

Cholera: Mechanism of Infection, History and Treatment

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Abstract

The bacterium *Vibrio cholerae* causes the diarrheal disease known as cholera. This gram-negative bacterium produces an endotoxin which locks a G protein into the configuration promoting constant cAMP formation. Cyclic AMP elevation causes inhibition of sodium and chloride into intestinal cells thereby causing them to release water, resulting in the massive diarrhea that characterizes the illness. Cholera symptoms vary with the percentage of body fluid loss. Treating cholera mainly involves replacing the fluids lost to diarrhea. The illness spreads via the fecal-oral route and can be passed by ingesting either contaminated food or water. Vaccines are available, but travelers from developed countries are said to be at low risk of contracting the disease.

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The term cholera designates the severe diarrheal disease caused by the bacterium *Vibrio cholerae*. The bacterium's name originates from the Greek words meaning "flow of bile." *V. cholerae* is a gram-negative flagellated bacillus that colonizes the intestinal lumen. The bacteria do not invade the intestinal epithelia. Instead, the endotoxin they produce, designated cholera toxin (CT), invades these cells and causes the disastrous symptoms. Cholera spreads via the fecal-oral route and proves to have more devastating consequences in areas lacking proper sanitation of water and human waste.

Italian scientist Filippo Pacini first described cholera during an 1854

outbreak in Florence. He performed autopsies of cholera patients and microscopically examined histological sections of the intestinal mucosa. He discovered a comma-shaped bacillus and published a paper the same year entitled "Microscopical observations and pathological deductions on cholera." Pacini supported the germ theory and insisted that cholera was contagious. Until the 1965 acknowledgement of Pacini's earlier discovery by the international committee on nomenclature, Robert Koch had been credited with discovering the bacterium that caused cholera. During the 1883 epidemic in Egypt, he discovered a bacillus present in the intestinal lumen of

patients who had died of cholera but not other causes. He followed the epidemic to India, and in 1884 finally isolated the bacillus in pure culture. He, like Pacini, noted the bent shape of the bacillus.¹

Also important to the elucidation of cholera's causative organism is the work of John Snow. At medical conferences in 1849 and 1853 he had argued that diseases such as cholera spread via drinking water rather than air. His ideas received few supporters. During the 1854 cholera epidemic in London, however, he probed his water-borne theory further. Through methodical epidemiological investigation, Snow eventually determined the primary source of contamination to be the Broad Street pump. Upon the removal of its handle, the number of new cases of cholera fell dramatically, and then disappeared. Investigation of the pump revealed that a sewer pipe passed within a few feet of the well. The pipe had gradually leaked its raw sewage into the drinking water supply.²

Vibrio cholerae can be subdivided into serogroups based upon antigens on its somatic surface. The serogroups O1 and O139 comprise the pathogenic strains. The O1 serogroup can be further divided into three serotypes according to somatic antigens and then into two biotypes according to specific phenotypes. Ogawa (somatic antigens A and C), Inaba (A and B), and Hikojima (A, B and C) designate the serotypes. El Tor and classic designate the biotypes. The O1 El Tor biotype predominates in most current infections.³ Serogroups O1 and O139 prove pathogenic due to their production of an enterotoxin that promotes the secretion of fluids and electrolytes into the intestinal lumen, resulting in diarrhea once the amount

overcomes the body's resorptive mechanisms.

This cholera toxin contains two A subunits and five B subunits. The B subunits allow binding to a ganglioside (GM₁) receptor on the intestinal epithelial cells. The B pentamer must bind to five corresponding GM₁ receptors. This binding occurs on cell surface areas termed lipid rafts, due to concentration of specific lipids. Membrane cholesterol is critical to these interactions and endocytosis of the A subunits. In an experiment removing cholesterol from epithelial membranes, researchers found that its presence facilitates the CT-GM₁ complex formation and endocytosis.⁴ Another experiment proved that the lipid rafts help anchor the toxin to the membrane for endocytosis of the A subunits. They help traffic the toxin into the cell, and to the basolateral surface, where it acts.⁵ Once internalized, the A subunits proteolytically cleave into A1 and A2 peptides. The A1 peptide ADP-ribosylates a GTP-binding protein, thereby preventing its inactivation. The always active G protein causes adenylate cyclase to continue forming cAMP. This increase in intracellular cAMP blocks absorption of sodium and chloride by microvilli and promotes the secretion of water from the intestinal crypt cells to preserve osmotic balance.⁶ This water secretion causes the watery diarrhea with electrolyte concentrations isotonic to plasma. The fluid loss occurs in the duodenum and upper jejunum, with the ileum less affected. The colon is less sensitive to the toxin, and is therefore still able to absorb some fluid. The large volume, however, overwhelms the colon's absorptive capacity.⁷ Symptoms of cholera begin following a 24- to 48-hour incubation period.

