support of this recommendation, the WHO and international health authorities established an international cholera vaccine stockpile in 2013.[128] By 2017, more than 12 million doses of oral cholera vaccine had been distributed from the stockpile as part of 46 deployments. The number of cholera vaccines shipped each year has roughly doubled since inception of the stockpile.[129] In 2017, a global strategy on cholera control with a target to reduce cholera deaths by 90% by 2030 was launched by the Global Task Force on Cholera Control.[129a]

In 2016 a live attenuated oral cholera vaccine was approved by the US Food and Drug Administration for use in adults in the United States (Vaxchora [CVD 103 HgR]; PaxVax, San Diego, CA).[130] This vaccine provided 90% protective efficacy against experimental challenge in North American adult volunteers within 10 days of vaccination and 80% protective efficacy against challenge at 90 days.[131] The duration of protection and vaccine efficacy in endemic zone populations is currently unknown. Additional live attenuated oral cholera vaccines are in various stages of development and analysis including Peru-15 (Haikou VTI Biological Institute, China), HaitiV,[131a] and several others.[132–138,139,140,141] A number of other cholera vaccines including subunit and conjugate vaccines are in various stages of development.[142]

Key References

The complete reference list is available online at Expert Consult.

1. Pollitzer R, Swaroop S, Burrows W. Cholera. *Monogr Ser World Health Organ*. 1959;58:1001–1019.
7. Devault AM, Golding GB, Waglechner N, et al. Second-pandemic strain of *Vibrio cholerae* from the Philadelphia cholera outbreak of 1849. *N Engl J Med*. 2014;370:334–340.
8. Heidelberg JF, Eisen JA, Nelson WC, et al. DNA sequence of both chromosomes of the cholera pathogen *Vibrio cholerae*. *Nature*. 2000;406:477–483.
9. Egan ES, Waldor MK. Distinct replication requirements for the two *Vibrio cholerae* chromosomes. *Cell*. 2003;114:521–530.
11. Colwell RR. Global climate and infectious disease: the cholera paradigm. *Science*. 1996;274:2025–2031.
14. Taylor RK, Miller VL, Furlong DB, et al. Use of phoA gene fusions to identify a pilus colonization factor coordinately regulated with cholera toxin. *Proc Natl Acad Sci USA*. 1987;84:2833–2837.
24. Waldor MK, Mekalanos JJ. Lysogenic conversion by a filamentous phage encoding cholera toxin. *Science*. 1996;272:1910–1914.
26. Lee SH, Hava DL, Waldor MK, et al. Regulation and temporal expression patterns of *Vibrio cholerae* virulence genes during infection. *Cell*. 1999;99:625–634.
28. Merrell DS, Butler SM, Qadri F, et al. Host-induced epidemic spread of the cholera bacterium. *Nature*. 2002;417:642–645.
30. Mutreja A, Kim DW, Thomson NR, et al. Evidence for several waves of global transmission in the seventh cholera pandemic. *Nature*. 2011;477:462–465.
31. Bik EM, Bunschoten AE, Gouw RD, et al. Genesis of the novel epidemic *Vibrio cholerae* O139 strain: evidence for horizontal transfer of genes involved in polysaccharide synthesis. *EMBO J*. 1995;14:209–216.
34. Chin CS, Sorenson J, Harris JB, et al. The origin of the Haitian cholera outbreak strain. *N Engl J Med*. 2011;364:33–42.
35. Piarroux R, Barrais R, Faucher B, et al. Understanding the cholera epidemic. *Haiti Emerg Infect Dis*. 2011;17:1161–1168.
39a. World Health Organization. Weekly epidemiological record: cholera, 2017. No. 38. 2018;93:489–500. http://www.who.int/wer/2018/wer9338/en/. Accessed April 19, 2019.
45. Deen JL, Von SL, Sur D, et al. The high burden of cholera in children: comparison of incidence from endemic areas in Asia and Africa. *PLoS Negl Trop Dis*. 2008;2:e173.
46. Harris JB, Khan AI, Larocque RC, et al. Blood group, immunity, and risk of infection with *Vibrio cholerae* in an area of endemicity. *Infect Immun*. 2005;73:7422–7427.
49. Harris JB, Larocque RC, Chowdhury F, et al. Susceptibility to *Vibrio cholerae* infection in a cohort of household contacts of patients with cholera in Bangladesh. *PLoS Negl Trop Dis*. 2008;2:e221.
50. Chowdhury F, Khan AI, Harris JB, et al. A comparison of clinical and immunologic features in children and older patients hospitalized with severe cholera in Bangladesh. *Pediatr Infect Dis J*. 2008;27:986–992.
53. Larocque RC, Sabeti P, Duggal P, et al. A variant in long palate, lung and nasal epithelium clone 1 is associated with cholera in a Bangladeshi population. *Genes Immun*. 2009;10:267–272.
56. World Health Organization. The Treatment of Diarrhoea, a Manual for Physicians and Other Senior Health Workers. 4th revision. 2005. http://whqlibdoc.who.int/publications/2005/9241593180.pdf. 2005.
58. Qadri F, Wenneras C, Albert MJ, et al. Comparison of immune responses in patients infected with *Vibrio cholerae* O139 and O1. *Infect Immun*. 1997;65:3571–3576.
59. Benenson AS, Islam MR, Greenough WB 3rd. Rapid identification of *Vibrio cholerae* by darkfield microscopy. *Bull World Health Organ*. 1964;30:827–831.
62. Mahalanabis D, Wallace CK, Kallen RJ, et al. Water and electrolyte losses due to cholera in infants and small children: a recovery balance study. *Pediatrics*. 1970;45:374–385.
65. Gill DM, Meren R. ADP-ribosylation of membrane proteins catalyzed by cholera toxin: basis of the activation of adenylate cyclase. *Proc Natl Acad Sci USA*. 1978;75:3050–3054.
67. Ramakrishna BS, Venkataraman S, Srinivasan P, et al. Amylase-resistant starch plus oral rehydration solution for cholera. *N Engl J Med*. 2000;342:308–313.
68. Global Task Force on Cholera Control. *First Steps for Managing an Outbreak of Acute Diarrhoea*. Geneva: World Health Organization; 2010.
74. Roy SK, Hossain MJ, Khatun W, et al. Zinc supplementation in children with cholera in Bangladesh: randomised controlled trial. *BMJ*. 2008;336:266–268.
77. Cholera Outbreak Training and Shigellosis Program (COTS Program). 2011. http://www.cotsprogram.com.
78. Comstock LE, Maneval D Jr, Panigrahi P, et al. The capsule and O antigen in *Vibrio cholerae* O139 Bengal are associated with a genetic region not present in *Vibrio cholerae* O1. *Infect Immun*. 1995;63:317–323.
79. Glass RI, Svennerholm AM, Khan MR, et al. Seroepidemiological studies of El Tor cholera in Bangladesh: association of serum antibody levels with protection. *J Infect Dis*. 1985;151:236–242.
80. Johnson RA, Uddin T, Aktar A, et al. Comparison of immune responses to the O-specific polysaccharide and lipopolysaccharide of *Vibrio cholerae* O1 in Bangladeshi adult patients with cholera. *Clin Vaccine Immunol*. 2012;19:1712–1721.
82. Pasetti MF, Levine MM. Insights from natural infection-derived immunity to cholera instruct vaccine efforts. *Clin Vaccine Immunol*. 2012;19:1707–1711.
84. Patel SM, Rahman MA, Mohasin M, et al. Memory B cell responses to *Vibrio cholerae* O1 lipopolysaccharide are associated with protection against infection from household contacts of patients with cholera in Bangladesh. *Clin Vaccine Immunol*. 2012;19:842–848.
85a. Aktar A, Rahman MA, Afrin S, et al. Plasma and memory B cell responses targeting O-specific polysaccharide (OSP) are associated with protection against Vibrio cholerae O1 infection among household contacts of cholera patients in Bangladesh. *PLoS Negl Trop Dis*. 2018;12:e0006399.
85b. Islam K, Hossain M, Kelly M, et al. Anti-O-specific polysaccharide (OSP) immune responses following vaccination with oral cholera vaccine CVD 103-HgR correlate with protection against cholera after infection with wild-type Vibrio cholerae O1 El Tor Inaba in North American volunteers. *PLoS Negl Trop Dis*. 2018;12:e0006376.
92. Leung DT, Rahman MA, Mohasin M, et al. Comparison of memory B cell, antibody-secreting cell, and plasma antibody responses in young children, older children, and adults with infection caused by *Vibrio cholerae* O1 El Tor Ogawa in Bangladesh. *Clin Vaccine Immunol*. 2011;18:1317–1325.
102. Saha A, Chowdhury MI, Khanam F, et al. Safety and immunogenicity study of a killed bivalent (O1 and O139) whole-cell oral cholera vaccine Shanchol, in Bangladeshi adults and children as young as 1 year of age. *Vaccine*. 2011;29:8285–8292.
103. Sur D, Lopez AL, Kanungo S, et al. Efficacy and safety of a modified killed-whole-cell oral cholera vaccine in India: an interim analysis of a cluster-randomised, double-blind, placebo-controlled trial. *Lancet*. 2009;374:1694–1702.
104. Kanungo S, Paisley A, Lopez AL, et al. Immune responses following one and two doses of the reformulated, bivalent, killed, whole-cell, oral cholera vaccine among adults and children in Kolkata, India: a randomized, placebo-controlled trial. *Vaccine*. 2009;27:6887–6893.
106. Clemens JD, Sack DA, Harris JR, et al. Field trial of oral cholera vaccines in Bangladesh: results from three-year follow-up. *Lancet*. 1990;335:270–273.
107. Van Loon FP, Clemens JD, Chakraborty J, et al. Field trial of inactivated oral cholera vaccines in Bangladesh: results from 5 years of follow-up. *Vaccine*. 1996;14:162–166.
111. Lucas ME, Deen JL, von Seidlein L, et al. Effectiveness of mass oral cholera vaccination in Beira, Mozambique. *N Engl J Med*. 2005;352:757–767.
114. Hashim R, Khatib AM, Enwere G, et al. Safety of the recombinant cholera toxin B subunit, killed whole-cell (rBS-WC) oral cholera vaccine in pregnancy. *PLoS Negl Trop Dis*. 2012;6:e1743.
119. Longini IM Jr, Nizam A, Ali M, et al. Controlling endemic cholera with oral vaccines. *PLoS Med*. 2007;4:e336.
120. Ali M, Emch M, von Seidlein L, et al. Herd immunity conferred by killed oral cholera vaccines in Bangladesh: a reanalysis. *Lancet*. 2005;366:44–49.
123. Reyburn R, Deen JL, Grais RF, et al. The case for reactive mass oral cholera vaccinations. *PLoS Negl Trop Dis*. 2011;5:e952.
124. Miller Neilan RL, Schaefer E, Gaff H, et al. Modeling optimal intervention strategies for cholera. *Bull Math Biol*. 2010;72:2004–2018.
128. World Health Organization. WHO technical working group on creation of an oral cholera vaccine stockpile: meeting report, Geneva, 26-27 April 2012. 2012. Pandemic and epidemic diseases, health security and environment. WHO/HSE/PED. 2012;2:1-23.
129a. Global Task Force on Cholera Control. Ending cholera: a global roadmap to 2030. https://www.who.int/cholera/publications/global-roadmap.pdf. Accessed April 19, 2019.
131a. Hubbard TP, Billings G, Dörr T, et al. A live vaccine rapidly protects against cholera in an infant rabbit model. *Sci Transl Med*. 2018;10:eaap8423.
138. Richie EE, Punjabi NH, Sidharta YY, et al. Efficacy trial of single-dose live oral cholera vaccine CVD 103-HgR in North Jakarta, Indonesia, a cholera-endemic area. *Vaccine*. 2000;18:2399–2410.
139. Qadri F, Chowdhury MI, Faruque SM, et al. Peru-15, a live attenuated oral cholera vaccine, is safe and immunogenic in Bangladeshi toddlers and infants. *Vaccine*. 2007;25:231–238.
144. Saha D, Karim MM, Khan WA, et al. Single-dose azithromycin for the treatment of cholera in adults. *N Engl J Med*. 2006;354:2452–2462.
145. Kaushik JS, Gupta P, Faridi MM, et al. Single dose azithromycin versus ciprofloxacin for cholera in children: a randomized controlled trial. *Indian Pediatr*. 2010;47:309–315.

References

1. Pollitzer R, Swaroop S, Burrows W. Cholera. *Monogr Ser World Health Organ*. 1959;58:1001–1019.
2. Greenough WBIII. The human, societal, and scientific legacy of cholera. *J Clin Invest*. 2004;113:334–339.
3. Howard-Jones N. Robert Koch and the cholera vibrio: a centenary. *Br Med J (Clin Res Ed)*. 1984;288:379–381.
4. Kaper JB, Morris JG Jr, Levine MM. Cholera. *Clin Microbiol Rev*. 1995;8:48–86.
5. Wang J, Villeneuve S, Zhang J, et al. On the antigenic determinants of the lipopolysaccharides of *Vibrio cholerae* O:1, serotypes Ogawa and Inaba. *J Biol Chem*. 1998;273:2777–2783.
6. Karlsson SL, Thomson N, Mutreja A, et al. Retrospective analysis of serotype switching of *Vibrio cholerae* O1 in a cholera endemic region shows it is a non-random process. *PLoS Negl Trop Dis*. 2016;10:e0005044. Oct 5.
7. Devault AM, Golding GB, Waglechner N, et al. Second-pandemic strain of *Vibrio cholerae* from the Philadelphia cholera outbreak of 1849. *N Engl J Med*. 2014;370:334–340.
8. Heidelberg JF, Eisen JA, Nelson WC, et al. DNA sequence of both chromosomes of the cholera pathogen *Vibrio cholerae*. *Nature*. 2000;406:477–483.
9. Egan ES, Waldor MK. Distinct replication requirements for the two *Vibrio cholerae* chromosomes. *Cell*. 2003;114:521–530.
10. Mekalanos JJ, Rubin EJ, Waldor MK. Cholera: molecular basis for emergence and pathogenesis. *FEMS Immunol Med Microbiol*. 1997;18:241–248.
11. Colwell RR. Global climate and infectious disease: the cholera paradigm. *Science*. 1996;274:2025–2031.
12. Nelson EJ, Harris JB, Morris JG, et al. Cholera transmission: the host, pathogen and bacteriophage dynamic. *Nat Rev Microbiol*. 2009;7:693–702.
13. Ritchie JM, Waldor MK. *Vibrio cholerae* interactions with the gastrointestinal tract: lessons from animal studies. *Curr Top Microbiol Immunol*. 2009;337:37–59.
14. Taylor RK, Miller VL, Furlong DB, et al. Use of phoA gene fusions to identify a pilus colonization factor coordinately regulated with cholera toxin. *Proc Natl Acad Sci USA*. 1987;84:2833–2837.
15. Krebs SJ, Taylor RK. Protection and attachment of *Vibrio cholerae* mediated by the toxin-coregulated pilus in the infant mouse model. *J Bacteriol*. 2011;193:5260–5270.
16. Ng D, Harn T, Altindal T, et al. The *Vibrio cholerae* minor pilin TcpB initiates assembly and retraction of the toxin-coregulated pilus. *PLoS Pathog*. 2016;12:e1006109.
17. Kovach ME, Shaffer MD, Peterson KM. A putative integrase gene defines the distal end of a large cluster of ToxR-regulated colonization genes in *Vibrio cholerae*. *Microbiology*. 1996;142(Pt 8):2165–2174.
18. Karaolis DK, Johnson JA, Bailey CC, et al. A *Vibrio cholerae* pathogenicity island associated with epidemic and pandemic strains. *Proc Natl Acad Sci USA*. 1998;95:3134–3139.
19. Fu Y, Waldor MK, Mekalanos JJ. Tn-Seq analysis of *Vibrio cholerae* intestinal colonization reveals a role for T6SS-mediated antibacterial activity in the host. *Cell Host Microbe*. 2013;14:652–663.
20. Levine MM, Kaper JB, Black RE, et al. New knowledge on pathogenesis of bacterial enteric infections as applied to vaccine development. *Microbiol Rev*. 1983;47:510–550.
21. Lencer WI, Tsai B. The intracellular voyage of cholera toxin: going retro. *Trends Biochem Sci*. 2003;28:639–645.
22. Sanchez J, Holmgren J. Cholera toxin—a foe and a friend. *Indian J Med Res*. 2011;133:153–163.
23. Rui H, Ritchie JM, Bronson RT, et al. Reactogenicity of live-attenuated *Vibrio cholerae* vaccines is dependent on flagellins. *Proc Natl Acad Sci USA*. 2010;107:4359–4364.
24. Waldor MK, Mekalanos JJ. Lysogenic conversion by a filamentous phage encoding cholera toxin. *Science*. 1996;272:1910–1914.
25. Matson JS, Withey JH, DiRita VJ. Regulatory networks controlling *Vibrio cholerae* virulence gene expression. *Infect Immun*. 2007;75:5542–5549.
26. Lee SH, Hava DL, Waldor MK, et al. Regulation and temporal expression patterns of *Vibrio cholerae* virulence genes during infection. *Cell*. 1999;99:625–634.
27. Schild S, Tamayo R, Nelson EJ, et al. Genes induced late in infection increase fitness of *Vibrio cholerae* after release into the environment. *Cell Host Microbe*. 2007;2:264–277.
28. Merrell DS, Butler SM, Qadri F, et al. Host-induced epidemic spread of the cholera bacterium. *Nature*. 2002;417:642–645.
29. Hsiao A, Ahmed AM, Subramanian S, et al. Members of the human gut microbiota involved in recovery from *Vibrio cholerae* infection. *Nature*. 2014;515:423–426. Nov 20.
30. Mutreja A, Kim DW, Thomson NR, et al. Evidence for several waves of global transmission in the seventh cholera pandemic. *Nature*. 2011;477:462–465.
31. Bik EM, Bunschoten AE, Gouw RD, et al. Genesis of the novel epidemic *Vibrio cholerae* O139 strain: evidence for horizontal transfer of genes involved in polysaccharide synthesis. *EMBO J*. 1995;14:209–216.
32. Domman D, Quilici ML, Dorman MJ, et al. Integrated view of *Vibrio cholerae* in the Americas. *Science*. 2017;358:789–793.
33. Newton AE, Heiman KE, Schmitz A, et al. Cholera in United States associated with epidemic in Hispaniola. *Emerg Infect Dis*. 2011;17:2166–2168.
34. Chin CS, Sorenson J, Harris JB, et al. The origin of the Haitian cholera outbreak strain. *N Engl J Med*. 2011;364:33–42.
35. Piarroux R, Barrais R, Faucher B, et al. Understanding the cholera epidemic. *Haiti Emerg Infect Dis*. 2011;17:1161–1168.
36. Hendriksen RS, Price LB, Schupp JM, et al. Population genetics of *Vibrio cholerae* from Nepal in 2010: evidence on the origin of the Haitian outbreak. *MBio*. 2011;2:e00157-11.
37. Qadri F, Islam T, Clemens JD. Cholera in Yemen—an old foe rearing its ugly head. *N Engl J Med*. 2017;377:2005–2007.
38. World Health Organization Global Task Force on Cholera Control. *Prevention and Control of Cholera Outbreaks: WHO Policy and Recommendations*. Geneva: WHO; 2011.
39. Cholera vaccines: WHO position paper—August 2017. *Wkly Epidemiol Rec*. 2017;92:477–498. Aug 25.

39a. World Health Organization. Weekly epidemiological record: cholera, 2017. No. 38. 2018;93:489–500. http://www.who.int/wer/2018/wer9338/en/. Accessed April 19, 2019.

