

Some pathogenic organisms damage humans by producing poisonous substances called toxins. *Toxins*, many of which are proteins, exert their effects by several methods:

1. damage to cell membranes,
2. disruption of various intracellular functions, and
3. inhibition of function at nerve cell synapses.

Toxins that act directly on cell membranes, called cytolytic toxins, disturb and ultimately kill the target cells. Produced by many organisms (e.g., bacteria, fungi, plants, fish, and snakes), cytolytic toxins may cause damage in several ways. For example, streptolysin O (67,000 D), produced by the bacterium *Streptococcus pyogenes*, causes pores to form in the target cell membranes. Affected cells are rapidly lysed because the cell membrane is much more permeable to ions such as Na^+ . Streptolysin O is believed to cause some of the damage in rheumatic fever.

Cell membrane destruction may also be caused by toxic enzymes. For example, many organisms secrete enzymes, called phospholipases, which cause the hydrolysis of membrane lipid molecules. Phospholipase A2 is found in the venom of several snakes.

Many toxins interfere with intracellular functions. The best-characterized of these are diphtheria toxin and cholera toxin, produced by the bacteria *Corynebacterium diphtheriae* and *Vibrio cholerae*, respectively. Both of these toxins contain two subunits, called A and B. The A subunit is responsible for the toxic effect, whereas the B subunit binds to the target cell. Once diphtheria toxin has entered the target cell, the A and B subunits split apart. The A subunit, which is an enzyme, catalyzes a reaction that prevents protein synthesis. The cell dies because it cannot synthesize proteins. The host organisms dies because cardiac, kidney, and nervous tissue are destroyed.

The B subunit of cholera toxin, which is made up of five identical subunits (Figure 5A), attaches to the membranes of intestinal cells. The A subunit is then inserted into these cells, where it activates an enzyme that increases the concentration of a nucleotide called cyclic AMP (cAMP). High sustained concentrations of cAMP, a molecule that opens membrane chloride channels, causes severe diarrhea. (Loss of chloride results in water loss because of osmotic pressure. Several gallons of fluid per day may be lost.) If left untreated, severe dehydration may cause death within 48 hours, by circulatory shock brought on by low blood volume.

Several toxic proteins act as neurotoxins by disrupting the activity of synapses. (A synapse is a junction between two neurons or between a neuron and a muscle cell.) The pain, tremors, and irritability that result from black widow spider bites are caused by α -latrotoxin (125,000 D). This molecule, a single polypeptide, stimulates a massive release of the neurotransmitter acetylcholine (ACh). In contrast, ACh release is inhibited by botulinum toxin, a mixture of several proteins produced by the bacterium *Clostridium botulinum*. Botulism, a malady most commonly caused by eating contaminated canned food, is characterized by vomiting, dizziness, and sometimes paralysis and death. A related species,

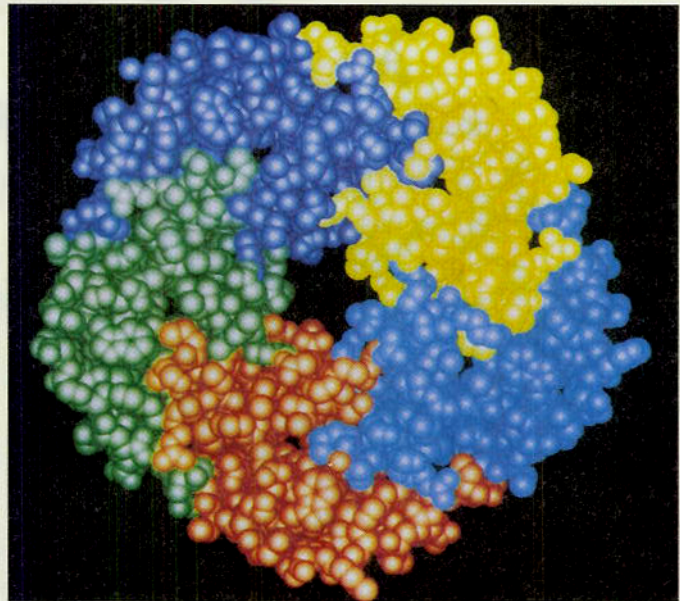


FIGURE 5A

The B-Subunit of the Cholera Toxin.

Clostridium tetani, produces another deadly neurotoxin. Tetanus toxin causes severe paralysis by blocking neurotransmitter release (primarily glycine and γ -aminobutyric acid) in the central nervous system.

Cholera: A Short History

Toxin-producing organisms, such as *V. cholerae*, do not kill only individual humans. Under certain circumstances they can affect an entire civilization. Cholera has had lasting effects on the Western world. Because of improved transportation, the 1817 cholera epidemic in India reached Europe. Traveling at an average speed of five miles a day, the disease left millions dead. Cholera claimed its first British victim in the port city of Sunderland in October 1831. The 22,000 deaths that followed during the next 2 years were due largely to horrendous living conditions during the Industrial Revolution (Figure 5B). Despite intense but misdirected efforts, cholera epidemics often occurred during the decades that followed. How the disease spread was not discovered until the newly emerging science of statistics revealed that poor sanitation and polluted water were responsible. Public pressure, driven by the seemingly endless deaths caused by cholera, eventually forced the British government to assume some responsibility for public health, a relatively modern concept. In 1859 the British Parliament contracted to build an elaborate sewer system in the city of London, at that time the largest municipal project of its kind ever undertaken. Cholera never returned to that city, and the relationship between sanitation and infectious disease was firmly established. In 1883 the German researcher Robert Koch identified the causative agent.