Patients experience a sudden onset of high volume watery diarrhea that is often followed by vomiting. Abdominal cramps may accompany these symptoms. The diarrhea has a characteristic “rice water” appearance. Its volume in severe manifestations of the disease can exceed 250 mL/kg in the first twenty-four hours. Adults typically present as afebrile, while children may have a fever. Other clinical signs correspond to the patient’s degree of dehydration on presentation. With a fluid loss of 3-5% of the body weight, the patient complains of excessive thirst. With a 5-8% loss, the patient experiences hypotension, tachycardia, weakness, fatigue, and dry mucous membranes. With greater than a 10% loss, the patient’s urine output decreases significantly; the eyes appear glassy and sunken; fontanelles in infants appear sunken; the pulse becomes weak, thready or absent; the skin appears wrinkled; the patient seems excessively sleepy or may lapse into a coma.⁵

Treatment of cholera proves fairly simple: patients must be rehydrated to avoid hypovolemic shock and death. This process occurs in two phases, primary rehydration and maintenance. Patients who can tolerate oral rehydration solution (ORS) should receive it, while severely dehydrated patients can receive Lactated Ringer’s solution or saline IV. Achieving normal hydration status should take less than four hours, with the rate for severely dehydrated patients approaching 50-100 mL/kg/hour. Upon attainment of normal hydration status, the patient must continue to receive oral ORS or IV fluid therapy until the diarrhea abates. Treatment guidelines recommend a rate in adults of 500-1000 mL/hour of oral rehydration solution to maintain normal

hydration status. Antibiotics can be useful in treatment of the infection, but they are not curative. Tetracycline, ciprofloxacin and erythromycin can shorten the duration and volume of fluid loss, but the disease still must run its course. Drug resistance becomes a concern in areas with endemic cholera.⁵ Treatment with appropriate fluids saves lives of patients, and with no treatment 25-50% of typical cases are fatal.⁸

Cholera infection requires a relatively high infectious dose of the bacteria. In a water vehicle, the infectious dose is 10^3 to 10^6 organisms, while a food vehicle requires only 10^2 to 10^4 organisms. This high dose results from the bacteria’s susceptibility to the acidity of the stomach. The risk of cholera infection increases in those persons who use antacids, histamine receptor blockers, and proton pump inhibitors as these drugs decrease gastric acidity. The same risk applies to patients with chronic gastritis due to *H. pylori* infection or those who have had a gastrectomy. Persons with type O blood also have an increased risk of infection, and this susceptibility is not fully understood.⁵

Equally important to the treatment of cholera is the prevention of future infections. Since cholera spreads via the fecal-oral route, creating or maintaining a potable water supply is critical during an epidemic. Boiling and chlorination of water can help kill the bacteria in water supplies. Proper disposal and/or sterilization of infectious waste, proper hygiene in food preparation, and cooking foods to high enough temperatures also help to attenuate the epidemic. Infection with the O1 serogroup seems to convey immunity against future infection with the same strain of bacteria, however this does not hold true for the O139 serogroup. Vaccines can help limit

epidemics in endemic areas where poor sanitation seems difficult to overcome. The oral route of vaccination seems most effective. The WC/rBS vaccine consists of killed, whole cell *V. cholerae* as well as the purified recombinant B subunit of the cholera toxin. This particular vaccine offers less protection against the El Tor biotype. Another oral vaccine, CVD 103-HgR, uses live, attenuated cholera bacteria. So far, this vaccine seems to be well tolerated and extremely immunogenic. New vaccines are being tested against El Tor and O139. Vaccination is no longer required for international travelers, as the risk of an international traveler from a developed country contracting cholera is so small (1 case in 500,000 travelers).^{5,9}

Cholera causes disease in many third world countries lacking proper sanitation methods, but it is relatively easy to treat. Nonetheless, further investigation into vaccine possibilities would no doubt aid in preventing these infections. Further understanding of the role of lipid rafts in the mechanism of disease may one day help in development of drugs or vaccines. The more pressing issue, however, is adequate funding and manpower to control the present cholera pandemics in Asia and Central America.

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