40. Ali M, et al. Updated global burden of cholera in endemic countries. *PLoS Negl Trop Dis*. 2015;9:e0003832.
41. Weill FX, Domman D, Njamkepo E, et al. Genomic history of the seventh pandemic of cholera in Africa. *Science*. 2017;358:785–789. Nov 10.
42. Wang F, Butler T, Rabbani GH, et al. The acidosis of cholera. Contributions of hyperproteinemia, lactic acidemia, and hyperphosphatemia to an increased serum anion gap. *N Engl J Med*. 1986;315:1591–1595.
43. Ryan ET, Dhar U, Khan WA, et al. Mortality, morbidity, and microbiology of endemic cholera among hospitalized patients in Dhaka, Bangladesh. *Am J Trop Med Hyg*. 2000;63:12–20.
44. Chowdhury F, Kuchta A, Khan AI, et al. The increased severity in patients presenting to hospital with diarrhea in Dhaka, Bangladesh since the emergence of the hybrid strain of Vibrio cholerae O1 is not unique to cholera patients. *Int J Infect Dis*. 2015;40:9–14.
45. Deen JL, Von SL, Sur D, et al. The high burden of cholera in children: comparison of incidence from endemic areas in Asia and Africa. *PLoS Negl Trop Dis*. 2008;2:e173.
46. Harris JB, Khan AI, Larocque RC, et al. Blood group, immunity, and risk of infection with *Vibrio cholerae* in an area of endemicity. *Infect Immun*. 2005;73:7422–7427.
47. Shahinian ML, Passaro DJ, Swerdlow DL, et al. *Helicobacter pylori* and epidemic *Vibrio cholerae* O1 infection in Peru. *Lancet*. 2000;355:377–378.
48. Clemens J, Albert MJ, Rao M, et al. Impact of infection by *Helicobacter pylori* on the risk and severity of endemic cholera. *J Infect Dis*. 1995;171:1653–1656.
49. Harris JB, Larocque RC, Chowdhury F, et al. Susceptibility to *Vibrio cholerae* infection in a cohort of household contacts of patients with cholera in Bangladesh. *PLoS Negl Trop Dis*. 2008;2:e221.
50. Chowdhury F, Khan AI, Harris JB, et al. A comparison of clinical and immunologic features in children and older patients hospitalized with severe cholera in Bangladesh. *Pediatr Infect Dis J*. 2008;27:986–992.
51. Chowdhury F, Begum YA, Alam MM, et al. Concomitant enterotoxigenic *Escherichia coli* infection induces increased immune responses to *Vibrio cholerae* O1 antigens in patients with cholera in Bangladesh. *Infect Immun*. 2010;78:2117–2124.
52. Harris JB, Podolsky MJ, Bhuiyan TR, et al. Immunologic responses to *Vibrio cholerae* in patients co-infected with intestinal parasites in Bangladesh. *PLoS Negl Trop Dis*. 2009;3:e403.
53. Larocque RC, Sabeti P, Duggal P, et al. A variant in long palate, lung and nasal epithelium clone 1 is associated with cholera in a Bangladeshi population. *Genes Immun*. 2009;10:267–272.
54. Shin OS, Uddin T, Citorik R, et al. LPLUNC1 modulates innate immune responses to *Vibrio cholerae*. *J Infect Dis*. 2011;204:1349–1357.
55. Karlsson EK, Harris JB, Tabrizi S, et al. Natural selection in a Bangladeshi population from the cholera-endemic Ganges river delta. *Sci Transl Med*. 2013;5:192ra86. Jul 3.
56. World Health Organization. The Treatment of Diarrhoea, a Manual for Physicians and Other Senior Health Workers. 4th revision. 2005. http://whqlibdoc.who.int/publications/2005/9241593180.pdf. 2005.
57. Lesmana M, Rockhill RC, Sutanti D, et al. An evaluation of alkaline peptone water for enrichment of *Vibrio cholerae* in feces. *Southeast Asian J Trop Med Public Health*. 1985;16:265–267.
58. Qadri F, Wenneras C, Albert MJ, et al. Comparison of immune responses in patients infected with *Vibrio cholerae* O139 and O1. *Infect Immun*. 1997;65:3571–3576.
59. Benenson AS, Islam MR, Greenough WB 3rd. Rapid identification of *Vibrio cholerae* by darkfield microscopy. *Bull World Health Organ*. 1964;30:827–831.
60. Dick MH, Guillerm M, Moussy F, et al. Review of two decades of cholera diagnostics—how far have we really come? *PLoS Negl Trop Dis*. 2012;6:e1845.
61. Harris JB, Larocque RC, Qadri F, et al. Cholera. *Lancet*. 2012;379:2466–2476.
62. Mahalanabis D, Wallace CK, Kallen RJ, et al. Water and electrolyte losses due to cholera in infants and small children: a recovery balance study. *Pediatrics*. 1970;45:374–385.
63. World Health Organization. First steps for managing an outbreak of acute diarrhea, 2010. http://www.who.int/cholera/en.
64. *Guidelines for the Inpatient Treatment of Severely Malnourished Children*. Geneva: World Health Organization; 2003. http://www.who.int/nutrition/publications/guide_inpatient_text.pdf.
65. Gill DM, Meren R. ADP-ribosylation of membrane proteins catalyzed by cholera toxin: basis of the activation of adenylate cyclase. *Proc Natl Acad Sci USA*. 1978;75:3050–3054.
66. Alam NH, Yunus M, Faruque AS, et al. Symptomatic hyponatremia during treatment of dehydrating diarrheal disease with reduced osmolarity oral rehydration solution. *JAMA*. 2006;296:567–573.
67. Ramakrishna BS, Venkataraman S, Srinivasan P, et al. Amylase-resistant starch plus oral rehydration solution for cholera. *N Engl J Med*. 2000;342:308–313.
68. Global Task Force on Cholera Control. *First Steps for Managing an Outbreak of Acute Diarrhoea*. Geneva: World Health Organization; 2010.
69. Bennish ML, Azad AK, Rahman O, et al. Hypoglycemia during diarrhea in childhood. Prevalence, pathophysiology, and outcome. *N Engl J Med*. 1990;322:1357–1363.
70. Nelson EJ, Nelson DS, Salam MA, et al. Antibiotics for both moderate and severe cholera. *N Engl J Med*. 2011;364:5–7.
71. Lindenbaum J, Greenough WB, Islam MR. Antibiotic therapy of cholera in children. *Bull World Health Organ*. 1967;37:529–538.
72. Greenough IWB, Gordon RS, Rosenberg I, et al. Tetracycline in the treatment of cholera. *Lancet*. 1964;1:355–357.
73. Yamamoto T, Nair GB, Albert MJ, et al. Survey of in vitro susceptibilities of *Vibrio cholerae* O1 and O139 to antimicrobial agents. *Antimicrob Agents Chemother*. 1995;39:241–244.
74. Roy SK, Hossain MJ, Khatun W, et al. Zinc supplementation in children with cholera in Bangladesh: randomised controlled trial. *BMJ*. 2008;336:266–268.
75. Siddique AK. Cholera epidemic among Rwandan refugees: experience of ICDDR,B in Goma, Zaire. *Glimpse*. 1994;16:3–4.
76. Harris JB, Larocque RC, Charles RC, et al. Cholera's western front. *Lancet*. 2010;376:1961–1965.
77. Cholera Outbreak Training and Shigellosis Program (COTS Program). 2011. http://www.cotsprogram.com.
78. Comstock LE, Maneval D Jr, Panigrahi P, et al. The capsule and O antigen in *Vibrio cholerae* O139 Bengal are associated with a genetic region not present in *Vibrio cholerae* O1. *Infect Immun*. 1995;63:317–323.
79. Glass RI, Svennerholm AM, Khan MR, et al. Seroepidemiological studies of El Tor cholera in Bangladesh: association of serum antibody levels with protection. *J Infect Dis*. 1985;151:236–242.
80. Johnson RA, Uddin T, Aktar A, et al. Comparison of immune responses to the O-specific polysaccharide and lipopolysaccharide of *Vibrio cholerae* O1 in Bangladeshi adult patients with cholera. *Clin Vaccine Immunol*. 2012;19:1712–1721.
81. Losonsky GA, Yunyongying J, Lim V, et al. Factors influencing secondary vibriocidal immune responses: relevance for understanding immunity to cholera. *Infect Immun*. 1996;64:10–15.
82. Pasetti MF, Levine MM. Insights from natural infection-derived immunity to cholera instruct vaccine efforts. *Clin Vaccine Immunol*. 2012;19:1707–1711.
83. Mosley WH, McCormack WM, Ahmed A, et al. Report of the 1966-67 cholera vaccine field trial in rural East Pakistan. 2. Results of the serological surveys in the study population—the relationship of case rate to antibody titre and an estimate of the inapparent infection rate with

Vibrio cholerae. *Bull World Health Organ*. 1969;40:187–197.

84. Patel SM, Rahman MA, Mohasin M, et al. Memory B cell responses to *Vibrio cholerae* O1 lipopolysaccharide are associated with protection against infection from household contacts of patients with cholera in Bangladesh. *Clin Vaccine Immunol*. 2012;19:842–848.
85. Aktar A, Rahman MA, Afrin S, et al. O-specific polysaccharide-specific memory B cell responses in young children, older children, and adults infected with *Vibrio cholerae* O1 Ogawa in Bangladesh. *Clin Vaccine Immunol*. 2016;23:427–435. May 6.
85a. Aktar A, Rahman MA, Afrin S, et al. Plasma and memory B cell responses targeting O-specific polysaccharide (OSP) are associated with protection against Vibrio cholerae O1 infection among household contacts of cholera patients in Bangladesh. *PLoS Negl Trop Dis*. 2018;12:e0006399.
85b. Islam K, Hossain M, Kelly M, et al. Anti-O-specific polysaccharide (OSP) immune responses following vaccination with oral cholera vaccine CVD 103-HgR correlate with protection against cholera after infection with wild-type Vibrio cholerae O1 El Tor Inaba in North American volunteers. *PLoS Negl Trop Dis*. 2018;12:e0006376.
86. Arifuzzaman M, Rashu R, Leung DT, et al. Antigen-specific memory T cell responses after vaccination with an oral killed cholera vaccine in Bangladeshi children and comparison to responses in patients with naturally acquired cholera. *Clin Vaccine Immunol*. 2012;19:1304–1311. Aug.
87. Ellis CN, LaRocque RC, Uddin T, et al. Comparative proteomic analysis reveals activation of mucosal innate immune signaling pathways during cholera. *Infect Immun*. 2015;83:1089–1103. Mar.
88. Leung DT, Bhuiyan TR, Nishat NS, et al. Circulating mucosal associated invariant T cells are activated in *Vibrio cholerae* O1 infection and associated with lipopolysaccharide antibody responses. *PLoS Negl Trop Dis*. 2014;8:e3076. Aug 21.
89. Charles RC, Nakajima R, Liang L, et al. Plasma and mucosal immunoglobulin M, immunoglobulin A, and immunoglobulin G responses to the *Vibrio cholerae* O1 protein immunome in adults with cholera in Bangladesh. *J Infect Dis*. 2017;216:125–134. Jul 1.
90. Kauffman RC, Bhuiyan TR, Nakajima R, et al. Single-cell analysis of the plasmablast response to *Vibrio cholerae* demonstrates expansion of cross-reactive memory B cells. *MBio*. 2016;7:Dec 20, pii: e02021-16.
91. Leung DT, Chowdhury F, Calderwood SB, et al. Immune responses to cholera in children. *Expert Rev Anti Infect Ther*. 2012;10:435–444.
92. Leung DT, Rahman MA, Mohasin M, et al. Comparison of memory B cell, antibody-secreting cell, and plasma antibody responses in young children, older children, and adults with infection caused by *Vibrio cholerae* O1 El Tor Ogawa in Bangladesh. *Clin Vaccine Immunol*. 2011;18:1317–1325.
93. Waldman RJ, Mintz ED. Papowitz HE. The cure for cholera—improving access to safe water and sanitation. *N Engl J Med*. 2013;368:592–594.
94. World Health Organization. Water Sanitation and Health (WSH): Health through Safe Drinking Water and Basic Sanitation. http://www.who.int/water_sanitation_health/mdg1/en/. Accessed June 19, 2014.
95. Ryan ET. The cholera pandemic, still with us after half a century: time to rethink. *PLoS Negl Trop Dis*. 2011;5:e1003.
96. Graves PM, Deeks JJ, Demicheli V, et al. Vaccines for preventing cholera: killed whole cell or other subunit vaccines (injected). *Cochrane Database Syst Rev*. 2010;(8):CD000974.
97. Baik YO, Choi SK, Olveda RM, et al. A randomized, non-inferiority trial comparing two bivalent killed, whole cell, oral cholera vaccines (Euvichol vs Shanchol) in the Philippines. *Vaccine*. 2015;33:6360–6365.
98. Anh DD, Canh do G, Lopez AL, et al. Safety and immunogenicity of a reformulated Vietnamese bivalent killed, whole-cell, oral cholera vaccine in adults. *Vaccine*. 2007;25:1149–1155.
99. Mahalanabis D, Lopez AL, Sur D, et al. A randomized, placebo-controlled trial of the bivalent killed, whole-cell, oral cholera vaccine in adults and children in a cholera endemic area in Kolkata. *India PLoS One*. 2008;3:e2323.
100. Trach DD, Clemens JD, Ke NT, et al. Field trial of a locally produced, killed, oral cholera vaccine in Vietnam. *Lancet*. 1997;349:231–235.
101. Thiem VD, Deen JL, Von SL, et al. Long-term effectiveness against cholera of oral killed whole-cell vaccine produced in Vietnam. *Vaccine*. 2006;24:4297–4303.
102. Saha A, Chowdhury MI, Khanam F, et al. Safety and immunogenicity study of a killed bivalent (O1 and O139) whole-cell oral cholera vaccine Shanchol, in Bangladeshi adults and children as young as 1 year of age. *Vaccine*. 2011;29:8285–8292.
103. Sur D, Lopez AL, Kanungo S, et al. Efficacy and safety of a modified killed-whole-cell oral cholera vaccine in India: an interim analysis of a cluster-randomised, double-blind, placebo-controlled trial. *Lancet*. 2009;374:1694–1702.
104. Kanungo S, Paisley A, Lopez AL, et al. Immune responses following one and two doses of the reformulated, bivalent, killed, whole-cell, oral cholera vaccine among adults and children in Kolkata, India: a randomized, placebo-controlled trial. *Vaccine*. 2009;27:6887–6893.
105. Peltola H, Siitonen A, Kyronseppa H, et al. Prevention of travellers' diarrhoea by oral B-subunit/whole-cell cholera vaccine. *Lancet*. 1991;338:1285–1289.
106. Clemens JD, Sack DA, Harris JR, et al. Field trial of oral cholera vaccines in Bangladesh: results from three-year follow-up. *Lancet*. 1990;335:270–273.
107. Van Loon FP, Clemens JD, Chakraborty J, et al. Field trial of inactivated oral cholera vaccines in Bangladesh: results from 5 years of follow-up. *Vaccine*. 1996;14:162–166.
108. Clemens JD, Sack DA, Harris JR, et al. Impact of B subunit killed whole-cell and killed whole-cell-only oral vaccines against cholera upon treated diarrhoeal illness and mortality in an area endemic for cholera. *Lancet*. 1988;1:1375–1379.
109. Clemens JD, Sack DA, Ivanoff B. Misleading negative findings in a field trial of killed, oral cholera vaccine in Peru. *J Infect Dis*. 2001;183:1306–1309.
110. Taylor DN, Cardenas V, Sanchez JL, et al. Two-year study of the protective efficacy of the oral whole cell plus recombinant B subunit cholera vaccine in Peru. *J Infect Dis*. 2000;181:1667–1673.
111. Lucas ME, Deen JL, von Seidlein L, et al. Effectiveness of mass oral cholera vaccination in Beira, Mozambique. *N Engl J Med*. 2005;352:757–767.
112. Sanchez JL, Vasquez B, Begue RE, et al. Protective efficacy of oral whole-cell/recombinant-B-subunit cholera vaccine in Peruvian military recruits. *Lancet*. 1994;344:1273–1276.
113. Bhattacharya SK, Sur D, Ali M, et al. 5 year efficacy of a bivalent killed whole-cell oral cholera vaccine in Kolkata, India: a cluster-randomised, double-blind, placebo-controlled trial. *Lancet Infect Dis*. 2013;13:1050–1056. Dec.
114. Hashim R, Khatib AM, Enwere G, et al. Safety of the recombinant cholera toxin B subunit, killed whole-cell (rBS-WC) oral cholera vaccine in pregnancy. *PLoS Negl Trop Dis*. 2012;6:e1743.
115. Ivers LC, Charles RC, Hilaire IJ, et al. Immunogenicity of the bivalent oral cholera vaccine Shanchol in Haitian adults with HIV infection. *J Infect Dis*. 2015;212:779–783. Sep 1.
116. Qadri F, Wierzba TF, Ali M, et al. Efficacy of a single-dose, inactivated oral cholera vaccine in Bangladesh. *N Engl J Med*. 2016;374:1723–1732. May 5.
117. Saha A, Khan A, Salma U, et al. The oral cholera vaccine Shanchol when stored at elevated temperatures maintains the safety and immunogenicity profile in Bangladeshi participants. *Vaccine*. 2016;34:1551–1558. Mar 18.
118. Kanungo S, Desai SN, Nandy RK, et al. Flexibility of oral cholera vaccine dosing-a randomized controlled trial measuring immune responses following alternative vaccination schedules in a cholera hyper-endemic zone. *PLoS Negl Trop Dis*. 2015;9:e0003574. Mar 12.
119. Longini IM Jr, Nizam A, Ali M, et al. Controlling endemic cholera with oral vaccines. *PLoS Med*. 2007;4:e336.
120. Ali M, Emch M, von Seidlein L, et al. Herd immunity conferred by killed oral cholera vaccines in Bangladesh: a reanalysis. *Lancet*. 2005;366:44–49.
121. Andrews JR, Basu S. Transmission dynamics and control of cholera in Haiti: an epidemic model. *Lancet*. 2011;377:1248–1255.
122. Tuite AR, Tien J, Eisenberg M, et al. Cholera epidemic in Haiti, 2010: using a transmission model to explain spatial spread of disease and identify optimal control interventions. *Ann Intern Med*. 2011;154:593–601.
123. Reyburn R, Deen JL, Grais RF, et al. The case for reactive mass oral cholera vaccinations. *PLoS Negl Trop Dis*. 2011;5:e952.
124. Miller Neilan RL, Schaefer E, Gaff H, et al. Modeling optimal intervention strategies for cholera. *Bull Math Biol*. 2010;72:2004–2018.
125. Chao DL, Halloran ME, Longini IM Jr. Vaccination strategies for epidemic cholera in Haiti with implications for the developing world. *Proc Natl Acad Sci USA*. 2011;108:7081–7085.
126. Jeuland M, Cook J, Poulos C, et al. Cost-effectiveness of new-generation oral cholera vaccines: a multisite analysis. *Value Health*. 2009;12:899–908.
127. Schaetti C, Weiss MG, Ali SM, et al. Costs of illness due to cholera, costs of immunization and cost-effectiveness of an oral cholera mass vaccination campaign in Zanzibar. *PLoS Negl Trop Dis*. 2012;6:e1844.
128. World Health Organization (WHO). WHO technical working group on creation of an oral cholera vaccine stockpile: meeting report, Geneva, 26-27 April 2012. 2012. Pandemic and epidemic diseases, health security and environment. WHO/HSE/PED. 2012;2:1-23.
129. Deployments from the oral cholera vaccine stockpile, 2013–2017. *Wkly Epidemiol Rec*. 2017;92:437–442. Aug 11.
129a. Global Task Force on Cholera Control. Ending cholera: a global roadmap to 2030. https://www.who.int/cholera/publications/global-roadmap.pdf. Accessed April 19, 2019.
130. Wong KK, Burdette E, Mahon BE, et al. Recommendations of the Advisory Committee on Immunization Practices for Use of Cholera Vaccine. *MMWR Morb Mortal Wkly Rep*. 2017;66:482–485. May 12.
131. Chen WH, Cohen MB, Kirkpatrick BD, et al. Single-dose live oral cholera vaccine CVD 103-HgR protects against human experimental infection with *Vibrio cholerae* O1 El Tor. *Clin Infect Dis*. 2016;62:1329–1335. Jun 1.
131a. Hubbard TP, Billings G, Dörr T, et al. A live vaccine rapidly protects against cholera in an infant rabbit model. *Sci Transl Med*. 2018;10:eaap8423.
132. Ryan ET, Calderwood SB, Qadri F. Live attenuated oral cholera vaccines. *Expert Rev Vaccines*. 2006;5:483–494.
133. Cohen MB, Giannella RA, Bean J, et al. Randomized, controlled human challenge study of the safety, immunogenicity, and protective efficacy of a single dose of Peru-15, a live attenuated oral cholera vaccine. *Infect Immun*. 2002;70:1965–1970.
134. Levine MM, Kaper JB, Herrington D, et al. Safety, immunogenicity, and efficacy of recombinant live oral cholera vaccines, CVD 103 and CVD 103-HgR. *Lancet*. 1988;2:467–470.
135. Kotloff KL, Wasserman SS, O'Donnell S, et al. Safety and immunogenicity in North Americans of a single dose of live oral cholera vaccine CVD 103-HgR: results of a randomized, placebo-controlled, double-blind crossover trial. *Infect Immun*. 1992;60:4430–4432.
136. Kenner JR, Coster TS, Taylor DN, et al. Peru-15, an improved live attenuated oral vaccine candidate for *Vibrio cholerae* O1. *J Infect Dis*. 1995;172:1126–1129.
137. Sack DA, Sack RB, Shimko J, et al. Evaluation of Peru-15, a new live oral vaccine for cholera, in volunteers. *J Infect Dis*. 1997;176:201–205.
138. Richie EE, Punjabi NH, Sidharta YY, et al. Efficacy trial of single-dose live oral cholera vaccine CVD 103-HgR in North Jakarta, Indonesia, a cholera-endemic area. *Vaccine*. 2000;18:2399–2410.
139. Qadri F, Chowdhury MI, Faruque SM, et al. Peru-15, a live attenuated oral cholera vaccine, is safe and immunogenic in Bangladeshi toddlers and infants. *Vaccine*. 2007;25:231–238.
140. Garcia L, Jidy MD, Garcia H, et al. The vaccine candidate *Vibrio cholerae* 638 is protective against cholera in healthy volunteers. *Infect Immun*. 2005;73:3018–3024.
141. Liang W, Wang S, Yu F, et al. Construction and evaluation of a safe, live, oral *Vibrio cholerae* vaccine candidate, IEM108. *Infect Immun*. 2003;71:5498–5504.
142. Sayeed MA, Bufano MK, Xu P, et al. A cholera conjugate vaccine containing O-specific polysaccharide (OSP) of *V. cholerae* O1 inaba and recombinant fragment of tetanus toxin heavy chain (OSP:rTTHc) induces serum, memory and lamina proprial responses against OSP and is protective in mice. *PLoS Negl Trop Dis*. 2015;9:e0003881. Jul 8.
143. Molla AM, Rahman M, Sarker SA, et al. Stool electrolyte content and purging rates in diarrhea caused by rotavirus, enterotoxigenic *E. coli*, and *V. cholerae* in children. *J Pediatr*. 1981;98:835–838.
144. Saha D, Karim MM, Khan WA, et al. Single-dose azithromycin for the treatment of cholera in adults. *N Engl J Med*. 2006;354:2452–2462.
145. Kaushik JS, Gupta P, Faridi MM, et al. Single dose azithromycin versus ciprofloxacin for cholera in children: a randomized controlled trial. *Indian Pediatr*. 2010;47:309–315.
146. Saha D, Khan WA, Karim MM, et al. Single-dose ciprofloxacin versus 12-dose erythromycin for childhood cholera: a randomised controlled trial. *Lancet*. 2005;366:1085–1093.
147. Khan WA, Bennish ML, Seas C, et al. Randomised controlled comparison of single-dose ciprofloxacin and doxycycline for cholera caused by *Vibrio cholerae* 01 or 0139. *Lancet*. 1996;348:296–300.
148. Islam MS, Midzi SM, Charimari L, et al. Susceptibility to fluoroquinolones of *Vibrio cholerae* O1 isolated from diarrheal patients in Zimbabwe. *JAMA*. 2009;302:2321–2322.

215 Other Pathogenic Vibrios

Marguerite A. Neill

SHORT VIEW SUMMARY

Definition
- Vibrios are gram-negative rods whose environmental niche is primarily marine and estuarine waters.
- They may also be found in freshwater sources.
- The halophilic vibrios require higher concentrations of salt for growth.

Epidemiology
- The most common *Vibrio* species causing human illness are *Vibrio parahemolyticus* and *Vibrio vulnificus.*
- The incidence of vibriosis in the United States has been increasing, indicating that current prevention efforts are not effective.
- Illness is more common in the warmer summer months when *Vibrio* populations are higher and there is increased human exposure from shellfish consumption and recreational water exposure.
- *Vibrio* species primarily cause diarrhea of varying severity, soft tissue infection, and/or bacteremia.
- *V. vulnificus* causes a distinct soft tissue infection with rapidly developing hemorrhagic bullae.
- *Vibrio* infections are more severe and more often fatal in people with cirrhosis, other underlying liver disease, or immune compromise.

