolytica*, *Toxoplasma gondii*
- **C** *Plasmodium* spp., *Trichomonas vaginalis*, *E. histolytica*, *Toxoplasma gondii*
- **D** All of the above

10 The commensal flora is always harmless.
True/False

Exercise 1.2

Choose a pathogen that has caused problems/has the potential to cause problems in your own clinical setting.

- What are the problems/potential problems?
- Apply the chain of infection to suggest solutions.
- Identify lessons for future clinical practice, policy and education in your organisation.

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