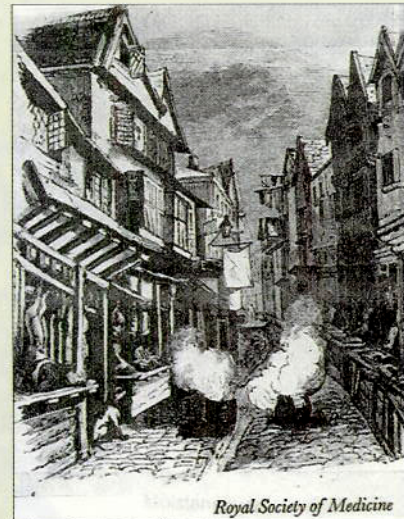
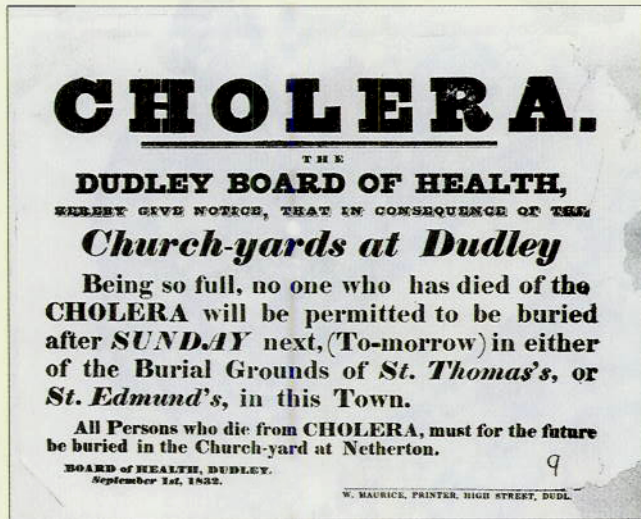


FIGURE 5B

Devastating Effect of Cholera on Britain in the Mid-Nineteenth Century.

(a) A poster attests to the severity of the epidemic. (b) Burning tar to kill infection in the air in Exeter. Other “methods” for preventing the spread of cholera included tobacco fumes, and cleaning with turpentine vinegar.

Bioterrorism and Anthrax

Bioterrorism is the use of bacteria, viruses, or toxins to intimidate or coerce human populations. Bioterrorists are criminals who seek to achieve political goals that are unobtainable by legitimate, non-violent means. In its latest incarnation, bioterrorism has taken the form of anthrax-contaminated letters sent to prominent individuals in government and the media in the weeks after the World Trade Center disaster. Anthrax is a disease, primarily affecting livestock (sheep, cattle, and horses), that is caused by the Gram-positive bacterium *Bacillus anthracis*. Known since biblical times (it is believed to be one of the ten plagues mentioned in the Book of Exodus, Chapter 9), anthrax has played an important role in the history of medicine and microbiology. In 1876 Robert Koch demonstrated that *B. anthracis* is the causative agent of anthrax. The techniques he developed while investigating anthrax and later tuberculosis led to the development of Koch’s postulates, the method still currently used to identify the causes of newly discovered infectious diseases. Louis Pasteur developed the first artificial vaccine against the disease, and in 1881, he conclusively demonstrated to a skeptical scientific community that vaccinated sheep and cattle could withstand the otherwise fatal injection of the live bacterium.

Anthrax is initiated by exposure to heat-resistant endospores of *B. anthracis* (a dormant form of the organism) that can exist in soil or animal products for decades. The spores enter the body through skin abrasions (cutaneous anthrax), the lungs (inhalation anthrax), or the ingestion of contaminated food (gastrointestinal anthrax). After inhalation of the endospores (the most deadly form of the disease) they are absorbed by macrophages, immune sys-

tem cells that ordinarily ingest and destroy invading bacteria and other foreign material. The macrophages, however, are unable to destroy the endospores because their capsular coating is made of an indigestible polymer composed of D-glutamic acid residues. Instead the endospores germinate into vegetative (actively dividing), disease-causing bacteria that divide until the macrophage bursts. If the exposure to the endospores is sufficient, the rapidly dividing bacteria can overwhelm the immune system and spread throughout the body. Systemic anthrax causes, within days after the first flulike symptoms appear, severe hypotension (low blood pressure), shock, and (in some cases) meningitis. The organism’s capacity to inflict such massive damage is made possible by three toxins, which together inactivate critical immune defenses, break into cells, and disrupt normal signaling mechanisms. Once the bacterial cells are released into the blood, they secrete their toxins. Protective antigen (PA), named before its role was discovered, binds to cell-surface receptors. Once on the cell surface, seven PA toxin molecules undergo proteolytic activation and assemble into a doughnut-shaped structure. This complex then binds the toxic enzymes lethal factor (LF) and edema factor (EF) and inserts them into the cell in an endocytosis-like process. LF, the principal cause of death, is a zinc-dependent protease (an enzyme that breaks peptide bonds in proteins) that disrupts the intricate intracellular signaling system. Its most damaging effect is to cause macrophages to release massive amounts of inflammatory molecules that induce shock. EF causes massive swelling (edema) in affected tissues. If the infection is not arrested by antibiotics such as penicillin, the combined effects of these toxins cause death.