Microbiology
- The major pathogenic vibrios grow well in blood culture media and common nonselective media used for wound cultures.
- Culture-independent diagnostic testing by DNA amplification panels is increasing the frequency of detecting *Vibrio* infections in the United States.

Diagnosis
- *Vibrio* infection should be suspected in people with diarrhea, sepsis, or wound infection after seafood consumption or marine water exposure.

Therapy
- Rapid recognition of possible *V. vulnificus* infection is key to improving survival and requires antimicrobial treatment as well as surgical débridement.
- Survival is improved in *V. vulnificus* infection when a third-generation cephalosporin is added to doxycycline or a quinolone, provided débridement of necrotic tissue is done.
- Treatment with doxycycline or a quinolone can decrease symptoms and fecal shedding in people with diarrhea continuing for 5 days.

Prevention
- No vaccine is available for the noncholera vibrios.
- Thorough cooking of seafood with prompt consumption or refrigeration is needed to decrease the risk for *Vibrio* infection.

Vibrio species are ubiquitous in estuarine waters in the temperate zones. Plankton blooms and temperature upshifting in the spring are followed by rapid outgrowth of most vibrios. Molluscan shellfish, which are filter feeders, acquire vibrios as part of their normal microbiota during the warmer months. Shellfish contamination by these vibrios thus occurs as a consequence of the normal climate-associated changes in *Vibrio* prevalence in coastal waters. In addition to this mechanism, sewage contamination of shellfish beds in epidemic cholera can contribute significantly to disease burden.

In the United States, illnesses caused by the commonly isolated pathogenic vibrios have a marked seasonal peak, with more than 90% of cases occurring between April and October. This presumably reflects seasonal changes in shellfish consumption, recreational water exposure, and the increase in densities of vibrios in marine waters during the warmer months. The incidence of vibriosis in the United States has been increasing despite prevention efforts.[1] This may be reflective of both host and exposure factors, such as increases in water-related outdoor activities (swimming, boating), natural disasters (hurricanes, flooding), increased consumption of seafood, and more persons living with chronic liver disease and immunocompromising conditions. Recently, more widespread use of DNA amplification panels for detection of diarrheal pathogens has led to a 54% increase in the number of *Vibrio* infections reported in 2017 compared with the prior 2 years.[2]

In addition to *Vibrio cholerae* serotype O1 and *Campylobacter fetus* (formerly known as *Vibrio fetus*), other vibrios have been clearly associated with human disease. The best known among the halophilic (salt-requiring) vibrios are *V. parahaemolyticus* and *V. vulnificus,* both of which can cause serious illness and death. Three other halophilic vibrios, *V. alginolyticus, V. fluvialis,* and *V. furnissii,* cause infections of appreciable frequency but lesser clinical severity. The former *V. hollisae* and *V. damselae* have been reclassified and are now *Grimontia hollisae* and *Photobacterium damselae,* respectively. *V. metschnikovii, V. cincinnatiensis,* and *V. harveyi* (*V. carchariae*) rarely cause human illness. The nonhalophilic vibrios include non-O1/O139 *V. cholerae* and *V. mimicus;* like *V. cholerae,* they cannot grow at higher salt concentrations. They are worldwide in distribution and have frequently been incriminated in human illness.

VIBRIO PARAHAEMOLYTICUS

V. parahaemolyticus, a halophilic vibrio, is a major cause of acute diarrheal disease in Japan.[3] In the United States, *V. parahaemolyticus* was the most commonly isolated *Vibrio* species and the most common of the speciated vibrios reported.[1] This pathogen was the most common bacterial cause of foodborne disease in Taiwan, accounting for 35% of all outbreaks[4]; in less-developed countries, it has been incriminated in up to 20% of acute diarrheal illnesses. A specific serotype, *V. parahaemolyticus* O3:K6, emerged as an important cause of human illness in Southeast Asia[5] and, since 1996, became established globally by a pandemic clone.[6] In 1998, this serotype first appeared in the United States,[7] causing a large multistate outbreak that prompted regulatory changes in programs for bacteriologic monitoring of shellfish.[8] In 2004, an outbreak of gastroenteritis caused by serotype O6:K18 was traced to raw oysters from the Gulf of Alaska, harvested when the water temperature was higher than 15°C.[9] Whether global warming trends portend locally acquired *V. parahaemolyticus* infection even at high northern latitudes is a concern and emphasizes the need for surveillance and appropriate diagnostics, a recurring theme in emerging infectious diseases.

The genome of *V. parahaemolyticus* has been sequenced.[10] Like *V. cholerae, V. parahaemolyticus* has two circular chromosomes, but unlike *V. cholerae,* a type III secretion system has been found on both of the two chromosomes of *V. parahaemolyticus.*[11] This finding likely underlies the inflammatory diarrhea seen in *V. parahaemolyticus,* but

not *V. cholerae,* infection, and suggests distinctly different mechanisms for diarrhea from these two pathogenic vibrios.

As suggested both by clinical manifestations and by animal studies, *V. parahaemolyticus* has the genetic endowment to produce an enterotoxin and to cause an inflammatory reaction in the small bowel mucosa. Major degrees of intestinal fluid loss are seldom seen, and the tissue damage that this vibrio causes is generally less extensive than that observed in shigellosis. Two hemolysins, thermostable direct hemolysin (TDH) and TDH-related hemolysin, have enterotoxin activity; strains negative for TDH and TDH-related hemolysin are usually avirulent.

Epidemiology

Because of lack of specificity of the clinical features of the illness, the epidemiologic history usually provides the most important clue to diagnosis. *V. parahaemolyticus* is ubiquitous in coastal waters,[3] although it typically is not recovered from estuarine waters during winter months in temperate zones. During periods of low temperature or nutrient deprivation, it enters a viable but nonculturable state.[12]

Consumption of raw or undercooked shellfish is the most common means of acquiring *V. parahaemolyticus* infection. In the United States, raw oysters are the most common vehicle.[13] In a microbiologic survey of oysters harvested from US waters, the frequency of *V. parahaemolyticus* contamination was consistently greater than that for *V. vulnificus.*[14,15] Inadequately cooked seafood can harbor small numbers of surviving vibrios, as can food contaminated by seawater on ships. *V. parahaemolyticus* can proliferate rapidly to reach high colony counts in contaminated foods held at ambient temperature for a few hours. This presumably contributes to the high attack rates seen in common source outbreaks.

Person-to-person transmission has not been documented, suggesting that the infective dose for normal persons is relatively high. *V. parahaemolyticus* has rarely been cultured from asymptomatic people, and no carrier state has been identified.

Clinical Manifestations

Gastroenteritis is the most common clinical illness associated with *V. parahaemolyticus* infection; wound infections and septicemia may be seen, but much less frequently. Enteric illness ranges from mild watery diarrhea to a frank dysentery-like syndrome. Illness commonly begins with the acute onset of explosive watery diarrhea, generally within 24 to 72 hours of ingestion of the contaminated seafood. Often, the diarrhea is accompanied by mild to moderately severe cramping and abdominal pain; low-grade fever, mild chills, and headache occur in fewer than half of infected people. The fluid loss is rarely severe, and death caused by *V. parahaemolyticus* is rare, usually occurring in very young children, older adults, or people with underlying disease.

Laboratory Findings

The diarrheal fluid is characteristically watery, sometimes mucoid, and occasionally bloody (<15% of cases). Fecal leukocytes are often present. *V. parahaemolyticus* is a pleomorphic gram-negative rod that is a facultative anaerobe. It is readily identified on the selective thiosulfate citrate–bile salts–sucrose (TCBS) agar, on which it appears as distinct opaque green colonies. While culture-independent diagnostic testing has greater sensitivity (lower limit of detection) compared with stool cultures, it does not provide antibiotic susceptibility or molecular subtyping results to compare isolates. Reflex cultures from fecal specimens positive for *Vibrio* species by DNA amplification panels were only positive in 38% of cultures.[2]

Nearly all clinical isolates of *V. parahaemolyticus* cause β-hemolysis of human erythrocytes (the Kanagawa reaction), which is caused by the production of TDH. Interestingly, growth in bile salt–containing environments enhances the expression of several virulence traits in *V. parahaemolyticus.*[16]

In patients with *V. parahaemolyticus* enteric infection, both serum and coproantibody responses to lipopolysaccharide and to TDH were detected.[17] Mucosal biopsies from the duodenum and rectum showed inflammatory changes, suggesting that the small and large intestines are affected. Tumor necrosis factor-α (TNF-α) levels were noted to be elevated acutely in *V. parahaemolyticus* infection, similar to observations in shigellosis but in contrast to cholera infections, in which TNF-α levels are not elevated.

Differential Diagnosis

Because *V. parahaemolyticus* is ubiquitous in coastal waters throughout the temperate and tropical zones of the world, this pathogen must be considered in the differential diagnosis of all acute diarrheal illnesses that follow the ingestion of seafood. There are no clinical features that, in the individual case, reliably distinguish diarrhea caused by *V. parahaemolyticus* from that caused by enterotoxigenic *Escherichia coli* or from milder cases of shigellosis or salmonellosis. Vomiting is characteristically less prominent than in disease caused by staphylococcal enterotoxin, and the cramping abdominal pain is generally less severe than that typical of food poisoning caused by *Clostridium perfringens.*

Therapy

No treatment is required by most patients because the gastroenteritis is usually self-limited. However, antimicrobial therapy could be considered for those patients with diarrhea lasting longer than 5 days.[18] Therapy with doxycycline or a quinolone would be expected to shorten the clinical course and duration of pathogen excretion. In the United States, 16% to 20% of patients with *V. parahemolyticus* infection were hospitalized; the overall case fatality rate was <1%.[1] Occasionally patients, usually at the extremes of age, may become sufficiently dehydrated that oral or intravenous rehydration is needed. Wound infections due to *V. parahaemolyticus* are managed similarly among all the vibrios causing soft tissue infection, including antibiotic treatment and débridement as needed.

Prevention

V. parahaemolyticus can remain viable in shrimp or crab meat for several minutes at temperatures as high as 80°C, and it is especially important when cooking large quantities of such foods to ensure that all portions of the seafood are heated to cooking temperatures adequate to kill *Vibrio* species. Of only slightly less importance is the necessity of refrigerating cooked seafood if it is not to be eaten immediately after cooking. Shipboard outbreaks can be prevented by avoiding the use of untreated seawater in galleys. Irradiation at a dose of 3.0 kGy can reduce *V. parahaemolyticus* levels by 6 logs without killing the oysters or adversely affecting their organoleptic qualities[19]; however, lower doses may not inactivate viruses.

It is not known whether protective immunity is conferred by clinical infection. No effective vaccine is currently available.

VIBRIO VULNIFICUS

Like other pathogenic halophilic vibrios, *V. vulnificus* is part of the normal marine microbiota and, in the temperate zones, reaches sufficient concentrations to cause clinical illness only in the warmer months of the year. Almost all oysters harvested in the summer from the Chesapeake Bay contain this pathogen, as do 10% of crabs. In the United States, 25% of all *Vibrio* infections were from a non-foodborne source and of these, *V. vulnificus* was the most common species isolated. Almost half of all *V. vulnificus* infections were non-foodborne.[20] After Hurricane Katrina in 2005, 18 cases of wound infection caused by vibrios were reported; 14 (82%) were caused by *V. vulnificus,* resulting in three deaths.[21] Infections from *V. vulnificus* may be increasing in the United States, particularly in association with Gulf coast oyster consumption,[22] and warmer water temperatures (>22°C) in the Gulf of Mexico may be contributing in part to the increase.

Over 80% of reported *V. vulnificus* cases were hospitalized in the United States; the case-fatality rate of 30% is the highest among infections caused by any *Vibrio* species.[1] *V. vulnificus* is estimated to account for 90% of all seafood-related deaths in the United States. Hippocrates may have provided the first description of a *V. vulnificus* infection in the 5th century BC when he described the rapid progression of a severe foot cellulitis with black blisters in Criton of Thasos, which was fatal in 48 hours.[23]

Clinical Manifestations

V. vulnificus is primarily associated with a severe, distinctive soft tissue infection or septicemia or both, rather than diarrheal illness.[24] In

compromised hosts, especially patients with cirrhosis, *V. vulnificus* can invade the bloodstream without causing gastrointestinal symptoms. The clinical picture is one of abrupt onset of chills and fever, often (in 33% of cases) followed by hypotension, usually (in 75%) followed by the development of metastatic cutaneous lesions within 36 hours after onset. These begin as erythematous lesions and rapidly evolve to hemorrhagic bullae or vesicles and then to necrotic ulcers (Fig. 215.1). *V. vulnificus* bacteremia has been fatal in over 50% of cases in which it occurred. More than 90% of such patients had a history of having consumed raw oysters in the 7 days before illness onset; concentrations of *V. vulnificus* of 10^3 colony-forming units per gram of oysters have produced illness. Although oysters harbor genetically heterogeneous populations of *V. vulnificus,* only one strain type has been recovered from human tissues in invasive infections.[25] In addition to cirrhosis, other risk factors for the septicemic form of *V. vulnificus* infection include other liver diseases; iron overload states such as hemochromatosis, hemolytic anemia, or chronic renal failure; malignancy; human immunodeficiency virus infection; and immunosuppressive medications.

Posttraumatic wound infections with *V. vulnificus* usually occur on the legs and arms; the diagnosis can be suspected from the distinct clinical picture (see Fig. 215.1).[21] After contamination of a superficial wound by warm seawater, *V. vulnificus* can cause a rapidly developing, intense cellulitis, necrotizing vasculitis, and ulcer formation in both healthy people and immunocompromised hosts, and bacteremia is frequent. This was graphically illustrated in a report of a surgeon with a minor cut to his finger while saltwater fishing, with severe advancing cellulitis developing <12 hours later. The rapidity and severity (arm amputation despite appropriate antibiotics and emergency débridements) may have been related to adalimumab (a TNF-α inhibitor), which he was receiving for psoriatic arthritis.[27] In another case, *V. vulnificus* apparently survived on intact skin for more than 24 hours after handling tilapia fish, with the patient subsequently developing a necrotic cellulitis after traumatic injury to the hand while working on a motor vehicle engine.[28,29]

V. vulnificus is an infrequent cause of acute, self-limited diarrheal illness in individuals receiving gastric acid suppression therapy,[30] ocular infections usually occurring after an injury from molluscan shell fragments,[31] and, rarely, septic arthritis.[32]

The major determinant of virulence in *V. vulnificus* is its polysaccharide capsule, which renders the bacterium resistant to phagocytosis and directly stimulates release of inflammatory cytokines such as TNF-α.[33] Other contributors to pathogenicity include a variety of extracellular proteins and cell wall lipopolysaccharide. Host-derived factors that contribute to the pathogen's virulence include the availability of iron and at least one inflammatory mediator. *V. vulnificus* can alter expression of several genes differentially in response to low-iron and iron-rich conditions and, in the latter, can multiply rapidly.[34] The predilection of this *Vibrio* species to cause disease in patients with iron overload states can be explained by its ability to sequester iron from hemoglobin and 100% saturated (but not 30%, or normally saturated) transferrin.

FIG. 215.1 Cellulitis (A) and hemorrhagic bullae (B) in *Vibrio vulnificus* infection. *(From Centers for Disease Control and Prevention. Vibrio illnesses after Hurricane Katrina—multiple states, August-September, 2005.* MMWR Morb Mortal Wkly Rep. *2005;54:928–931.)*

Differential Diagnosis

V. vulnificus should be suspected in any immunocompromised host (but especially with cirrhosis or underlying liver disease) who develops a septicemic illness associated with necrotizing cutaneous lesions within 1 to 3 days after the ingestion of oysters. Although rare, the clinical syndrome is distinct and should suggest this diagnosis.

Similarly, the development of cellulitis in humans occupationally or recreationally exposed to seawater should suggest infection with *V. vulnificus* (especially in the presence of severe, necrotizing cellulitis). Other *Vibrio* species may cause soft tissue infections, but these are usually less severe.

V. vulnificus grows readily in blood culture media, on blood or other nonselective agar for wound cultures, and on MacConkey and TCBS media; final identification is made by standard biochemical tests. Because *V. vulnificus* ferments lactose, it can be overlooked in cultures grown on MacConkey agar unless the laboratory is advised to specifically look for it; thus communication between the clinician and microbiology laboratory personnel is key. *V. vulnificus* can also be identified from stool specimens by DNA amplification panels that increasingly are being adopted by clinical laboratories for syndromic testing (e.g., diarrhea).[35]

A real-time quantitative polymerase chain reaction (PCR) assay to detect the *toxR* gene of *V. vulnificus* was capable of detecting as few as five copies per microliter of serum; positive PCR results were obtained in 5 of 22 cases with negative blood cultures but in which *V. vulnificus* was isolated from soft tissue.[36] In a retrospective study using real-time PCR for *V. vulnificus toxR,* DNA copy numbers in tissue were considerably higher than in blood. Such testing could add diagnostic speed as well as specificity, especially in persons who had received antibiotics prior to obtaining cultures.[37]

Therapy

Early administration of antibiotics in *V. vulnificus* infection is critical because the cellulitis can advance very rapidly. Immediate surgical consultation should be obtained because débridement of all devitalized tissue or even amputation may be lifesaving.[27] In one retrospective case series, temporizing surgical management for those patients with shock at admission was associated with lower mortality versus aggressive débridement (26% vs. 60%), but choice and timing of antimicrobial therapy were not detailed.[38]

Fluoroquinolones, third-generation cephalosporins, and doxycycline are highly active against *V. vulnificus,* and the combination of ciprofloxacin and cefotaxime exhibited synergy.[39] Survival in *V. vulnificus* necrotizing fasciitis was higher in those treated with minocycline or ciprofloxacin added to a third-generation cephalosporin, provided that prompt surgical intervention was obtained.[40,41]

Prevention

Although patients with underlying liver disease and other chronic illnesses should be warned of the hazards of eating raw oysters, this has not been accomplished effectively in the United States, even when required by law.[1] A capsular polysaccharide conjugate vaccine has been developed, but studies indicate that polyclonal immunoglobulin, actively or passively derived, was necessary for cross-protection among capsular types of *V. vulnificus*.[42] At present, thorough cooking of seafood remains the only effective means of prevention.

VIBRIO ALGINOLYTICUS

V. alginolyticus has been predominantly associated with cellulitis and acute otitis media or externa rather than gastroenteritis.[20] These infections generally occurred after local trauma in otherwise healthy seawater from swimmers or fishermen and responded well to appropriate antibiotics.[43,44] In a review of *V. alginolyticus* infection in the United States from 1988 to 2012, there was a 12-fold increase during this period, such that it is now the second most common *Vibrio* infection. Most infections were reported from coastal states, with the highest rates from the Gulf coast.[45]

Necrotizing fasciitis has developed after a soft tissue injury from a coral reef to the leg of an apparently immunocompetent individual.[46] *V. alginolyticus* caused a pacemaker pocket infection in a man who swam off the Atlantic coast of France,[47] showing that *Vibrio* infections also can be diseases of medical progress. Isolation is similar to that for *V. vulnificus;* however, *V. alginolyticus* does not ferment lactose.

HALOPHILIC VIBRIOS

Several other *Vibrio* species are recognized as causative agents of human disease acquired through ingestion of contaminated seafood or contact of traumatized skin with seawater or brackish water. *Grimontia hollisae* (formerly *V. hollisae*) primarily causes a moderate to severe diarrheal illness, often requiring hospital admission.[1,48,49] *Photobacterium damselae* (formerly *V. damselae*) causes serious wound infections, with findings reminiscent of the clinical picture of *V. vulnificus*.[50,51] Such cases were previously reported with exposures in warmer southern waters; a fatal case with an extremely aggressive necrotizing fasciitis due to *P. damselae* was reported in New England.[52] *V. fluvialis* previously was primarily associated with sporadic gastroenteritis; more recently, it has caused peritonitis (one case of which was in association with continuous ambulatory peritoneal dialysis)[53,54] and severe hemorrhagic cellulitis.[55] Of potentially greater significance is the finding of carbapenem resistance in clinical isolates of *V. fluvialis* from persons with diarrhea in India. These isolates produced New Delhi metallo-β-lactamase 1 (NDM-1) and were resistant to nearly all antibiotics tested except azithromycin and doxycycline.[56]

V. furnissii has been rarely isolated in sporadic cases of diarrhea.[14] However, a nonfatal case in a patient with poorly controlled diabetes described *V. furnissii* bacteremia and skin lesions similar to those in *V. vulnificus* infection.[57] *V. metschnikovii* has been a rare cause of severe infections, including pneumonia, cellulitis, and bacteremia[58–60]; *V. cincinnatiensis* has caused bacteremia and meningitis.[61] *V. harveyi* (formerly *Vibrio carchariae*) has caused cellulitis after a shark bite[62] and sepsis in a pediatric patient with a central venous catheter.[63]

NONHALOPHILIC VIBRIOS: NON-O1/O139 *VIBRIO CHOLERAE* AND *VIBRIO MIMICUS*

Vibrios that are biochemically similar to *V. cholerae* but that do not agglutinate in *V. cholerae* O1 or O139 antiserum are taxonomically included in the species *V. cholerae* and are referred to as non-O1/O139 *V. cholerae*. *V. mimicus* is closely related to non-O1 *V. cholerae* but differs biochemically in being sucrose negative and Voges-Proskauer reaction negative. These nonhalophilic vibrios require only trace amounts of sodium chloride for growth in culture medium; this characteristic distinguishes them from the true halophilic vibrios *V. parahaemolyticus, V. vulnificus,* and *V. alginolyticus,* which require higher concentrations of sodium chloride in culture media and have the remarkable ability to grow in 10% sodium chloride.

Epidemiology

V. cholerae organisms are worldwide in distribution and ubiquitous in seawater and estuarine water sources, but they also have been isolated from fresh water. The burden of disease from these strains has been in the severity of illness in individual patients. They have not been observed to cause sweeping epidemics, as have *V. cholerae* O1 and O139, although a few non-O1 strains have caused outbreaks. The molecular basis for this epidemiologic behavioral difference resides in a large (39.5-kb) *Vibrio* pathogenicity island (VPI) that contains gene clusters responsible for cholera toxin acquisition and expression as well as sequences involved in colonization.[64] The group of virulence genes within the VPI is strongly tied to epidemic ability. The VPI is present in epidemic and pandemic *V. cholerae* O1 strains, in Bengal O139, and in two non-O1 *V. cholerae* strains that caused outbreaks. The VPI was absent in sporadic diarrheal and nontoxigenic environmental isolates of non-O1 *V. cholerae*.[64] Horizontal gene transfer of this pathogenicity island may be an initial step for the acquisition of epidemic capability by non-O1 *V. cholerae* strains.

In temperate regions, vibrio infections are usually seasonal; the higher number of cases in summer months is associated with warmer sea surface temperatures (>15°C) that promote growth of vibrio species. Northern Europe has seen an increase in vibrio infections (despite being nonreportable), particularly in years with heat waves.[65] There was a substantial increase in non-O1/O139 *V. cholerae* infections in Sweden and Finland in 2014, a year that had the highest (Sweden) and second highest (Finland) recorded temperatures in >150 years. The cases, negative for cholera toxin, were reported from within 100 miles of the Arctic Circle.[66]

Clinical Manifestations

Non-O1 *V. cholerae* organisms produce a wide spectrum of diarrheal illness, ranging from severe watery diarrhea indistinguishable from that with cholera to the milder traveler's diarrhea of the type commonly associated with enterotoxigenic *E. coli.* Some clinical isolates of non-O1 *V. cholerae* produce cholera toxin, but most are nontoxigenic. No clinical features distinguish the severe diarrheal illnesses caused by enterotoxin-producing non-O1 *V. cholerae* from those caused by classic *V. cholerae.* This was aptly illustrated in a report of eight sporadic cases of severe diarrhea with dehydration in the southeastern United States; *V. cholerae* serotype O75 was isolated from the patients and environmental samples, and all isolates produced cholera toxin.[67] Non-O1 *V. cholerae* strains can rarely cause bacteremia, almost invariably in patients with liver disease[68,69] but occasionally also in healthy people.

V. mimicus has caused sporadic cases of acute diarrheal illness in the United States and the tropics. Illness was associated with ingestion of raw seafood, including turtle eggs[70] and crayfish.[71]

Laboratory Findings

With intestinal infections caused by both non-O1 *V. cholerae* and *V. mimicus,* the diarrheal fluid varies from the watery isotonic fluid characteristic of cholera gravis to loose stools in which small numbers of leukocytes and erythrocytes may be seen. The organisms are readily identified on TCBS agar, on which *V. cholerae* appears as opaque yellow colonies and *V. mimicus* shows as green colonies; final speciation is made by biochemical tests and lack of agglutination in O1 antisera.

Therapy

Other than oral fluid and electrolyte repletion, no treatment is required by the large majority of patients with diarrheal disease, and antimicrobial agents have not been shown to shorten the clinical course. In occasional patients, especially those in the developing world, the intestinal fluid loss is sufficient to require intravenous electrolyte therapy. In this situation, therapy is guided by the same principles used for the treatment of cholera.

Prevention

Because non-O1/O139 *V. cholerae* organisms exist in a variety of water sources, ranging from freshwater rivers to the oceans, purification of water sources and adequate cooking of fish and other seafood provide the only certain protection against these occasional pathogens.

Key References

The complete reference list is available online at Expert Consult.

1. Newton A, Kendall M, Vugia DJ, et al. Increasing rates of vibriosis in the United States, 1996-2010: review of surveillance data from 2 systems. *Clin Infect Dis*. 2012;54:S391–S395.
6. Nair GB, Ramamurthy T, Bhattacharya SK, et al. Global dissemination of *Vibrio parahaemolyticus* serotype O3:K6 and its serovariants. *Clin Microbiol Rev*. 2007;20:39–48.
7. Daniels NA, Ray B, Easton A, et al. Emergence of a new *Vibrio parahaemolyticus* serotype in raw oysters. *JAMA*. 2000;284:1541–1545.
8. Oliver JF, Ostroff SM. Preventing *Vibrio parahaemolyticus* infection. *JAMA*. 2001;285:42–43.
9. McLaughlin JB, Depaola A, Bopp CA, et al. Outbreak of *Vibrio parahaemolyticus* gastroenteritis associated with Alaskan oysters. *N Engl J Med*. 2005;353:1463–1470.
11. Honda T, Ilida T, Akeda Y, et al. Sixty years of *Vibrio parahaemolyticus* research. *Microbe.* 2008;3:462–466.
13. Daniels NA, MacKinnon L, Bishop R, et al. *Vibrio parahaemolyticus* infections in the United States, 1973-1998. *J Infect Dis*. 2000;181:1661–1666.
17. Qadri F, Alam MS, Nishibuchi M, et al. Adaptive and inflammatory immune responses in patients infected with strains of *Vibrio parahaemolyticus*. *J Infect Dis*. 2003;187:1085–1096.
18. Morris JG. Cholera and other types of vibriosis: a story of human pandemics and oysters on the half shell. *Clin Infect Dis*. 2003;37:272–280.
20. Dechet AM, Yu PA, Koram N, et al. Nonfoodborne *Vibrio* infections: an important cause of morbidity and mortality in the United States, 1997-2006. *Clin Infect Dis*. 2008;46:970–976.
21. Centers for Disease Control and Prevention. *Vibrio* illnesses after Hurricane Katrina—multiple states, August-September, 2005. *MMWR Morb Mortal Wkly Rep*. 2005;54:928–931.
22. Shapiro RL, Altekruse S, Hutwagner L, et al. The role of Gulf Coast oysters harvested in warmer months in *Vibrio vulnificus* infections in the United States, 1988-1996. *J Infect Dis*. 1998;178:752–759.
23. Baethge BA, West BC. *Vibrio vulnificus:* did Hippocrates describe a fatal case? *Rev Infect Dis*. 1988;10:614–615.
25. Jackson JK, Murphree RL, Tamplin ML. Evidence that mortality from *Vibrio vulnificus* infection results from single strains among heterogeneous populations in shellfish. *J Clin Microbiol*. 1997;35:2098–2101.
33. Powell JL, Wright AC, Wasserman SS, et al. Release of tumor necrosis factor alpha in response to *Vibrio vulnificus* capsular polysaccharide in in vivo and in vitro models. *Infect Immun*. 1997;65:3713–3718.
34. Alice AF, Naka H, Crosa JH. Global gene expression as a function of the iron status of the bacterial cell: influence of differentially expressed genes in the virulence of the human pathogen *Vibrio vulnificus*. *Infect Immun*. 2008;76:4019–4037.
35. Marano NN, Daniels NA, Easton AN, et al. A survey of stool culturing practices for *Vibrio* species at clinical laboratories in Gulf Coast states. *J Clin Microbiol*. 2000;38:2267–2270.
40. Chen SC, Lee YT, Tsai SJ, et al. Antibiotic therapy for necrotizing fasciitis caused by *Vibrio vulnificus:* retrospective analysis of an 8 yr period. *J Antimicrob Chemother*. 2012;67:488–493.
49. Hinestrosa F, Madeira RG, Bourbeau PP. Severe gastroenteritis and hypovolemic shock caused by *Grimontia* (*Vibrio*) *hollisae* infection. *J Clin Microbiol*. 2007;45:3462–3463.
51. Yamane K, Asato J, Kawade N, et al. Two cases of fatal necrotizing fasciitis caused by *Photobacterium damsela* in Japan. *J Clin Microbiol*. 2004;42:1370–1372.
53. Lee JY, Park JS, Oh SH, et al. Acute infectious peritonitis caused by *Vibrio fluvialis*. *Diagn Microbiol Infect Dis*. 2008;62:216–218.
59. Hansen W, Freney J, Benyagoub H, et al. Severe human infections caused by *Vibrio metschnikovii*. *J Clin Microbiol*. 1993;31:2529–2530.
60. Wallet F, Tachon M, Nseir S, et al. *Vibrio metschnikovii* pneumonia. *Emerg Infect Dis*. 2005;10:1641–1642.
61. Bode RB, Brayton PR, Colwell RR, et al. A new *Vibrio* species, *Vibrio cincinnatiensis,* causing meningitis: successful treatment in an adult. *Ann Intern Med*. 1986;104:55–56.
62. Pavia AT, Bryan JA, Maher KL, et al. *Vibrio carchariae* infection after a shark bite. *Ann Intern Med*. 1989;111:85–86.
64. Karaolis DK, Johnson JA, Bailey CC, et al. A *Vibrio cholerae* pathogenicity island associated with epidemic and pandemic strains. *Proc Natl Acad Sci USA*. 1998;95:3134–3139.
67. Tobin-D'Angelo M, Smith AR, Bulens SN, et al. Severe diarrhea caused by cholera toxin-producing *Vibrio cholerae* serogroup O75 infections acquired in the southeastern United States. *Clin Infect Dis*. 2008;47:1035–1040.
71. Centers for Disease Control and Prevention. Notes from the field: *Vibrio mimicus* infection from consuming crayfish—Spokane, Washington, June 2010. *MMWR Morb Mortal Wkly Rep*. 2010;59:1374.

References

1. Newton A, Kendall M, Vugia DJ, et al. Increasing rates of vibriosis in the United States, 1996-2010: review of surveillance data from 2 systems. *Clin Infect Dis*. 2012;54:S391–S395.
2. Marder EP, Griffin PM, Cieslak PR, et al. Preliminary incidence and trends of infections with pathogens transmitted commonly through food – Foodborne Diseases Active Surveillance Network, 10 U.S. sites, 2006-2017. *MMWR Morb Mortal Wkly Rep*. 2018;67:324–328.
3. Zen-Yoji H, Sakai S, Terayama T, et al. Epidemiology, enteropathogenicity, and classification of *Vibrio parahaemolyticus*. *J Infect Dis*. 1965;115:436–444.
4. Pan T-M, Wang T-K, Lee C-L, et al. Food-borne disease outbreaks due to bacteria in Taiwan, 1986 to 1995. *J Clin Microbiol*. 1997;35:1260–1262.
5. Okuda J, Ishibashi M, Hayakawa E, et al. Emergence of a unique O3:K6 clone of *Vibrio parahaemolyticus* in Calcutta, India, and isolation of strains from the same clonal group from Southeast Asian travelers arriving in Japan. *J Clin Microbiol*. 1997;35:3150–3155.
6. Nair GB, Ramamurthy T, Bhattacharya SK, et al. Global dissemination of *Vibrio parahaemolyticus* serotype O3:K6 and its serovariants. *Clin Microbiol Rev*. 2007;20:39–48.
7. Daniels NA, Ray B, Easton A, et al. Emergence of a new *Vibrio parahaemolyticus* serotype in raw oysters. *JAMA*. 2000;284:1541–1545.
8. Oliver JF, Ostroff SM. Preventing *Vibrio parahaemolyticus* infection. *JAMA*. 2001;285:42–43.
9. McLaughlin JB, Depaola A, Bopp CA, et al. Outbreak of *Vibrio parahaemolyticus* gastroenteritis associated with Alaskan oysters. *N Engl J Med*. 2005;353:1463–1470.
10. Makino K, Oshima K, Kurokawa K, et al. Genome sequence of *Vibrio parahaemolyticus:* a pathogenic mechanism distinct from that of V. cholerae. *Lancet*. 2003;361:743–749.
11. Honda T, Ilida T, Akeda Y, et al. Sixty years of *Vibrio parahaemolyticus* research. *Microbe*. 2008;3:462–466.
12. Jiang X, Chai T-J. Survival of *Vibrio parahaemolyticus* at low temperatures under starvation conditions and subsequent resuscitation of viable, nonculturable cells. *Appl Environ Microbiol*. 1996;62:1300–1305.
13. Daniels NA, MacKinnon L, Bishop R, et al. *Vibrio parahaemolyticus* infections in the United States, 1973-1998. *J Infect Dis*. 2000;181:1661–1666.
14. Cook DW, O'Leary P, Hunsucker JC, et al. *Vibrio vulnificus* and *Vibrio parahaemolyticus* in U.S. retail shell oysters: a national survey from June 1998 to July 1999. *J Food Prot*. 2002;65:79–87.
15. Nishibuchi M, Kaper JB. Thermostable direct hemolysin gene of *Vibrio parahaemolyticus:* a virulence gene acquired by a marine bacterium. *Infect Immun*. 1995;63:2093–2099.
16. Pace JL, Chai T-J, Rossi HA, et al. Effect of bile on *Vibrio parahaemolyticus*. *Appl Environ Microbiol*. 1997;63:2372–2377.
17. Qadri F, Alam MS, Nishibuchi M, et al. Adaptive and inflammatory immune responses in patients infected with strains of *Vibrio parahaemolyticus*. *J Infect Dis*. 2003;187:1085–1096.
18. Morris JG. Cholera and other types of vibriosis: a story of human pandemics and oysters on the half shell. *Clin Infect Dis*. 2003;37:272–280.
19. Jakabi M, Gelli DS, Torre J. Inactivation by ionizing radiation of *Salmonella enteritidis, Salmonella infantis,* and *Vibrio parahaemolyticus* in oysters (*Crassostrea brasiliana*). *J Food Prot*. 2003;66:1025–1029.
20. Dechet AM, Yu PA, Koram N, et al. Nonfoodborne *Vibrio* infections: an important cause of morbidity and mortality in the United States, 1997-2006. *Clin Infect Dis*. 2008;46:970–976.
21. Centers for Disease Control and Prevention. *Vibrio* illnesses after Hurricane Katrina—multiple states, August-September, 2005. *MMWR Morb Mortal Wkly Rep*. 2005;54:928–931.
22. Shapiro RL, Altekruse S, Hutwagner L, et al. The role of Gulf Coast oysters harvested in warmer months in *Vibrio vulnificus* infections in the United States, 1988-1996. *J Infect Dis*. 1998;178:752–759.
23. Baethge BA, West BC. *Vibrio vulnificus:* did Hippocrates describe a fatal case? *Rev Infect Dis*. 1988;10:614–615.
24. Tacket CO, Brenner F, Blake PA. Clinical features and an epidemiologic study of *Vibrio vulnificus* infections. *J Infect Dis*. 1984;149:558–561.
25. Jackson JK, Murphree RL, Tamplin ML. Evidence that mortality from *Vibrio vulnificus* infection results from single strains among heterogeneous populations in shellfish. *J Clin Microbiol*. 1997;35:2098–2101.
26. Deleted in review.
27. Park J, Lee CS. *Vibrio vulnificus* infection. *N Engl J Med*. 2018;379:4.
28. Scully EP, Earp BE, Miller AL, et al. Just a cut. *N Engl J Med*. 2016;375:1780–1786.
29. Colondner R, Chazan B, Kopelowitz J, et al. Unusual portal of entry of *Vibrio vulnificus:* evidence of its prolonged survival on the skin. *Clin Infect Dis*. 2002;34:714–715.
30. Johnston JM, Becker SF, McFarland LM. Gastroenteritis in patients with stool isolates of *Vibrio vulnificus*. *Am J Med*. 1986;80:336–338.
31. Penland RL, Boniuk M, Wilhelmus KR. *Vibrio* ocular infections on the U.S. Gulf Coast. *Cornea*. 2000;19:26–29.
32. Johnson RW, Arnett FC. A fatal case of *Vibrio vulnificus* presenting as septic arthritis. *Arch Intern Med*. 2002;161:2616–2618.
33. Powell JL, Wright AC, Wasserman SS, et al. Release of tumor necrosis factor alpha in response to *Vibrio vulnificus* capsular polysaccharide in in vivo and in vitro models. *Infect Immun*. 1997;65:3713–3718.
34. Alice AF, Naka H, Crosa JH. Global gene expression as a function of the iron status of the bacterial cell: influence of differentially expressed genes in the virulence of the human pathogen *Vibrio vulnificus*. *Infect Immun*. 2008;76:4019–4037.
35. Marano NN, Daniels NA, Easton AN, et al. A survey of stool culturing practices for *Vibrio* species at clinical laboratories in Gulf Coast states. *J Clin Microbiol*. 2000;38:2267–2270.
36. Kim HS, Kim DM, Neupane GP, et al. Comparison of conventional, nested, and real-time PCR assays for rapid and accurate detection of *Vibrio vulnificus*. *J Clin Microbiol*. 2008;46:2992–2998.
37. Lee JY, Kim SW, Kim DM, et al. Clinical usefulness of real-time polymerase chain reaction for the diagnosis of *Vibrio vulnificus* infection using skin and soft tissues. *Am J Trop Med Hyg*. 2017;97:443–446.
38. Hong GL, Dai XQ, Lu CJ, et al. Temporizing surgical management improves outcome in patients with *Vibrio* necrotizing fasciitis complicated with septic shock on admission. *Burns*. 2014;40:446–454.
39. Kim DM, Lym Y, Jang SJ, et al. In vitro efficacy of the combination of ciprofloxacin and cefotaxime against *Vibrio vulnificus*. *Antimicrob Agents Chemother*. 2005;49:3489–3491.
40. Chen SC, Lee YT, Tsai SJ, et al. Antibiotic therapy for necrotizing fasciitis caused by *Vibrio vulnificus:* retrospective analysis of an 8 yr period. *J Antimicrob Chemother*. 2012;67:488–493.
41. Liu JW, Lee IK, Tang HJ, et al. Prognostic factors and antibiotics in *Vibrio vulnificus* septicemia. *Arch Intern Med*. 2006;166:2117–2123.
42. Devi SJN, Hayat U, Powell JL, et al. Preclinical immunoprophylactic and immunotherapeutic efficacy of antisera to capsular polysaccharide-tetanus toxoid conjugate vaccines of *Vibrio vulnificus*. *Infect Immun*. 1996;64:2220–2224.
43. Schmidt U, Chmel H, Cobbs C. *Vibrio alginolyticus* infections in humans. *J Clin Microbiol*. 1979;10:666–668.
44. Opal SM, Saxon JR. Intracranial infection by *Vibrio alginolyticus* following injury in salt water. *J Clin Microbiol*. 1986;23:373–374.
45. Slifka KMJ, Newton AE, Mahon BE. *Vibrio alginoyticus* infections in the USA, 1988-2012. *Epidemiol Infect*. 2017;145:1491–1499.
46. Gomez JM, Fajardo R, Patino JF, et al. Necrotizing fasciitis due to *Vibrio alginolyticus* in an immunocompetent patient. *J Clin Microbiol*. 2003;41:3427–3429.
47. Flock F, Boutille D. Pacemaker infection due to *Vibrio alginolyticus*. *Eur J Intern Med*. 2008;19:e109–e110.
48. Morris JG, Miller HG, Wilson R, et al. Illness caused by *Vibrio damselae* and *Vibrio hollisae*. *Lancet*. 1982;1:1294–1297.
49. Hinestrosa F, Madeira RG, Bourbeau PP. Severe gastroenteritis and hypovolemic shock caused by *Grimontia* (*Vibrio*) *hollisae* infection. *J Clin Microbiol*. 2007;45:3462–3463.
50. Fraser SL, Purcell BK, Delgado B, et al. Rapidly fatal infection due to *Photobacterium* (*Vibrio*) *damsela*. *Clin Infect Dis*. 1997;25:935–936.
51. Yamane K, Asato J, Kawade N, et al. Two cases of fatal necrotizing fasciitis caused by *Photobacterium damsela* in Japan. *J Clin Microbiol*. 2004;42:1370–1372.
52. Goodell KH, Jordan MR, Graham R, et al. Rapidly advancing necrotizing fasciitis caused by *Photobacterium* (*Vibrio*) *damsela:* a hyperaggressive variant. *Crit Care Med*. 2004;32:278–281.
53. Lee JY, Park JS, Oh SH, et al. Acute infectious peritonitis caused by *Vibrio fluvialis*. *Diagn Microbiol Infect Dis*. 2008;62:216–218.
54. Ratnaraja N, Blackmore T, Byrne J, et al. *Vibrio fluvialis* peritonitis in a patient receiving continuous ambulatory peritoneal dialysis. *J Clin Microbiol*. 2005;43:514–515.
55. Huang KC, Hsu RWW. *Vibrio fluvialis* hemorrhagic cellulitis and cerebritis. *Clin Infect Dis*. 2005;40:e75–e77.
56. Chowdhury G, Pazhani GP, Sarkar A, et al. Carbapenem resistance in clonally distinct clinical strains of *Vibrio fluvialis* isolated diarrheal samples. *Emerg Infect Dis*. 2016;22:1754–1761.
57. Derber C, Coudron P, Tarr C, et al. *Vibrio furnissii:* an unusual cause of bacteremia and skin lesions after ingestion of seafood. *J Clin Microbiol*. 2011;49:2348–2349.
58. Jean-Jacques W, Rajashekaraiah KR, Farmer JJ 3rd. *Vibrio metschnikovii* bacteremia in a patient with cholecystitis. *J Clin Microbiol*. 1981;14:711–712.
59. Hansen W, Freney J, Benyagoub H, et al. Severe human infections caused by *Vibrio metschnikovii*. *J Clin Microbiol*. 1993;31:2529–2530.
60. Wallet F, Tachon M, Nseir S, et al. *Vibrio metschnikovii* pneumonia. *Emerg Infect Dis*. 2005;10:1641–1642.
61. Bode RB, Brayton PR, Colwell RR, et al. A new *Vibrio* species, *Vibrio cincinnatiensis,* causing meningitis: successful treatment in an adult. *Ann Intern Med*. 1986;104:55–56.
62. Pavia AT, Bryan JA, Maher KL, et al. *Vibrio carchariae* infection after a shark bite. *Ann Intern Med*. 1989;111:85–86.
63. Wilkins S, Millar M, Hemsworth S, et al. *Vibrio harveyi* sepsis in a child with cancer. *Pediatr Blood Cancer*. 2008;50:891–892.
64. Karaolis DK, Johnson JA, Bailey CC, et al. A *Vibrio cholerae* pathogenicity island associated with epidemic and pandemic strains. *Proc Natl Acad Sci USA*. 1998;95:3134–3139.
65. Baker-Austin C, Stockley L, Rangdale R, et al. Environmental occurrence and clinical impact of Vibrio vulnificus and Vibrio parahaemolyticus: a European perspective. *Environ Microbiol Rep*. 2010;2:7–18.
66. Baker-Austin C, Trinanes JA, Salmenlinna S, et al. Heat wave-associated vibriosis, Sweden and Finland, 2014. *Emerg Infect Dis*. 2016;22:1216–1220.
67. Tobin-D'Angelo M, Smith AR, Bulens SN, et al. Severe diarrhea caused by cholera toxin-producing *Vibrio cholerae* serogroup O75 infections acquired in the southeastern United States. *Clin Infect Dis*. 2008;47:1035–1040.
68. Safrin S, Morris JG, Adams M, et al. Non-O1 *Vibrio cholerae* bacteremia: case report and review. *Rev Infect Dis*. 1988;10:1012–1017.
69. Ko WC, Chuang YC, Huang GC, et al. Infections due to non-O1 *Vibrio cholerae* in southern Taiwan: predominance in cirrhotic patients. *Clin Infect Dis*. 1998;27:774–780.
70. Campos E, Bolanos H, Acuna MT, et al. *Vibrio mimicus* diarrhea following ingestion of raw turtle eggs. *Appl Environ Microbiol*. 1996;62:1141–1144.
71. Centers for Disease Control and Prevention. Notes from the field: *Vibrio mimicus* infection from consuming crayfish—Spokane, Washington, June 2010. *MMWR Morb Mortal Wkly Rep*. 2010;59:1374.

216 *Campylobacter jejuni* and Related Species

Ban Mishu Allos, Martin J. Blaser, Nicole M. Iovine, and Beth D. Kirkpatrick

SHORT VIEW SUMMARY

Definition

- *Campylobacter* spp. are small, curved, gram-negative rods that cause acute gastrointestinal illness that typically lasts from 1 to 7 days. The diarrhea is frequently bloody and is associated with abdominal pain. Postinfectious sequelae occur infrequently. *C. jejuni* is the most common human clinical isolate.

Epidemiology

- *Campylobacter* spp. are found in a variety of animals and are a common cause of human diarrheal disease worldwide. In developed nations, diarrhea occurs in all ages, whereas in low- and middle-income countries, symptomatic illness occurs mostly in young children. Consumption or handling of poultry is a common mode of transmission.

Microbiology

- Many, but not all, *Campylobacter* spp. thrive at 42°C. The gram-negative bacilli are small (0.3–0.6 µm in diameter) and motile. They are microaerophilic and grow best at an oxygen concentration of 5% to 10%.

Diagnosis

- The gold standard for diagnosis is isolation of the organism from a stool or occasionally a blood culture. However, many highly sensitive, non–culture based techniques are now used for diagnosis by clinical microbiology laboratories.

Therapy

- The most important tenet of treatment is to replace fluid and electrolytes to prevent dehydration. Antibiotics may shorten the duration of symptoms. Azithromycin 500 mg orally daily for 3 days will treat most infections.

Prevention

- The best way to prevent infection is by thoroughly cooking poultry and other foods of animal origin and taking other measures to ensure food and water are not contaminated with animal feces. No effective vaccine is currently available.

Campylobacteriosis refers to the group of infections caused by gram-negative bacteria of the genus *Campylobacter.* Among the most common bacterial infections of humans in all areas of the world, *Campylobacter* spp. cause both diarrheal and systemic illnesses and may be associated with long-term sequelae. Infection of domesticated animals with *Campylobacter* is widespread. The name *Campylobacter* is derived from the Greek *campylos,* meaning "curved", and *baktron,* meaning "rod". After the recognition of *Campylobacter jejuni* as a major human pathogen, numerous related *Campylobacter, Arcobacter,* and *Helicobacter* species have been identified. The solving of the first *Campylobacter jejuni* genomic sequence in 2000 opened new doors in our understanding of these organisms.[1] Whole-genome sequences for multiple *Campylobacter* spp. have been determined[2] and are available on various websites.

MICROBIOLOGY

Campylobacter organisms are motile, non–spore-forming, comma-shaped, gram-negative rods.[3] Originally isolated from aborted sheep fetuses in 1909, these and similar organisms were considered subspecies of *Vibrio fetus.* However, because these organisms did not ferment carbohydrates and differed in their guanine plus cytosine (G + C) DNA content from true members of the genus *Vibrio,* a new genus, *Campylobacter,* was created. Fourteen species have been recognized within the genus; however, in recent years, taxonomic studies have indicated that splitting the genus is more appropriate.[4] The genus *Arcobacter* has been created, which now includes *Arcobacter butzleri* and *Arcobacter skirrowi.*[5] *Helicobacter cinaedi* and *Helicobacter fennelliae* had been named *Campylobacter cinaedi* and *Campylobacter fennelliae* when first discovered.[6] Although transfer to the genus *Helicobacter* is more appropriate on taxonomic grounds, because these two species cause intestinal rather than gastric illnesses they are discussed in this chapter. *Helicobacter pylori,* previously named *Campylobacter pylori,* is discussed in Chapter 217. It is clear that new members of *Campylobacter* and related genera are being identified with regularity[7–9] and that many of these will be found to be human pathogens.

Table 216.1 lists the *Campylobacter* and related species most commonly associated with human disease and indicates the differentiating characteristics. Certain species, such as *Campylobacter nitrofigilis* and *Arcobacter cryaerophilus,* have not yet been associated with human illness. In contrast, the "nitrate-negative" campylobacters are associated with diarrheal illnesses, but the appropriate nomenclature for the organisms has not been determined. Two types of illnesses are directly associated with *Campylobacter* spp.: enteric and extraintestinal. For each of these illnesses, one *Campylobacter* species predominates and other species are less commonly present. The prototype for enteric infection is *C. jejuni;* for extraintestinal infection it is *Campylobacter fetus* (Table 216.2). Because the organisms causing enteric and extraintestinal illnesses are generally the same, they are considered together in the following discussion.

Campylobacters and related organisms grow best in an atmosphere containing 5% to 10% oxygen and are thus considered microaerophilic.[3,4] Although most of these organisms will not grow under aerobic or anaerobic conditions, *C. jejuni* can grow in candle jars, which permits isolation when the optimal atmosphere cannot be achieved. All campylobacters grow at 37°C; however, *C. jejuni* grows best at 42°C. Because *C. jejuni* is the most common enteric pathogen of humans, many laboratories have used incubation at 42°C for optimal isolation; however, use of this temperature will not permit detection of infections by many of the related species. In particular, *Campylobacter upsaliensis* may be missed.

Campylobacter spp. multiply more slowly than do the usual bacteria of the enteric flora and therefore cannot be isolated from fecal specimens unless selective techniques are used. The most common isolation methods use blood-based, antibiotic-containing media. Three such media—Skirrow medium, Butzler agar, and Campy-BAP medium—or variations of these have been in wide use.[4] The last two media contain cephalothin, which inhibits *C. fetus* and several other *Campylobacter* subspecies, but are best suited for isolating *C. jejuni.* Several enrichment broths have been developed, but because ill humans usually excrete 10^6 to 10^9 *C. jejuni* colony-forming units per gram of stool, enrichment usually is not necessary. Blood-free media can also be used.[10] Owing to their small

TABLE 216.1 Differential Characteristics of *Campylobacter* and Related Species Most Commonly Associated With Pathogenicity in Humans

	GROWTH				H_2S PRODUCTION			SUSCEPTIBILITY TO 30-μg DISK		
SPECIES	25°C	37°C	42°C	NITRATE REDUCTION	On TSI	On Lead Acetate Paper	HIPPURATE HYDROLYSIS	Cephalothin	Nalidixic Acid	C-19 FATTY ACID REDUCTION
Campylobacter jejuni	–	+	+	+	–	+	+[a]	R	S	+
Campylobacter coli	–	+	+	+	v	+	–	R	S	+
Campylobacter lari	–	+	+	+	–	+	–	R	R	+
Campylobacter fetus subsp. *fetus*	+	+	v	+	–	v	–	S	R	–
Campylobacter hyointestinalis	v	+	v	+	+	+	–	S	R	+
Helicobacter cinaedi	–	+	–	+	–	+	–	S	S	–
Campylobacter upsaliensis[b]	–	+	+[c]	+	–	+	–	S	S	–
Helicobacter fennelliae	–	+	–	–	–	+	–	S	S	–

[a]Approximately 5% to 10% of *C. jejuni* strains are hippurate negative.
[b]Catalase negative or weak.
[c]Occasional isolates fail to grow at 42°C.
–, Does not have the characteristic; +, has the characteristic; *R,* resistant; *S,* susceptible; *TSI,* triple sugar iron agar slant; *v,* variable (some strains show the characteristic).

TABLE 216.2 *Campylobacter, Helicobacter,* and *Arcobacter* Species Associated With Different Clinical Manifestations of Infection

ENTERIC DISEASE	EXTRAINTESTINAL DISEASE
Major Pathogen	**Major Pathogen**
Campylobacter jejuni	*Campylobacter fetus*
Minor Pathogens	**Minor Pathogens**
Campylobacter coli	*Campylobacter jejuni*
Campylobacter lari	*Campylobacter coli*
Campylobacter fetus	*Campylobacter lari*
Helicobacter fennelliae	*Helicobacter fennelliae*
Helicobacter cinaedi	*Helicobacter cinaedi*
Campylobacter upsaliensis	*Campylobacter sputorum*
Arcobacter butzleri	*Campylobacter hyointestinalis*
Arcobacter skirrowi	*Helicobacter rappini*
Arcobacter cryaerophilus	

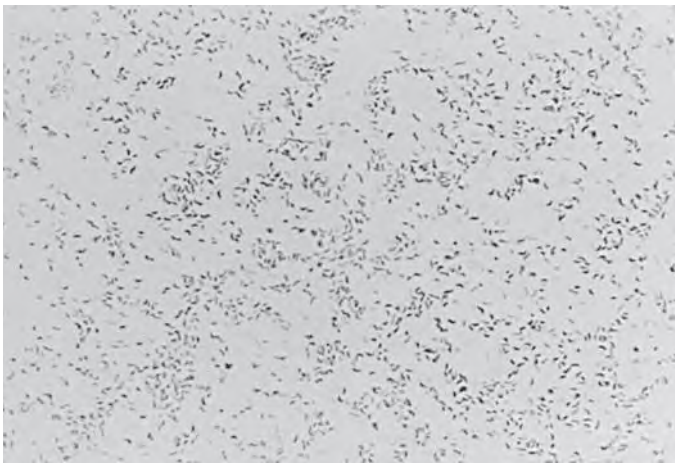

FIG. 216.1 Fine-curved, S-shaped, or spiral, lightly staining gram-negative appearance of *Campylobacter jejuni* in pure culture (×1000).

size (0.3–0.6 μm in diameter) and motility, *Campylobacter* spp. and related organisms pass through 0.45- or 0.65-μm filters that retard the usual enteric flora. Filtration methods permit isolation without use of antibiotic-containing media. It is now clear that use of filtration techniques and nonselective rich media such as chocolate agar, with incubation of plates at 37°C, improves stool culture yields of both *C. jejuni* and the "atypical" enteric *Campylobacter* spp.[11,12] The development of filtration techniques represents a significant advance over the use of selective media, and such techniques are now recommended for primary isolation of *Campylobacter* spp. from fecal specimens or swabs.

Visible colonies usually appear on the plating media within 24 to 48 hours, and usually longer for the "atypical" species. *Campylobacter* spp. can be distinguished from other microorganisms on the basis of several standard criteria and can be distinguished from one another on the basis of biochemical testing.[4,13] Organisms from young cultures have a typical vibrioid appearance (Fig. 216.1), but after 48 hours of incubation, organisms appear coccoid. The ability to hydrolyze hippurate distinguishes *C. jejuni* from most other members of the genus, but hippurate-negative *C. jejuni* isolates also occur. Isolation of organisms from sites without a normal microbiota, such as the bloodstream, is not difficult, although when *Campylobacter* is the suspected pathogen, incubation of cultures should be extended to 2 weeks. With radiometric detection systems, turbidity of the medium may not be present, and the increase in released radiolabeled substance may be less than usually specified thresholds, reflecting suboptimal conditions for certain of these organisms.[14] State-of-the-art identification to the species level should include polymerase chain reaction studies of 16S recombinant RNA (rRNA) or other targets for comparison with known species.[15–17] As discussed later (see "Diagnosis"), these techniques have been developed for culture confirmation and for typing of strains and may be more sensitive than traditional culture methods.[18–20]

As with other bacteria whose ecologic niche is the gastrointestinal tract of mammals and avian species, the serotypical diversity of *C. jejuni* is enormous. More than 90 different serotypes based on somatic (O) antigens and 50 different serotypes based on heat-labile (capsular and flagellar) antigens have been identified[4]; phase variation of flagellar

antigens occurs. O-antigen variation reflects the presence of differing genetic cassettes that contain the enzymes for O-antigen formation. No group somatic or flagellar antigen has been identified; however, several superficial proteins appear to have broad serotypical specificity, a factor that may aid in the development of a broadly specific vaccine.

C. jejuni cannot long withstand drying or freezing temperatures, which are characteristics that limit its transmission.[21] However, *C. jejuni* survives in milk or other foods or in water kept at 4°C for several weeks. Pasteurization effectively destroys the organism, as does chlorine at concentrations in standard use for water disinfection.

EPIDEMIOLOGY

Campylobacteriosis is a worldwide zoonosis. *Campylobacter* spp. are commonly found as commensals of the gastrointestinal tract in wild or domesticated cattle, sheep, swine, goats, dogs, cats, rodents, and all varieties of fowl.[3,21] *C. jejuni* has a very varied reservoir, but *Campylobacter coli* and *Campylobacter hyointestinalis* are most commonly isolated from swine, and *C. upsaliensis* is most commonly isolated from dogs.[22,23] *C. fetus* subsp. *fetus* has been isolated from sheep, cattle, poultry, reptiles, and swine.[3] Primary acquisition of *Campylobacter* spp. by animals often occurs early in life and may lead to morbidity or mortality, but in most colonized animals a lifelong carrier state develops. The vast reservoir in animals is probably the ultimate source for most enteric *Campylobacter* infections in humans. Meats originating from infected animals frequently become contaminated with intestinal contents during the slaughtering process.[21] In particular, commercially raised poultry is nearly always colonized with *C. jejuni,* slaughterhouse procedures amplify contamination, and chicken and turkey in supermarkets, ready for consumers to take home, frequently are contaminated.[21,24] Excreta from infected animals may contaminate soil or water. Most infections in humans probably result from consumption of contaminated food and water. Investigations of more than 50 outbreaks indicate that unpasteurized (raw) milk is such a vehicle.[21,25] Similarly, untreated surface water has been responsible for both endemic and epidemic campylobacteriosis. Backpackers in Wyoming who drank untreated water and developed acute diarrheal illnesses had three times more *Campylobacter* infections than *Giardia* infections.[26] Several large outbreaks have been traced to defects in municipal water systems.[27,28] Undercooked meats, especially poultry, have been associated with infection.[29–33] Other vehicles include raw clams, raw or undercooked beef, and unpasteurized cheeses and goat's milk. In one US study, children riding in grocery store shopping carts next to raw meat or poultry had higher rates of *Campylobacter* infections.[34,35] Nevertheless, consumption of undercooked poultry is estimated to be responsible for 50% to 70% of sporadic *Campylobacter* infections in developed countries. Increases in the isolation of *Campylobacter* spp. reflect both improved recognition and increased consumption of poultry in recent years.

Direct contact with infected animals may result in transmission. Household pets, especially young dogs and cats with diarrhea, have been implicated as vectors for campylobacteriosis.[19,22] In 2017, a large multistate US outbreak of multidrug-resistant *C. jejuni* infections occurred in puppies and was transmitted to more than 100 people.[36] Because healthy dogs, cats, rodents, and birds may excrete *Campylobacter* and related organisms, it is not surprising that human infections associated with these animals also have been reported. People with occupational exposure to cattle, sheep, and other farm animals are at increased risk for infection, and laboratory-acquired infections have been reported. *C. fetus* strains in reptiles and mammals probably diverged 200 million years ago[37]; however, humans may become infected with reptile strains, possibly due to consuming a food of reptile origin. Most reported strains in the United States and elsewhere have been from peoples of Asian origin,[38] suggesting that some particular contaminated food is involved. The reptile isolates are sufficiently distinct that a new subspecies has been proposed: *C. fetus* subsp. *testudinum.*

As with other enteric pathogens, fecal-oral person-to-person transmission of *C. jejuni* has been reported. People in contact with the excreta of infected individuals who are not feces continent (e.g., infants) are at risk for infection. Infected school-age children rarely may transmit *Campylobacter* infection. Transmission from infected food handlers who are asymptomatic is at best uncommon. Perinatal transmission from a mother who may not have been symptomatic may be due to exposure in utero, during passage through the birth canal, or during the first days of life.[39] Infection has been associated with blood transfusion from an infected patient.[40] Because of a variety of sexual practices, homosexual men appear to be at increased risk for infection caused by *H. cinaedi, H. fennelliae,* and other "atypical" *Campylobacter* spp.[41] Human immunodeficiency virus (HIV)–infected patients are at substantially increased risk for infection.[42] The standardization of serotyping methods[43] and the development of molecular methods for identification and typing of *C. jejuni* and related organisms[15–20,44,45] should improve our understanding of transmission.

C. jejuni infections occur year-round in the United States and other developed countries but with a sharp peak in summer and early fall. *C. fetus* infections show the same seasonal variation, but the peak is less marked. The reason for this seasonal variation is unclear. In developed countries, the incidence of infection is higher when air temperatures rise.[46] Flies also have been suggested as a potential source of transmission to humans.[47] In tropical countries, the seasonal variation of *C. jejuni* infection appears to be influenced by rainfall.

For many years, the incidence of *C. jejuni* infections continued to rise in the United States and Europe and exceeded rates of *Salmonella* and *Shigella* infections combined.[48,49] Beginning in the mid-1990s, the incidence has waxed and waned but *C. jejuni* remained one of the most frequently reported pathogens that cause foodborne illness. Improved hygienic practices on farms and especially in poultry slaughterhouses may have contributed to stabilization in disease frequency. Beginning in 2014, the incidence began to increase again.[50,51] Indeed, in 2017, the most recent year reported by the Centers for Disease Control and Prevention's Foodborne Diseases Active Surveillance Network (FoodNet), the incidence of *Campylobacter* rose by 10%, climbing to 19.1 per 100,000 population.[52] Over the past decade, the incidence of *Campylobacter* infections has also increased in Europe and Israel.[53,54] The reasons for these increases are not obvious, but increasing reliance on culture-independent diagnostic tests (CIDTs) could be playing a role. In the United States, the number of *Campylobacter* cases diagnosed by CIDTs alone increased by 114% in 2016 when compared with 2013 to 2015.[55] Because CIDT methods are quicker and more readily available, clinicians may order these tests more frequently, making year-to-year comparisons difficult. As a result, changes in *Campylobacter* incidence over time and determinations of the effectiveness of prevention strategies are becoming increasingly challenging. Nevertheless, *Campylobacter* infections have always been underdiagnosed vis-à-vis infections with Enterobacteriaceae, due to their slow and fastidious nature in culture, the need for specialized culture conditions, and the importance of laboratory experience in isolating and identifying these organisms. The increasing use of CIDTs is permitting a better assessment of *C. jejuni* infections, but unless the constituents of the panels are broadened in the future, will fail to identify infections due to the less common species, further underdiagnosing these infections. Other technologic advancements, such as whole-genome sequencing of *Campylobacter* isolates, may be available in the future for use in outbreak investigations and in further characterizing the epidemiology of these infections.[56]

Based on current estimates, there are probably more than 2 million *Campylobacter* infections annually in the United States, although there is great geographic variability in incidence rates, even within the United States.[57] Population-based studies show peak incidence in children younger than age 1 year and in people 15 to 29 years old[58]; however, cases have been reported in patients of all ages. The incidence in males may be higher. The prevalence of infection in healthy people is very low (<1%).

The epidemiology of infection in developing countries is markedly different. *C. jejuni* is often isolated from healthy people, and the infection is especially common during the first 5 years of life.[59–61] During the first 2 years of life, most children have numerous *Campylobacter* infections, but those occurring early in life frequently are symptomatic, whereas later infections are mostly asymptomatic.[60] The source of these frequent infections has not been defined, but preliminary evidence suggests that human-to-human transmission may be more common than in developed countries. The substantial age-related difference in the infection-to-illness ratios in developed and developing countries

appears primarily to be due to differences in age- or exposure-related immunity of the populations rather than to differences in the isolates.[62,63] Areas where *Campylobacter* infection is most prevalent are associated with growth shortfalls among children.[64] Interestingly, even in developed nations, the incidence of infection in rural areas is higher, similar to patterns observed in developing nations, and possibly associated with more direct contact with animal vectors of the infection.[65] *C. jejuni* and other *Campylobacter* spp. are important causes for the acute diarrheal illnesses suffered by travelers.[66]

PATHOGENESIS AND PATHOLOGIC CHARACTERISTICS

The absence of a nonprimate animal model that is closely analogous to human infection makes understanding *Campylobacter* pathogenesis more difficult. However, it is clear from outbreak investigations and from volunteer studies that not all *Campylobacter* infections produce illness. Although all factors responsible for this phenomenon are not known, three of the most important appear to be the dose of organisms reaching the small intestine, the virulence of the infecting strain, and the specific immunity of the host to the pathogen ingested. Among exposed people who become ill, the incubation period varies from 1 to 7 days, a characteristic that is probably inversely related to the dose ingested. Although there are reports of illness occurring within 24 hours of exposure,[67,68] most infections occur 2 to 4 days after exposure. In one study, volunteers became ill after ingesting as few as 500 organisms, but with a dose of less than 10^4 organisms, illness was infrequent.[69,70] *C. jejuni,* like *Salmonella typhimurium,* is susceptible to hydrochloric acid.[70] Taken together, these data suggest that the infectious dose for *C. jejuni* is similar to that for *Salmonella.* The acidic milieu of the stomach provides an effective barrier against *Campylobacter* infection. However, vehicles such as milk, fatty foods, and water that favor passage through the gastric acid barrier may permit some infections to occur at relatively low doses. Similarly, patients who use proton pump inhibitors or histamine type 2 blockers are more susceptible to infection.[71,72] Also, some *Campylobacter* spp. appear to be well adapted to survival outside animal hosts and are more resilient to physical stresses[73]; these strains may be more available to infect humans.

C. jejuni multiplies in human bile[70] a characteristic that aids colonization of the bile-rich upper small intestine early in infection. The sites of tissue injury include the jejunum, ileum, and colon, with similar pathologic features in each. Inspection of affected tissues may reveal a diffuse, bloody, edematous, and exudative enteritis,[74] but pathologic examinations are generally only performed on specimens from patients with the most severe cases. Microscopic examination of rectal biopsy specimens has shown a nonspecific colitis with an inflammatory infiltrate of neutrophils, mononuclear cells, and eosinophils in the lamina propria; degeneration, atrophy, loss of mucus, and crypt abscesses in the epithelial glands; and ulceration of the mucosal epithelium.[75,76] Rectal biopsy samples with these nonspecific features have been interpreted as showing acute ulcerative colitis or Crohn disease. In other cases, the appearance of the rectal biopsy sample has been similar to that of specimens obtained in *Salmonella* or *Shigella* infections. In a series of 124 patients with *C. jejuni* infection, 18 of the most severely ill patients underwent sigmoidoscopic examination or rectal biopsy; 17 of these procedures showed colonic involvement.[77] Some patients have terminal ileitis as well as colitis. Unspecified host factors are also clearly important; in volunteers, a single strain produced a wide spectrum of clinical manifestations.[69]

The presence of bacteremia in some patients, the finding of cellular infiltration in biopsy specimens, and the presence of blood in stools from patients with *Campylobacter* colitis also suggest that tissue invasion occurs. The process of *C. jejuni* invasion is multifactorial. Some evidence suggests *C. jejuni* breaches epithelial cell barriers via a paracellular route by disruption of tight junctions.[78] Other evidence exists for basolateral and apical transcellular routes of invasion.[79] Invasion requires microtubule-dependent mechanisms,[80,81] and subsequently *Campylobacter* organisms are contained within a compartment that does not fuse with lysosomes but allows release of the organisms to the basolateral side of the epithelial cell and underlying lamina propria.[82] Subsequent detection of the organisms by the host immune system leads to an influx of inflammatory cells and cytokines/chemokines and to tissue destruction, which ultimately produce the disease signs and symptoms. The bacteria's flagellae are also important virulence factors because they promote the motility and chemotaxis needed for *C. jejuni* to colonize the intestinal tract.[83,84] The bacterial flagellar export apparatus is involved in the secretion of a number of proteins that affect invasion.[85–87] Some secreted proteins produce rapid apoptotic death of cell cultures[88]; others may modulate the immune response to favor bacterial survival.[89] However, the role of these proteins in causing diarrhea is not known. Unlike other enteric pathogens such as *Salmonella, Campylobacter* flagellins are not recognized by the host pattern recognition receptor Toll-like receptor (TLR) 5 due to differences in specific regions of flagellin genes, and do not elicit production of the proinflammatory cytokine interleukin-8, suggesting that flagellins are involved in evasion of innate immune responses in their reservoir hosts.[90,91]

Unlike other pathogens such as *Escherichia coli*, the *C. jejuni* genome does not encode any conventional known enterotoxins.[1] Some strains express the cytolethal distending toxin (Cdt), named for its effect on mammalian cell lines,[91] but its role in pathogenesis is unclear because it is not always present in strains isolated from patients with diarrhea.[92] However, this toxin may affect cell cycle kinetics[93,94] and may play a role in suppressing innate immunity by inducing death of macrophages.[95,96]

A high-molecular-weight plasmid (pVir) encodes proteins involved in secretion and also enhances the invasive capabilities of *C. jejuni* virulence.[97,98] It has been identified in some clinical *Campylobacter* isolates and has been significantly associated with bloody stools, although strains lacking pVir may remain virulent.[99,100] Several epithelial cell adhesins have been identified in *C. jejuni.* The superficial antigen (PEB1) that appears to be the major adhesin[101] and is conserved among *C. jejuni* strains also is a target of the immune response,[102] and may represent a vaccine candidate.[103,104] Other important adhesins include JIpA, a surface-exposed lipoprotein,[105] and CadF, which mediates adhesion by binding to fibronectin.[106] Acquisition of ferrous and ferric iron in the gut is critical for colonization by *C. jejuni,* and the molecules involved in the process may be considered as virulence factors and targets for interventions as well.[107]

Campylobacter outer membranes contain lipopolysaccharides (LPSs) with typical endotoxic activity.[108–110] The structure of the LPS O antigen is highly variable.[109,110] Many *C. jejuni* O antigens possess sialic acid–containing structures that are recognized by host immune cells bearing sialoadhesins.[111–113] The close resemblance of these sialylated structures to those seen in human gangliosides such as GM1, GD1a, GD3, and GT1a and their presence in strains isolated from patients who developed the Guillain-Barré syndrome (GBS) support a role in the pathogenesis of this disorder, via molecular mimicry.[111,112,114] Certain capsular genotypes are also associated with the sialylated lipooligosaccharide structures implicated in GBS, raising the possibility that capsule also may contribute to GBS pathogenesis.[115] Sialylated lipooligosaccharides confer resistance to host cationic antimicrobial peptides and proteins[116] and may also be associated with more severe gastrointestinal disease.[117]

Bacteremia can sometimes be detected in patients with *Campylobacter* infections, whether or not they show signs of systemic illness. Most bacteremias reported to the Centers for Disease Control and Prevention have been due to *C. fetus* subsp. *fetus,* whereas *C. jejuni* is by far the more common pathogen overall. One explanation for the apparently greater tendency of *C. fetus* to cause bacteremia is that it is usually resistant to the bactericidal activity present in normal human serum.[118] Important risk factors for *C. fetus* bacteremia and meningitis include immunosuppression and occupational exposure to infant animals that are the reservoir hosts for *C. fetus.*[119,120] *C. fetus* is covered with a surface (S)–layer protein that functions as a capsule.[121,122] Virtually all human isolates of *C. fetus* possess an S-layer protein that completely disrupts C3b binding to these organisms.[123] Lack of C3b binding explains both serum and phagocytosis resistance.[120] *C. fetus* also has the ability to change the major S-layer protein expressed. This results in antigenic variation[124] and is facilitated by recombination among several highly homologous genes encoding full-length proteins.[125,126] The S-layer protein of *C. fetus* is the major virulence factor explaining its extraintestinal spread (Fig. 216.2).

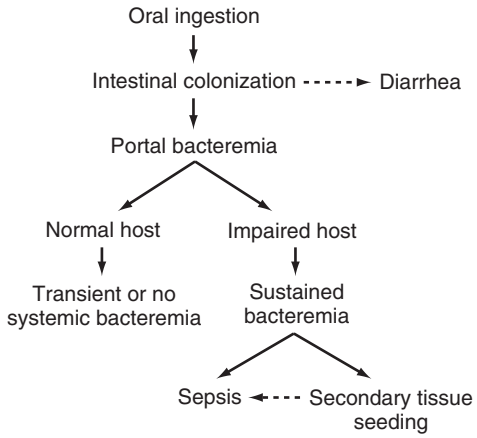

FIG. 216.2 Pathogenesis of *Campylobacter fetus* infections. *(From Blaser MJ. Campylobacter fetus: emerging infection and model system for bacterial pathogenesis at mucosal surfaces.* Clin Infect Dis. *1998;27:256–258.)*

IMMUNOLOGY

The adaptive human immune responses responsible for protection and recovery from *Campylobacter* infection are not completely understood. Immunocompetent individuals who develop *C. jejuni* diarrheal disease usually resolve the diarrhea and become asymptomatic without antibiotic treatment. Protection from recurrent disease may not develop following a single infection, as has been shown in human experimental infection and reinfection models of *C.jejuni*[127]; early antibiotic treatment might thwart the development of a full immune response to the infection. Patients infected with *Campylobacter* spp. develop specific immunoglobulin (Ig) G, IgM, and IgA antibodies in serum[128,129] and IgA antibodies in intestinal secretions.[129,130]

In low-income countries, where *C. jejuni* infection is hyperendemic, serum IgA levels rise progressively with age, and the decreasing case-to-infection ratio with age suggests acquisition of protective immunity following multiple exposures. Further supporting the importance of humoral immunity are multiple reports of severe and recurrent *C. jejuni* infection in patients with congenital or acquired hypogammaglobulinemia.[131,132] In HIV-infected patients as well, failure of *C. jejuni* infection to respond to antimicrobial therapy has been correlated with failure to produce a humoral response to infection.[131] Overall, patients with immunodeficiency from HIV have a markedly increased incidence of *C. jejuni,*[42,133] although the role of cellular immunity is not well understood. The heightened incidence is consistent with an epidemiologic model of wide circulation of the organism in human foods, but most often at low doses to which immunocompetent hosts are not susceptible, but to which immunocompromised hosts are.

Components of the innate immune response have been demonstrated to both limit infection and trigger inflammation. *C. jejuni* is serum sensitive, allowing for complement-mediated killing as well as opsonization followed by phagocytosis.[134] Phagocytes, intestinal epithelial cells, and many other cells express the key pattern recognition receptors TLRs 2 and 4, which recognize cell wall components and LPS/lipooligosaccharide. Engagement of these receptors triggers proinflammatory signaling. This leads to the production of inflammatory cytokines (e.g., interleukin-6, tumor necrosis factor-α) and chemokines (e.g., interleukin-8) that assist in the recruitment and activation of host cells that kill *C. jejuni* directly via neutrophilic phagocytosis, for example.[135,136] Unlike most other enteric pathogens, the flagellin of *C. jejuni* is not recognized by TLR5, and TLR9 is poorly activated by *C. jejuni* as well, suggesting these are means of immune evasion.[137,138] Nonetheless, *C. jejuni* is susceptible to killing by cationic antimicrobial agents, including α- and β-defensins produced by intestinal epithelial cells and phagocytes.[139]

As discussed earlier, one important virulence factor in *C. fetus* that is not found in *C. jejuni* is a proteinaceous paracrystalline array attached to the O antigen of LPS known as an S layer. This renders *C. fetus* serum resistant, owing to the inability of the complement component C3b to opsonize its surface.[140] Patients with *C. fetus* infections much more frequently have evidence of impaired immunity, including conditions such as chronic alcoholism, liver disease, old age, diabetes mellitus, and malignancies,[141,142] permitting an organism that a priori can resist innate immunity to take hold.

CLINICAL MANIFESTATIONS

Campylobacter jejuni Infections

The clinical manifestations of infections caused by all of the *Campylobacter* spp. that cause enteric illnesses appear identical; *C. jejuni* infection may be regarded as the prototype. Acute enteritis is the most common presentation of *C. jejuni* infection. Symptoms may last from 1 day to 1 week or longer. Often, there is a prodrome with fever, headache, myalgia, and malaise 12 to 24 hours before the onset of intestinal symptoms.[143] In some patients, the constitutional symptoms may coincide with the intestinal phase or, less often, may follow it. The most common symptoms are diarrhea, malaise, fever, and abdominal pain.[143–145] Diarrhea may range in severity from loose stools to massive watery or grossly bloody stools. In any patient, the entire spectrum of diarrhea may be seen. For most patients, there are 10 or more bowel movements on the worst day of the illness. Abdominal pain is usually cramping and is relieved by defecation; it may be the predominant manifestation of illness. *Campylobacter* enteritis is frequently self-limiting, with a gradual resolution of symptoms over several days; however, illness lasting longer than 1 week occurs in 10% to 20% of patients seeking medical attention, and relapse may be seen in another 5% to 10% of patients who do not receive treatment.[44,143–146]

Infection also may be manifested as an acute colitis, with symptoms of fever, abdominal cramps, and bloody diarrhea persisting for 1 week or longer.[75,145] Fever may be low grade or consist of daily peaks above 40°C (104°F). Initially, stools may be watery, but as the illness progresses they may become frankly bloody; tenesmus is a common symptom. In the most severe forms, patients appear very ill, and toxic megacolon has been reported.[147] Because of the propensity of *Campylobacter* infection to affect young adults and the characteristic clinical presentation, it may be readily confused with ulcerative colitis or Crohn disease.[75,143] The pathologic findings on rectal biopsy are nonspecific, and the clinical features and radiographic findings are also nondiagnostic. Therefore the clinician should have a high index of suspicion for *Campylobacter* infection in a patient who presents with this symptom complex. Because of the often fastidious nature of these organisms,[148,149] a single negative culture does not rule out infection, especially if optimal filtration methods are not used for primary isolation of a pathogen.

Occasionally, acute abdominal pain may be the major or only symptom of infection.[150] Although any quadrant of the abdomen may be affected, patients most often complain of pain in the right lower quadrant. As with *Yersinia enterocolitica* and *Salmonella enteritidis, C. jejuni* may cause pseudoappendicitis.[44,143] In most cases, the removed appendix has shown minimal or no inflammation. Enlarged mesenteric nodes (mesenteric adenitis) and terminal ileitis[44] also may be responsible for symptoms. Diagnosis is often made during the postoperative period, when diarrhea ensues. *Campylobacter* infection occasionally may present solely as a gastrointestinal hemorrhage.[151] Among neonates, *C. jejuni* infection may be manifested as one or more grossly bloody stools and no other symptoms, with findings suggesting intussusception,[152] or with extraintestinal foci.[153] Fever also may be the sole manifestation of *C. jejuni* infection. Temperature elevation may be so severe and persistent that typhoid fever is the initial diagnosis until *C. jejuni* is isolated from stools. Febrile convulsions in young children before the onset of the enteric phase of illness also may occur.[154]

Bacteremia has been noted in less than 1% of patients with *C. jejuni* infection. In part, this low frequency reflects the fact that physicians rarely perceive diarrheal illness as an indication for blood culture, even when fever is present. Nevertheless, bacteremia appears to be more common in infections in people at the extremes of age.[58,155,156] Meningitis and endocarditis are rare manifestations of *C. jejuni* infection. In general, three patterns of extraintestinal *C. jejuni* infection have been noted.[157] First, there may be a transient bacteremia in a normal host with acute *Campylobacter* enteritis. The bacteremia may be discovered several days after blood cultures are obtained, by which time the patient usually has completely recovered. The course is benign, and no specific treatment

based on the positive blood culture result is usually indicated. Second, there may be a sustained bacteremia or deep focus of infection in a previously normal host; usually the patient has an acute enteritis as well. The *C. jejuni* isolates are generally relatively or absolutely serum resistant.[157] Bacteremia usually has its origin in the intestinal tract inflammation and responds to antimicrobial therapy. Third, sustained bacteremia or deep infection may occur in an immunocompromised host; many such patients do not have an acute enteritis. *C. jejuni* isolates are usually serum sensitive.[157] However, as with other gram-negative bacteria, *C. jejuni* bacteremia may produce fever and shock.[158] Antimicrobial therapy, which may need to be prolonged, is required for elimination or suppression of this infection.

C. jejuni may cause septic abortion,[159] but sustained bacteremia in a pregnant patient does not necessarily imply fetal infection or a bad outcome.[157] There have been infrequent reports of *C. jejuni* infections manifesting as acute cholecystitis,[160] pancreatitis,[161] and cystitis.[162] People with immunoglobulin deficiencies often develop prolonged, severe, and recurrent *C. jejuni* infections,[131,132] often with bacteremia and other extraintestinal manifestations such as erysipelas-like skin lesions or osteomyelitis.[163]

Long-term colonization by immunocompromised hosts, such as common variable immunodeficiency, has been recognized and may last for years.[164] Symptomatic and asymptomatic recrudescence of *C. jejuni* infection following appropriate antibiotics in healthy adults is also a newly recognized phenomenon, and suggests that *C. jejuni* both may be shed in the feces for longer than recognized postinfection and may persist in the host in a protected site such as intestinal biofilms.[165]

C. jejuni infections have been associated with multiple postinfectious sequelae, especially GBS but also reactive arthritis, irritable bowel syndrome, and immunoproliferative disorders of the small intestine. GBS is a strongly associated but uncommon consequence of *C. jejuni* infection (estimated at 1 case per 2000 infections) that usually occurs 2 or 3 weeks after the diarrheal illness.[166,167] From 20% to 50% of GBS cases follow *C. jejuni* infections, reflecting in part the high incidence of these infections.[166–170] A particular group of *C. jejuni* organisms marked by LPS serotype O:19 is overrepresented among people who develop GBS.[170–172] A recent study demonstrated that *C. jejuni* O:19 chaperone proteins share high primary sequence homology with heat shock proteins found on human peripheral nerves; these structures as well as capsular and LPS glycolipids may be involved in the molecular mimicry triggering GBS.[173] Serotype O:41 has also been implicated, and other sporadic cases may be due to specific *C. jejuni* strains with sialylation of their LPS molecules.[174–176] Postinfectious reactive arthritis may occur up to several weeks after infection, and prolonged rheumatic symptoms have also been reported. The relation of this phenomenon to the presence of HLA-B27 histocompatibility antigens is not clear.[177–179] The organism was detected by polymerase chain reaction assay in small intestinal biopsy specimens from patients with mucosa-associated lymphoid tissue (MALT),[180] analogous to the role of *H. pylori* in gastric MALT lymphomas. Myopericarditis,[181,182] hepatitis,[183] cellulitis,[184] interstitial nephritis, the hemolytic-uremic syndrome, and IgA nephropathy[185] are other reported complications.

Campylobacter fetus Infections

Although *C. fetus* may occasionally cause diarrheal disease, it has the propensity to cause dissemination, including bacteremia, endovascular infections, and cellulitis, and may do so in the absence of intestinal symptoms.[141,186] As summarized in Table 216.3, the clinical, laboratory, and epidemiologic characteristics of *C. jejuni* infections differ significantly from those of *C. fetus* subsp. *fetus,* which often produce systemic manifestations. *C. fetus* infections may cause intermittent diarrhea or nonspecific abdominal pain without localizing signs. The diarrheal illness may manifest exactly like *C. jejuni* infection and is more common than was suspected several years ago. Clinical manifestations are similar and sequelae uncommon. Nearly all affected patients survive the infections when appropriate antibiotic treatment is given and usually do well without antibiotic treatment. *C. fetus* also may cause a prolonged relapsing illness characterized by fever, chills, and myalgias, in which a source of infection cannot be demonstrated.[142,187] Occasionally, secondary seeding of an organ will occur, leading to a more complicated infection[187,188] and sometimes to a fulminant, fatal course.

TABLE 216.3 Biologic and Clinical Characteristics of *Campylobacter jejuni* and *Campylobacter fetus* subsp. *fetus*

FEATURE	*CAMPYLOBACTER JEJUNI*	*CAMPYLOBACTER FETUS* SUBSP. *FETUS*
Epidemiologic Characteristics		
Major reservoir	Avian species, food animals	Cattle and sheep (reptiles)
Affected hosts	Normal hosts; all ages affected; often in clusters of cases	Opportunistic agent in debilitated hosts; clustering rare; healthy hosts may be affected
Laboratory Characteristics		
Range of growth temperatures	32°–42°C	25°–37°C[a]
Usual source of isolation	Feces	Bloodstream
Clinical Characteristics		
As a cause for diarrheal illness	Common	Uncommon
Clinical manifestations	Acute gastroenteritis, colitis	Systemic illness with bacteremia, meningitis, vascular infections, abscesses; gastroenteritis
Outcome of infection	Usually self-limited	May be fatal in debilitated hosts

[a]Occasionally grows at 42°C.

C. fetus infections appear to have a predilection for vascular sites; vascular necrosis occurs in patients with endocarditis and pericarditis resulting from this organism.[189–191] Mycotic aneurysms of the abdominal aorta and, rarely, peripheral arteries also occur.[192] Thrombophlebitis may be associated with *C. fetus* bacteremia, but whether it is the primary event or a secondary manifestation of the infection is uncertain. Patients with a bacteremic illness without localization should be carefully evaluated for the presence of septic thrombophlebitis, because the response is good when this condition is treated with appropriate antibiotics. Infections during pregnancy primarily have been manifested as upper respiratory tract symptoms, pneumonitis, fever, and bacteremia. However, four of five *C. fetus*–infected second-trimester patients were delivered of dead infants despite antibiotic therapy. One patient received antibiotic therapy and had a normal term infant. All the mothers survived their infection.[193]

Central nervous system (CNS) infections with *C. fetus* occur in neonates and adults. The prognosis is poor for premature infants, but five of six full-term neonates in one series survived infection. Infection is manifested as a meningoencephalitis with a cerebrospinal fluid polymorphonuclear pleocytosis. Subdural effusion may complicate infection. Meningoencephalitis is also the most common CNS manifestation of *C. fetus* infection in adults.[194] Cerebrovascular accidents, subarachnoid hemorrhages, and brain abscesses also occur. The prognosis is better in adults than in neonates, with a survival rate of approximately 67%, although neurologic sequelae are frequent. *C. fetus* has been shown to cause a variety of other types of localized infections, including septic arthritis, spontaneous bacterial peritonitis, salpingitis, lung abscess, empyema, cellulitis, urinary tract infection, vertebral osteomyelitis, and cholecystitis.[195,196] Although most patients with these illnesses recovered with appropriate antibiotics and drainage procedures, the clinical course was frequently prolonged and relapsing. Antibiotic resistance to fluoroquinolones may develop in immunocompromised patients who receive monotherapy regimens.[197] Nevertheless, in other patients, self-limiting bacteremia without any sequelae has been observed. Hypogammaglobulinemic patients may have persistent bacteremia and local symptoms unless given chronic suppressive therapy with antibiotics.

C. fetus is found in multiple animals and animal products, mainly cattle and sheep, which are probably the main source of human infection.[198]

Foodborne sources of infection likely include raw milk products, raw liver, and raw meat of such animals.[199] Similarly, reptiles may carry a different group of *C. fetus* strains,[200,201] and human infections with these organisms have been reported as well.[198,202] Reports of a cluster of *C. fetus* cases among men who have sex with men suggest the possibility of person-to-person transmission.[203]

Infection Caused by Other Enteric *Campylobacter* Species

The clinical manifestations of infection caused by other enteric *Campylobacter* spp. overlap substantially with those of *C. jejuni* infection.[149,204,205] On average, *C. coli* may produce milder disease.[62] In one series of homosexual men, *H. cinaedi* and *H. fennelliae* infections were more often asymptomatic than were those caused by *C. jejuni.*[206] Among immunocompromised patients, especially those with acquired immunodeficiency syndrome or hematologic malignancies, these "atypical" *Helicobacter* spp. have caused cellulitis, osteomyelitis, and bacteremia.[207–209] As with *C. fetus, C. upsaliensis* mostly causes diarrheal diseases in previously normal people[22,23,210] and bacteremia in immunocompromised hosts[211]; most strains of the latter species are serum resistant. Other extraintestinal manifestations, such as breast abscess, have been observed.[212] Although dogs have long been considered an important source of human *C. upsaliensis* infections, genetics studies have shown that human and canine strains are distinct.[213] Cellulitis may occur in immunocompromised hosts infected with any of a variety of these "atypical" species.[209] *C. hyointestinalis,* which resembles *C. fetus* in its biochemical characteristics,[214] also may cause bacteremia in immunocompromised hosts. *A. butzleri* may cause abdominal cramps without diarrheal illness.[215] *Helicobacter (Flexispira) rappini* has been reported to cause bacteremia in immunocompromised hosts.[216]

C. fetus subsp. *venerealis,* which had never been considered a human pathogen, was reported to have been isolated from stools from two homosexual men in Australia and from two women with bacterial vaginosis. *C. fetus* subsp. *fetus* has been isolated from two other patients with vaginosis. *Campylobacter curvus* has caused septicemia, liver abscess, and possibly chronic (Brainerd) diarrhea.[217,218] *Campylobacter sputorum* subsp. *sputorum,* which is indigenous to the human mouth and intestine, has been isolated from perianal boils and lung abscesses. *C. sputorum* subsp. *bubulus,* a commensal of sheep and cattle, has been isolated from boils and skin abscesses from humans. *Campylobacter insulaenigrae,* a recently identified species seen primarily in marine mammals, has been isolated from the stool and blood of a patient with end-stage renal and hepatic disease.[219] *Campylobacter concisus,* long believed to be part of the microbiota of healthy people, is now considered a possible cause of human gastrointestinal illness.[220–222] An increasing body of evidence also has linked *C. concisus* infection to childhood Crohn disease.[223–227] Other recently identified *Campylobacter* spp. that may be clinically relevant include *C. urealyticus,*[228–231] *C. troglodytis,*[232] *C. lari* subsp. *concheus, C. peloridis,*[233] *C. gracilis,*[234] and *C. fetus* subsp. *testudinum.*[235] Since these organisms are fastidious and can only be diagnosed by culture, it is likely that they are not as rare as it presently appears.

DIAGNOSIS

The gold standard for diagnosis of *Campylobacter* infection remains isolation by culture; however, multiple new non–culture-based techniques are now in use.

Bacteriologic Studies

Confirmation of the diagnosis of *C. jejuni* infection is based on a positive result on stool culture or, occasionally, blood culture, as described in the "Microbiology" section earlier. Because blood cultures are not often performed in the evaluation of patients presenting with diarrheal symptoms, the frequency of bacteremia is not known. *Campylobacter* spp. cannot be isolated from fecal specimens unless microaerobic incubation conditions and selective techniques that reduce the growth of competing microorganisms are used.[4,43] *C. fetus* is usually isolated from blood cultures 4 to 14 days after the specimen has been obtained.[142] Occasionally, *C. fetus* may be isolated from feces of patients with either diarrheal or systemic infections.[236] If *C. fetus* or another of the atypical species is suspected, incubation at 37°C and use of media without cephalosporins are necessary. Matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) mass spectrometry can also provide rapid identification of clinical *Campylobacter* species (e.g., VITEK 2 system; bioMerieux USA, Durham, NC).

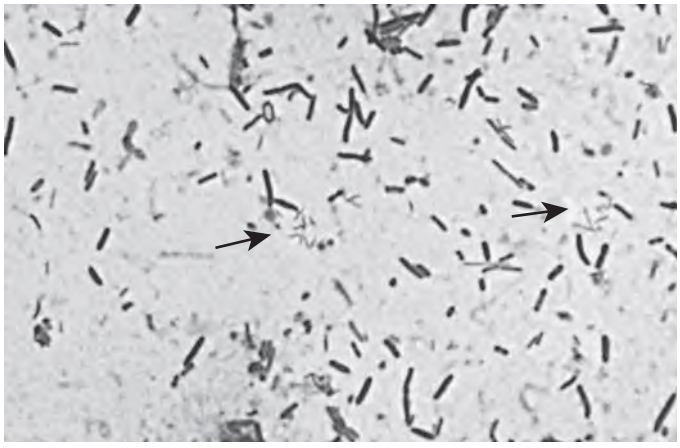

FIG. 216.3 Gram stain of fecal specimen from a patient with *Campylobacter* enteritis. Arrows point to typical gram-negative fine, small, spiral, and *Vibrio*-like organisms (×1024).

Molecular Techniques and Immunoassays

Use of enzyme immunoassays and nucleic acid amplification tests (NAATs, such as polymerase chain reaction) for *Campylobacter* diagnosis are also rapidly expanding in the clinical microbiology laboratory. Although no standardized guidelines exist for use, these techniques are significantly more rapid and sensitive than culture-based methods and often include at least *C. jejuni* and *C. coli.*[237] NAATs for *Campylobacter* diagnosis are usually part of a multiple enteric pathogen panel, including *Salmonella, Shigella*, and others.[237] However, when antibiotic resistance is a concern, the bacterial isolate is still needed and culture should be concurrently performed, especially in outbreak settings. In addition, NAATs detect bacterial DNA, not viable organisms, and should be interpreted with caution.

Direct Examination of Feces

Examination of diarrheal fecal specimens by darkfield or phase-contrast microscopy within 2 hours of passage can permit a rapid presumptive diagnosis of *Campylobacter* enteritis if the characteristic darting motility of the *Campylobacter* organism is seen.[145,238] This test is particularly useful in the acute phase of the illness. Similarly, the presence of vibrioid forms in Gram-stained stool specimens is a very specific diagnostic feature, although the sensitivity of this finding is 50% to 75% (Fig. 216.3).[239] Although infrequently performed, these techniques could be used by clinicians and laboratorians who want to make a rapid diagnosis in cases in which the index of suspicion for *Campylobacter* infections is high.

THERAPY

Fluid and electrolyte replacement constitutes the cornerstone of treatment of diarrheal illnesses. Patients with *Campylobacter* infections who are severely dehydrated should undergo rapid volume expansion using intravenous solutions of electrolytes in water. For patients with less serious volume depletion, oral rehydration using glucose and electrolyte solutions is indicated. People infected with *C. jejuni* who are ill enough to seek medical attention and from whom a fecal culture is obtained represent only a subset of all those infected. Nevertheless, even among these patients, fewer than half are candidates for specific antimicrobial therapy.[143] In a meta-analysis of 11 small randomized trials, antimicrobial agents reduced the duration of intestinal symptoms by approximately 1.3 days.[240] Because the greatest therapeutic benefit occurs when antimicrobial therapy is started early, the rapid presumptive diagnosis of *Campylobacter* infection by means of direct visualization of the organisms in stool is clinically relevant. Earlier diagnosis also has been

facilitated by the development of rapid CIDTs. Treatment with antibiotics seems prudent in those patients with high fever, bloody diarrhea, or more than eight stools per day; in patients whose symptoms have not lessened or are worsening at the time the diagnosis is made; or in those in whom symptoms have persisted for more than 1 week.[241]

In vitro, *C. jejuni* is susceptible to a wide variety of antimicrobial agents, including erythromycin, macrolides, the tetracyclines, the aminoglycosides, chloramphenicol, the quinolones, the nitrofurans, and clindamycin.[242,243] Erythromycin was once considered the agent of choice; however, because erythromycin, is primarily metabolized by cytochrome P-450 isoenzyme 3A4, there is a slight risk for sudden cardiac death. Therefore this agent is no longer recommended. We agree with the recommendation made in the 2017 Infectious Diseases Society of America guidelines that first-line therapy is either a macrolide or a fluoroquinolone.[244] Therapy with extended-spectrum macrolides such as clarithromycin or azithromycin should be equally effective.[245] The recommended dosage of azithromycin is 500 mg orally daily for 3 days; a single dose of 1000 mg may be equally effective, although this dosage is associated with more nausea. In children, the recommended dosage of azithromycin is 10 mg/kg per day for 3 days.

The dosage of ciprofloxacin is 500 mg orally twice daily for 5 to 7 days,[246] although as with azithromycin, a single dose (750 mg) may be just as effective.[247] Fluoroquinolones have the advantage of possessing activity across a broad spectrum of bacteria causing diarrheal illness, including *Campylobacter* spp. However, fluoroquinolones should be used with caution because rising rates of resistance to these agents have limited their usefulness in treating *Campylobacter* infections (see later section on "Resistance to Fluoroquinolones"). Another alternative agent is tetracycline, except in children younger than age 9 years; in such patients, clindamycin may be used. Most *C. jejuni* and *C. coli* isolates are not susceptible to most cephalosporins or penicillin, and these agents should not be used. However, amoxicillin or ticarcillin plus clavulanic acid appears to be universally effective.[248] Some macrolide- and fluoroquinolone-resistant *C. coli* strains may be susceptible to fosfomycin.[249] Susceptibility to sulfonamides and metronidazole is variable. Unlike in *Salmonella* infections, treatment with antimicrobial agents does not prolong carriage of *C. jejuni;* on the contrary, erythromycin eliminates carriage within 72 hours in most patients.[242]

H. cinaedi and those *Campylobacter* strains acquired in developing countries, especially *C. coli,* are more likely to be resistant to macrolides and tetracycline.[250] In such cases, when treatment is indicated, alternative agents should be used until susceptibility is known. Use of an antimotility agent appears to prolong duration of symptoms and has been associated with fatalities.[251] The necessity for treating septic or bacteremic episodes with agents other than macrolides has not been established. For those patients who appear very ill, treatment with gentamicin, imipenem, or chloramphenicol (where available) is indicated, but susceptibility tests should be performed. In hypogammaglobulinemic patients with recurrent *C. jejuni* bacteremias, fresh-frozen plasma with appropriate antibiotics may eradicate the infection[163]; oral immune globulin therapy may have some value as well for recurrent diarrheal illness.[252] Systemic *C. fetus* infections should be treated parenterally, but erythromycin should not be used.[253,254] Occasionally, systemic infections diagnosed only retrospectively by positive results on blood culture resolve after empirical oral therapy. In these cases, follow-up cultures are recommended; if results are no longer positive, further treatment is not required. If necessary, ampicillin treatment has been associated with good results. Patients with endovascular infections caused by *C. fetus* require at least 4 weeks of therapy, and gentamicin or ampicillin is probably the agent of choice.[254] Treatment with imipenem or meropenem constitutes another alternative. Infections of the CNS should be treated with ampicillin, imipenem, or chloramphenicol (where available) for 2 or 3 weeks. Patients with other serious infections should also receive parenteral gentamicin or another aminoglycoside, ampicillin, or imipenem for at least 2 weeks. Because antibodies to *C. fetus* are not usually present in serum from normal people, intravenous immune globulin is not helpful for this infection in immunodeficient patients.[188] For *C. fetus*–infected patients with diarrheal illness or other less severe infections, treatment need not be as intense or as prolonged.

TABLE 216.4 Resistance Mechanisms in *Campylobacter*

RESISTANCE MECHANISM	EXAMPLES
Modification of an antibiotic's target or alteration in the expression level of the target	Modification of the DNA gyrase target of quinolones/fluoroquinolones[258] Modification of the ribosomal A site target of tetracyclines via binding of bacterial TetO[259] Mutations in 23S ribosomal RNA target of macrolides[260,261] Variant forms of the dihydrofolate reductase target of trimethoprim[262]
Inability of the antibiotic to reach its target	Major outer membrane protein[263,264] Lipopolysaccharide and capsule-mediated exclusion[265]
Efflux of the antibiotic	Efflux of macrolides, fluoroquinolones, β-lactams, and tetracyclines via CmeABC[257]
Modification or inactivation of the antibiotic	Aminoglycoside-modifying enzymes[266] β-Lactamases[267–269]

Antibiotic Resistance

Campylobacter species are often naturally transformable organisms, allowing for the acquisition of resistance genes from other *Campylobacter* spp. as well as other gram-positive and gram-negative organisms.[255,256] These genetic elements represent a combination of endogenous and acquired genes that may be chromosomal-, transposon-, or plasmid-borne and include the resistance mechanisms shown in Table 216.4.[257–269]

High-level antibiotic resistance often occurs due to the combined effects of antibiotic efflux plus a second mechanism. The most well-studied efflux mechanism is a tripartite multidrug-resistance pump that also confers resistance to bile acids, known as CmeABC.[257,270] This pump is encoded by three genes: *cmeC,* corresponding to an outer membrane protein; *cmeA,* corresponding to the periplasmic portion, and *cmeB,* encoding the inner membrane drug transporter.[257,271] Additional potential efflux pumps that may contribute to antibiotic resistance (CmeDEF and CmeG) have also been reported, but their clinical significance is unclear.[272,273] Although there are numerous examples of resistance to specific antibiotics, the following discussion will be restricted to those antibiotics most commonly used to treat acute diarrhea: fluoroquinolones and macrolides.

Resistance to Fluoroquinolones

The fluoroquinolones (e.g., ciprofloxacin) are the most commonly used class of antibiotics used to treat acute bacterial diarrhea.[274] Because diarrhea due to *Campylobacter* is generally clinically indistinguishable from other causes of acute diarrhea, many cases are treated empirically with fluoroquinolones. Therefore fluoroquinolone resistance is of concern.

Through the late 1980s into the early 1990s, fluoroquinolone resistance rates were generally low in the United States and worldwide.[275,276] In 2013, the Centers for Disease Control and Prevention designated *Campylobacter* as a microorganism with a serious antibiotic resistance threat.[277] This was driven principally by the increase in fluoroquinolone resistance observed in *C. jejuni* from 15% in 1998 to 25% in 2015.[278] Resistance rates in certain parts of the world are much higher than in the United States—as high as 75% in travelers to Asia[279]—highlighting the need for an accurate travel history when assessing the traveler with diarrhea. Inappropriate and overprescription of fluoroquinolones in humans, combined with increased fluoroquinolone use in the poultry industry in particular, have contributed to the increased prevalence of fluoroquinolone resistance. In 2004, the US Food and Drug Administration reversed its prior approval for the therapeutic use of the veterinary fluoroquinolone enrofloxacin because of the concern that the observed increase in fluoroquinolone resistance in human isolates was due to an increase in fluoroquinolone-resistant poultry isolates.[276,280] Only approximately 2% to 3% of US isolates are resistant to azithromycin, and so macrolides are the first-line therapies.[244] Unfortunately, macrolide resistance rates are much higher in parts of Asia and Africa; for example,

in studies in South Africa and Nigeria, 80% or more of isolates were resistant to macrolides.[281,282]

The target of fluoroquinolones is bacterial DNA gyrase, which unwinds double-stranded DNA to allow for DNA replication and transcription. The two main subunits of DNA gyrase are encoded by *gyrA* and *gyrB*.[283] Fluoroquinolones form a stable complex with these enzymes on DNA, thereby inhibiting DNA replication and transcription, leading to bacterial cell death.[284] Resistance in *Campylobacter* occurs most commonly via a Thr-86-Ile substitution in the quinolone resistance-determining region (QRDR) of *gyrA*. Unlike fluoroquinolone resistance in *Salmonella* and *E. coli,* this single mutation confers high-level resistance to nalidixic acid and fluoroquinolones.[258] Because only the single Thr-86-Ile mutation is needed to mediate high-level resistance and does so without an apparent (or large) fitness cost, fluoroquinolone resistance appears quickly in animals and humans.[285–288] In contrast, less common mutations in *gyrA* (i.e., Asp-90-Asn and Ala-70-Thr) confer only intermediate-level resistance.[289] However, if the efflux pump CmeABC is present also, synergy between these two mechanisms produces high-level fluoroquinolone resistance.[290–292] Finally, although mutations in *gyrB* occur, none confers resistance to fluoroquinolones.[293]

Resistance to Macrolides

Macrolides, specifically azithromycin, are the treatments of choice for diarrheal illnesses due to *Campylobacter,* owing to the unacceptably high rates of fluoroquinolone resistance.[274] These antibiotics are large molecules (>700 Da) that inhibit protein synthesis by reversibly binding to the P site on the 50S subunit of bacterial ribosomes, leading to bactericidal or bacteriostatic effects.

The two principal mechanisms that mediate macrolide resistance in *Campylobacter* are (1) modification of the target site on bacterial ribosomes and (2) efflux via the multidrug-resistance efflux pump CmeABC. These mechanisms can work synergistically to produce high-level macrolide resistance in *Campylobacter.*[294,295] Specific point mutations at positions 2074 or 2075 (positions 2058 and 2059 in *E. coli* numbering) of the peptidyl encoding region in domain V of the 23S rRNA gene leads to high-level resistance when all three copies of the 23S rRNA genes are mutated.[261,296–298] There appears to be a gene dosage effect because strains harboring mutations on two 23S rRNA genes exhibit only intermediate resistance[299,300]; mutation in a single rRNA gene has not been reported. The low spontaneous mutation rate of the 23S rRNA genes (approximately 10^{-10} per cell per generation) contributes to the apparent higher barrier to the emergence of resistance compared with fluoroquinolones.[295] In another contrast to fluoroquinolone resistance, competition experiments suggest that macrolide resistance imparts a fitness cost.[301–305]

The second major mechanism of macrolide resistance is via the multidrug-resistance efflux pump CmeABC.[257,261] High-level resistance can occur in concert with ribosomal mutations as well as by mutation in *cmeB* alone.[306] For example, in macrolide-resistant strains that express wild-type 23S rRNA, gene disruption or silencing of *cmeB* or *cmeA* causes reversion to a macrolide-susceptible phenotype.[294,307] In other instances, the CmeABC efflux pump and rRNA mutations function synergistically to effect high-level macrolide resistance.[294,308,309]

PROGNOSIS

The vast majority of patients recover fully after *C. jejuni* infections, either spontaneously or after appropriate antimicrobial therapy. The "reactive arthritis," formerly called Reiter syndrome, occurring in human leukocyte antigen HLA-B27–positive people closely resembles that seen after *Yersinia, Salmonella,* or *Shigella* infections and is not specific to *C. jejuni* infection. However, rheumatologic symptoms may persist for several months or possibly for years in a few affected people. GBS is an uncommon sequela of *Campylobacter* enteritis, but because of their high prevalence, *Campylobacter* infections are the most important recognized antecedent of this clinically devastating disorder. Occasional deaths after *C. jejuni* infections have been reported in developed countries[251]; in most cases, the victim was an elderly person or an immunocompromised host. However, fatalities in previously healthy young adults may occur, probably as a result of volume depletion. Some of the deaths that occur in GBS patients can be attributed to the consequences of *C. jejuni* infection. Because in developing countries most symptomatic *Campylobacter* infections occur in children younger than 2 years of age[60] and frequently produce a dysenteric picture, it is reasonable to conclude that *C. jejuni* infection may play a role in the dehydration and malnutrition that often accompany infantile diarrhea in these geographic areas. The outcome of infections caused by newly discovered *Campylobacter*-like organisms remains to be determined.

C. fetus infection may be lethal to patients with chronic compensated diseases such as cirrhosis or diabetes mellitus or may hasten the demise of seriously immunocompromised patients. For immunocompromised hosts with systemic *C. fetus* infections, prognosis is most dependent on the rapidity with which appropriate antimicrobial therapy is begun. Previously healthy people infected with *C. fetus* usually survive the illness without permanent sequelae.

PREVENTION

Because most human *Campylobacter* infections are acquired from consumption or handling of poultry, thorough cooking of chicken and other poultry products is an important step in preventing transmission of infection to people. Furthermore, meticulous attention to avoiding cross-contamination of utensils and cutting boards during food preparation also may reduce the risk of infection. Elimination or reduction of use of antibiotics in animals used for food production may reduce the rate of antibiotic resistance among *Campylobacter* species. Finally, because *C. jejuni* infections are "accidentally" acquired by humans, and because there is evidence for the natural development of immunity among people in developing countries, the goal of producing a vaccine is probably achievable.

Key References

The complete reference list is available online at Expert Consult.

1. Parkhill J, Wren BW, Mungall K, et al. The genome sequence of the food-borne pathogen *Campylobacter jejuni* reveals hyper-variable sequences. *Nature.* 2000;403:665–668.
18. Crofts AA, Poly FM, Ewing CP, et al. *Campylobacter jejuni* transcriptional and genetic adaption during human infection. *Nat Microbiol.* 2018;3:494–502.
22. Labarca JA, Sturgen J, Borenstein L, et al. *Campylobacter upsaliensis*: another pathogen for consideration in the United States. *Clin Infect Dis.* 2002;34:e59–e60.
29. Friedman CR, Hoekstra RM, Samuel M, et al. Risk factors for sporadic *Campylobacter* infection in the United States: a case-control study in FoodNet sites. *Clin Infect Dis.* 2004;38:S285–S296.
34. Fullerton KE, Ingram LA, Jones TF, et al. Sporadic *Campylobacter* infection in infants: a population-based surveillance case-control study. *Pediatr Infect Dis J.* 2007;26:19–24.
36. Centers for Disease Control and Prevention. Multistate Outbreak of Multidrug-Resistant *Campylobacter* Infections Linked to Contact with Pet Store Puppies; 2018. https://www.cdc.gov/campylobacter/outbreaks/puppies-9-17/index.html.
44. Nachamkin I, Blaser MJ, Szymanski C, eds. *Campylobacter jejuni.* 3rd ed. Washington, DC: American Society for Microbiology; 2008.
48. Blaser MJ, Wells JF, Feldman RA, et al. *Campylobacter* enteritis in the United States: a multicenter study. *Ann Intern Med.* 1983;98:360.
49. Mead PS, Slutsker L, Dietz V, et al. Food-related illness and death in the United States. *Emerg Infect Dis.* 1999;5:607–625.
52. Marder MPHEP, Griffin PM, Cieslak PR, et al. Preliminary incidence and trends of infections with pathogens transmitted commonly through food — Foodborne Diseases Active Surveillance Network, 10 U.S. sites, 2006–2017. *MMWR Morb Mortal Wkly Rep.* 2018;67:324–328.
55. Marder EP, Cieslak PR, Cronquist AB, et al. Incidence and trends of infections with pathogens transmitted commonly through food and the effect of increasing use of culture-independent diagnostic tests on surveillance — Foodborne Diseases Active Surveillance Network, 10 U.S. sites, 2013–2016. *MMWR Morb Mortal Wkly Rep.* 2017;66:397–403.
56. Llarena AK, Taboada E, Rossi M. Whole-genome sequencing in epidemiology of *Campylobacter jejuni* infections. *J Clin Microbiol.* 2017;55:1269.
58. Tauxe RV. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. In: Nachamkin I, Blaser MJ, Tompkins LS, eds. *Campylobacter jejuni: Current Status and Future Trends.* Washington, DC: American Society for Microbiology; 1992:9–19.
61. Platts-Mills JA, Babji S, Bodhidatta L, et al. Pathogen-specific burdens of community diarrhoea in developing countries: a multisite birth cohort study. *Lancet Glob Health.* 2015;3:e564–e575.
64. Amour C, Gtatz J, Mduma E, et al. Epidemiology and impact of *Campylobacter* infection and children in 8 low-resource settings: results from the MAL-ED study. *Clin Infect Dis.* 2016;63:1171–1179.
66. Shah N, DuPont HL, Ramsey DJ. Global etiology of travelers' diarrhea: systematic review from 1973 to the present. *Am J Trop Med Hyg.* 2009;80:609–614.
69. Black RE, Levine MM, Clements ML, et al. Experimental *Campylobacter jejuni* infection in humans. *J Infect Dis.* 1988;157:472.

90. de Zoete MR, Keestra AM, Wagenaar JA, et al. Reconstitution of a functional Toll-like receptor 5 binding site in *Campylobacter jejuni* flagellin. *J Biol Chem*. 2010;285:12149–12158.
92. Mortensen NP, Schiellerup P, Boisen N, et al. The role of *Campylobacter jejuni* cytolethal distending toxin in gastroenteritis: toxin detection, antibody production, and clinical outcome. *APMIS*. 2011;119:626–634.
95. Hickey TE, Majam G, Guerry P. Intracellular survival of *Campylobacter jejuni* in human monocytic cells and induction of apoptotic death by cytolethal distending toxin. *Infect Immun*. 2005;73:5194–5197.
100. Tracz DM, Keelan M, Ahmed-Bentley J, et al. pVir and bloody diarrhea in *Campylobacter jejuni* enteritis. *Emerg Infect Dis*. 2005;11838–11843.
103. Pei Z, Blaser MJ. PEB1, the major cell-binding factor of *Campylobacter jejuni,* is a homolog of the binding component in gram-negative nutrient transport systems. *J Biol Chem*. 1993;267:18717–18725.
110. Perez Perez GI, Blaser MJ. Lipopolysaccharide characteristics of pathogenic campylobacters. *Infect Immun*. 1985;47:353–359.
112. Aspinall GO, Fujimoto S, McDonald AG, et al. Lipopolysaccharides from *Campylobacter jejuni* associated with Guillain-Barré syndrome patients mimic human gangliosides in structure. *Infect Immun*. 1994;62:2122–2125.
114. Goodyear CS, O'Hanlon GM, Plomp JJ, et al. Monoclonal antibodies raised against Guillain-Barre syndrome-associated *Campylobacter jejuni* lipopolysaccharides react with neuronal gangliosides and paralyze muscle-nerve preparations. *J Clin Invest*. 1999;104:697–708.
115. Heikema AP, Islam Z, Horst-Kreft D, et al. *Campylobacter jejuni* capsular genotypes are related to Guillain-Barre syndrome. *Clin Microbiol Infect*. 2015;21:852, e1–9.
118. Blaser MJ, Smith PF, Kohler PA. Susceptibility of *Campylobacter* isolates to the bactericidal activity in human serum. *J Infect Dis*. 1985;151:227.
119. Wagenaar JA, van Bergen MA, Blaser MJ, et al. *Campylobacter fetus* infections in humans: exposure and disease. *Clin Infect Dis*. 2014;58:1579–1586.
120. Escher R, Brunner C, von Steiger N, et al. Clinical and epidemiological analysis of *Campylobacter fetus* subsp. *fetus* infections in humans and comparative genetic analysis with strains isolated from cattle. *BMC Infect Dis*. 2016;16:198.
123. Blaser MJ, Smith PF, Repine JE, et al. Pathogenesis of *Campylobacter fetus* infections: failure of C3b to bind explains serum and phagocytosis resistance. *J Clin Invest*. 1988;81:1434–1444.
127. Kirkpatrick BD, Lyon CE, Porter CK, et al. Lack of homologous protection against *Campylobacter jejuni* CG8421 in a human challenge model. *Clin Infect Dis*. 2013;57:1106–1113.
135. Havelaar AH, van Pelt W, Ang CW, et al. Immunity to *Campylobacter*: its role in risk assessment and epidemiology. *Crit Rev Microbiol*. 2009;35:1–22.
156. Nielsen H, Hansen KK, Gradel KO, et al. Bacteraemia as a result of *Campylobacter* species: a population-based study of epidemiology and clinical risk factors. *Clin Microbiol Infect*. 2010;16:57–61.
158. Bhattacharyya R, Flores EJ, Azar MM. Case 30-2017 — A 65-year-old woman with altered mental status, bacteremia, and acute liver failure. *N Engl J Med*. 2017;377:1274–1282.
164. Bloomfield SJ, et al. Long-term colonization by *Campylobacter jejuni* within a human host: evolution, antimicrobial resistance, and adaptation. *J Infect Dis*. 2018;217:103–111.
167. Rees JH, Soudain SE, Gregory NA, et al. *Campylobacter jejuni* infection and Guillain-Barré syndrome. *N Engl J Med*. 1995;333:1374–1379.
170. Kuroki S, Saida T, Nukina M, et al. *Campylobacter jejuni* strains from patients with Guillain-Barré syndrome belong mostly to Penner serogroup 19 and contain β-*N*-acetylglucosamine residues. *Ann Neurol*. 1993;33:243–247.
173. Loshaj-Shala A, Regazzoni L, Daci A, et al. Guillain Barre syndrome (GBS): new insights in the molecular mimicry between *C. jejuni* and human peripheral nerve (HPN) proteins. *J Neuroimmunol*. 2015;289:168–176.
174. Aspinall GO, Fujimoto S, McDonald AG, et al. Lipopolysaccharides from *Campylobacter jejuni* associated with Guillain-Barré syndrome patients mimic human gangliosides in structure. *Infect Immun*. 1994;62:2122–2125.
176. Goodyear CS, O'Hanlon GM, Plomp JJ, et al. Monoclonal antibodies raised against Guillain-Barré syndrome-associated *Campylobacter jejuni* lipopolysaccharides react with neuronal gangliosides and paralyze muscle-nerve preparations. *J Clin Invest*. 1999;104:697–708.
178. Pope JE, Krizova A, Garg AX, et al. *Campylobacter* reactive arthritis: a systematic review. *Semin Arthritis Rheum*. 2007;37:48–55.
180. Lecuit M, Abachin E, Martin A, et al. Immunoproliferative small intestinal disease associated with *Campylobacter jejuni*. *N Engl J Med*. 2004;350:239–248.
186. Pacanowski J, Lalande V, Lacombe K, et al. *Campylobacter* bacteremia: clinical features and factors associated with fatal outcome. *Clin Infect Dis*. 2008;47:790–796.
193. Fujihara N, Takakura S, Saito T, et al. A case of perinatal sepsis by *Campylobacter fetus* subsp. *fetus* infection successfully treated with carbapenem – case report and literature review. *J Infect*. 2006;53:e199–e202.
195. Cone LA, Dreisbach PB, Hirschberg J, et al. Cellulitis and septic arthritis caused by *Campylobacter fetus* and *Campylobacter jejuni*: report of 2 cases and review of the literature. *J Clin Rheumatol*. 2003;9:362–369.
203. Marchand-Senécal X, Bekal S, Pilon PA, et al. *Campylobacter fetus* cluster among men who have sex with men, Montreal, Quebec, Canada, 2014-2016. *Clin Infect Dis*. 2017;65:1751.
223. Ismail Y, Mahendran V, Octavia S, et al. Investigation of the enteric pathogenic potential of oral *Campylobacter concisus* strains isolated from patients with inflammatory bowel disease. *PLoS ONE*. 2012;7:e38217.
231. O'Donovan D, Corcoran GD, Lucey B, et al. *Campylobacter ureolyticus:* a portrait of the pathogen. *Virulence*. 2014;5:498–506.
240. Ternhag A, Asikainen T, Giesecke J, et al. A meta-analysis on the effects of antibiotic treatment on duration of symptoms caused by infection with *Campylobacter* species. *Clin Infect Dis*. 2007;44:696.
244. Shane AL, Mody RK, Crump JA, et al. 2017 Infectious Diseases Society of America clinical practice guidelines for the diagnosis and management of infectious diarrhea. *Clin Infect Dis*. 2017;65:e45.
245. Centers for Disease Control and Prevention. The pre-travel consultation: travelers' diarrhea. The Yellow Book: CDC health information for international travel, 2018. https://wwwnc.cdc.gov/travel/yellowbook/2018/the-pre-travel-consultation/travelers-diarrhea. Accessed September 5, 2017.
265. Post A, Martiny D, van Waterschoot N, et al. Antibiotic susceptibility profiles among *Campylobacter* isolates obtained from international travelers between 2007 and 2014. *Eur J Clin Microbiol Infect Dis*. 2017;36:2101–2107.

References

1. Parkhill J, Wren BW, Mungall K, et al. The genome sequence of the food-borne pathogen *Campylobacter jejuni* reveals hyper-variable sequences. *Nature*. 2000;403:665–668.
2. Poly F, Guerry P. Pathogenesis of *Campylobacter*. *Curr Opin Gastroenterol*. 2008;24:27–31.
3. Smibert RM. Genus *Campylobacter*. In: Krieg NR, Holt HG, eds. *Bergey's Manual of Systematic Bacteriology*. Baltimore: Williams & Wilkins; 1984:111–118.
4. Nachamkin I. *Campylobacter, Helicobacter*, and related spiral bacteria. In: Murray PR, ed. *Manual of Clinical Microbiology*. 6th ed. Washington, DC: American Society for Microbiology; 1996:402–409.
5. Vandamme P, Vancanneyt M, Pot B, et al. Polyphasic taxonomic study of the emended genus *Arcobacter* with *Arcobacter butzleri* comb. nov. and *Arcobacter skirrowi* sp. nov., an aerotolerant bacterium isolated from veterinary specimens. *Int J Syst Bacteriol*. 1992;42:344–356.
6. Fennell CL, Totten PA, Quinn TC, et al. Characterization of *Campylobacter*-like organisms isolated from homosexual men. *J Infect Dis*. 1984;149:58–66.
7. Burnens AP, Stanley J, Schaad UB, et al. Novel *Campylobacter*-like organism resembling *Helicobacter fennelliae* isolated from a boy with gastroenteritis and from dogs. *J Clin Microbiol*. 1993;31:1916–1917.
8. Foley JE, Solnick JV, LaPointe J-M, et al. Identification of a novel enteric *Helicobacter* species in a kitten with severe diarrhea. *J Clin Microbiol*. 1998;36:908–912.
9. Husman M, Gries C, Jehnichen P, et al. *Helicobacter* sp. strain *Mainz* isolated from an AIDS patient with septic arthritis: case report and nonradioactive analysis of 16S rRNA sequence. *J Clin Microbiol*. 1994;32:3037–3039.
10. Bolton GJ, Hutchinson DN, Coates D. Blood-free selective medium for isolation of *Campylobacter jejuni* from feces. *J Clin Microbiol*. 1984;19:169–171.
11. Steele TW, McDermott JN. Technical note: the use of membrane filters applied directly to the surface of agar plates for the isolation of *Campylobacter jejuni* from feces. *Pathology*. 1984;16:263–265.
12. Lastovica AJ, LeRoux E. Efficient isolation of *Campylobacter upsaliensis* from stools. *J Clin Microbiol*. 2001;39:4222–4223.
13. Burnens AP, Nicolet J. Three supplementary diagnostic tests for *Campylobacter* species and related organisms. *J Clin Microbiol*. 1993;31:708–710.
14. Wang WL, Blaser MJ. Detection of pathogenic *Campylobacter* species in blood culture systems. *J Clin Microbiol*. 1986;23:709.
15. van Camp G, Fierens H, Vandamme P, et al. Identification of enteropathogenic *Campylobacter* species by oligonucleotide probes and polymerase chain reaction based on 16S rRNA genes. *Syst Appl Microbiol*. 1993;16:30–36.
16. Ng L-K, Kingombe CIB, Yan W, et al. Specific detection and confirmation of *Campylobacter jejuni* by DNA hybridization and PCR. *Appl Environ Microbiol*. 1997;63:4558–4563.
17. Persson S, Petersen HM, Jespersgaard C, et al. Real-time TaqMan polymerase chain reaction-based genus-identification and pyrosequencing-based species identification of *Campylobacter jejuni, C. coli, C. lari, C. upsaliensis*, and *C. fetus* directly on stool samples. *Diagn Microbiol Infect Dis*. 2012;74:6–10.
18. Crofts AA, Poly FM, Ewing CP, et al. *Campylobacter jejuni* transcriptional and genetic adaption during human infection. *Nat Microbiol*. 2018;3:494–502.
19. Giesendorf BAJ, van Belkum A, Koeken A, et al. Development of species-specific DNA probes for *Campylobacter jejuni, Campylobacter coli*, and *Campylobacter lari* by polymerase chain reaction fingerprinting. *J Clin Microbiol*. 1993;31:1541–1546.
20. Ugarte-Ruiz M, Gómez-Barrero S, Porrero MC, et al. Evaluation of four protocols for the detection and isolation of thermophilic *Campylobacter* from different matrices. *J Appl Microbiol*. 2012;113:200–208.
21. Blaser MJ, Taylor DN, Feldman RA. Epidemiology of *Campylobacter jejuni* infections. *Epidemiol Rev*. 1983;5:157.
22. Labarca JA, Sturgen J, Borenstein L, et al. *Campylobacter upsaliensis*: another pathogen for consideration in the United States. *Clin Infect Dis*. 2002;34:e59–e60.
23. Patton CM, Shaffer N, Edmonds P, et al. Human disease associated with "*Campylobacter upsaliensis*" (catalase-negative or weakly positive *Campylobacter* species) in the United States. *J Clin Microbiol*. 1989;27:66–73.
24. Atabay HI, Corry JEL. The prevalence of campylobacters and arcobacters in broiler chickens. *J Appl Microbiol*. 1997;83:619–626.
25. Burakoff A, Brown K, Knutsen J, et al. Outbreak of fluoroquinolone-resistant *Campylobacter jejuni* infections associated with raw milk consumption from a herdshare dairy–Colorado, 2016. *MMWR Morb Mortal Wkly Rep*. 2018;67:146–148.
26. Taylor DN, McDermott KT, Little JR, et al. *Campylobacter* enteritis associated with drinking untreated water in back-country areas of the Rocky Mountains. *Ann Intern Med*. 1983;99:38.
27. Mentzing L-O. Waterborne outbreaks of *Campylobacter* enteritis in central Sweden. *Lancet*. 1981;2:352.
28. Richardson G, Thomas DR, Smith RM, et al. A community outbreak of *Campylobacter jejuni* infection from a chlorinated public water supply. *Epidemiol Infect*. 2007;135:1151–1158.
29. Friedman CR, Hoekstra RM, Samuel M, et al. Risk factors for sporadic *Campylobacter* infection in the United States: a case-control study in FoodNet sites. *Clin Infect Dis*. 2004;38:S285–S296.
30. Gallay A, Bosquet V, Siret V, et al. Risk factors for acquiring sporadic *Campylobacter* infection in France: results from a national case-control study. *J Infect Dis*. 2008;197:1477–1484.
31. Stafford RJ, Schluter PJ, Wilson AJ, et al. OzFoodNet Working Group. Population-attributable risk estimates for risk factors associated with *Campylobacter* infection, Australia. *Emerg Infect Dis*. 2008;14:895–901.
32. Deming MS, Tauxe RV, Blake PA, et al. *Campylobacter* enteritis at a university: transmission from eating chicken and from cats. *Am J Epidemiol*. 1987;126:526–534.
33. Little CL, Gormley FJ, Rawal N, et al. A recipe for disaster: outbreaks of campylobacteriosis associated with poultry liver pâté in England and Wales. *Epidemiol Infect*. 2010;138:1691–1694.
34. Fullerton KE, Ingram LA, Jones TF, et al. Sporadic *Campylobacter* infection in infants: a population-based surveillance case-control study. *Pediatr Infect Dis J*. 2007;26:19–24.
35. Skirrow MB. *Campylobacter* enteritis in dogs and cats: a "new" zoonosis. *Vet Res Commun*. 1981;5:13.
36. Centers for Disease Control and Prevention. Multistate Outbreak of Multidrug-Resistant *Campylobacter* Infections Linked to Contact with Pet Store Puppies; 2018. https://www.cdc.gov/campylobacter/outbreaks/puppies-9-17/index.html.
37. Tu Z-C, Dewhirst FE, Blaser MJ. Evidence that the *Campylobacter fetus sap* locus is an ancient genomic constituent with origins before mammals and reptiles diverged. *Infect Immun*. 2001;69:2237–2244.
38. Tu Z, Zeitlin G, Gagner J-P, et al. *Campylobacter fetus* of reptile origin as a human pathogen. *J Clin Microbiol*. 2004;42:4405–4407.
39. Vesikari T, Huttunen L, Maki R. Perinatal *Campylobacter fetus* ss. *jejuni* enteritis. *Acta Paediatr Scand*. 1981;70:261.
40. Pepersack F, Prigogyne T, Butzler JP, et al. *Campylobacter jejuni* posttransfusional septicemia. *Lancet*. 1979;2:911.
41. Totten PA, Fennell CL, Tenover FC, et al. *Campylobacter cinaedi* (sp. nov.) and *Campylobacter fennelliae* (sp. nov.): two new *Campylobacter* species associated with enteric disease in homosexual men. *J Infect Dis*. 1985;151:131.
42. Sorvillo FJ, Lieb LE, Waterman SH. Incidence of campylobacteriosis among patients with AIDS in Los Angeles County. *J Acquir Immune Defic Syndr*. 1991;4:598–602.
43. Penner JL. The genus *Campylobacter*: a decade of progress. *Clin Microbiol Rev*. 1988;1:157–172.
44. Nachamkin I, Blaser MJ, Szymanski C, eds. *Campylobacter jejuni*. 3rd ed. Washington, DC: American Society for Microbiology; 2008.
45. Champion OL, Gaunt MW, Gundogdu O, et al. Comparative phylogenomics of the food-borne pathogen *Campylobacter jejuni* reveals genetic markers predictive of infection source. *Proc Natl Acad Sci USA*. 2005;102:16043–16048.
46. Louis VR, Gillespie IA, O'Brien SJ, et al. Temperature-driven *Campylobacter* seasonality in England and Wales. *Appl Environ Microbiol*. 2005;71:85–92.
47. Nichols GL. Fly transmission of *Campylobacter*. *Emerg Infect Dis*. 2005;11:361–364.
48. Blaser MJ, Wells JF, Feldman RA, et al. *Campylobacter* enteritis in the United States: a multicenter study. *Ann Intern Med*. 1983;98:360.
49. Mead PS, Slutsker L, Dietz V, et al. Food-related illness and death in the United States. *Emerg Infect Dis*. 1999;5:607–625.
50. Centers for Disease Control and Prevention. Incidence and trends of infection with pathogens transmitted commonly through food—Foodborne Diseases Active Surveillance Network, 10 U.S. sites, 1996-2012. *MMWR Morb Mortal Wkly Rep*. 2013;2:283–287.
51. Crim SM, Griffin PM, Tauxe R, et al. Preliminary incidence and trends of infection with pathogens transmitted commonly through food—foodborne diseases active surveillance network, 10 U.S. sites, 2016-2014. *MMWR Morb Mortal Wkly Rep*. 2015;64:495–499.
52. Marder MPHEP, Griffin PM, Cieslak PR, et al. Preliminary incidence and trends of infections with pathogens transmitted commonly through food — Foodborne Diseases Active Surveillance Network, 10 U.S. sites, 2006–2017. *MMWR Morb Mortal Wkly Rep*. 2018;67:324–328.
53. Weinberger M, Moran-Gilad J, Rokney A, et al. Molecular epidemiology of *Campylobacter jejuni* infection in Israel—a nationwide study. *Clin Microbiol Infect*. 2016;22:1005, e9–1005.e15.
54. European Centre for Disease Prevention and Control. Annual epidemiologic report: Campybacteriosis, 2016. http://ecdc.europa.eu/en/healthtopics/campylobacteriosis/surveillance-data/Pages/Annual-surveillance-data.aspx. Accessed April 25, 2017.
55. Marder EP, Cieslak PR, Cronquist AB, et al. Incidence and trends of infections with pathogens transmitted commonly through food and the effect of increasing use of culture-independent diagnostic tests on surveillance — Foodborne Diseases Active Surveillance Network, 10 U.S. sites, 2013–2016. *MMWR Morb Mortal Wkly Rep*. 2017;66:397–403.
56. Llarena AK, Taboada E, Rossi M. Whole-genome sequencing in epidemiology of *Campylobacter jejuni* infections. *J Clin Microbiol*. 2017;55:1269.
57. Ailes E, Scallan E, Berkelman RL, et al. Do differences in risk factors, medical care seeking, or medical practices explain the geographic variation in campylobacteriosis in Foodborne Diseases Active Surveillance Network (FoodNet) sites? *Clin Infect Dis*. 2012;54:S464–S471.
58. Tauxe RV. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. In: Nachamkin I, Blaser MJ, Tompkins LS, eds. *Campylobacter jejuni: Current Status and Future Trends*. Washington, DC: American Society for Microbiology; 1992:9–19.
59. Glass RI, Stoll BJ, Huq MI, et al. Epidemiologic and clinical features of endemic *Campylobacter jejuni* infection in Bangladesh. *J Infect Dis*. 1983;148:292.
60. Calva JJ, Ruiz-Pallacios GM, Lopez-Vidal AB, et al. Cohort study of intestinal infection with *Campylobacter* in Mexican children. *Lancet*. 1988;1:503–506.
61. Platts-Mills JA, Babji S, Bodhidatta L, et al. Pathogen-specific burdens of community diarrhoea in developing countries: a multisite birth cohort study. *Lancet Glob Health*. 2015;9:e564–e575.
62. Taylor DN, Echeverria P, Pitarangsi C, et al. The influence of immunity and strain characteristics on the epidemiology of campylobacteriosis. *J Clin Microbiol*. 1988;26:863.
63. Taylor DN, Perlman D, Echeverria PD, et al. *Campylobacter* immunity and quantitative excretion rates in Thai children. *J Infect Dis*. 1993;168:754–758.
64. Amour C, Gtatz J, Mduma E, et al. Epidemiology and impact of *Campylobacter* infection and children in 8 low-resource settings: results from the MAL-ED study. *Clin Infect Dis*. 2016;63:1171–1179.
65. Ethelberg S, Simonsen J, Gerner-Smidt P, et al. Spatial distribution and registry-based case-control analysis of *Campylobacter* infections in Denmark, 1991-2001. *Am J Epidemiol*. 2005;162:1008–1015.
66. Shah N, DuPont HL, Ramsey DJ. Global etiology of travelers' diarrhea: systematic review from 1973 to the present. *Am J Trop Med Hyg*. 2009;80:609–614.
67. Hope KG, Merritt TD, Durrheim DN. Short incubation periods in *Campylobacter* outbreaks associated with poultry liver dishes. *Commun Dis Intell Q Rep*. 2014;38:E20–E23.
68. Parry A, Fearnley E, Denehy E. 'Surprise': outbreak of *Campylobacter* infection associated with chicken liver pate at a surprise birthday party, Adelaide, Australia, 2012. *Western Pac Surveill Response J*. 2012;3:16–19.
69. Black RE, Levine MM, Clements ML, et al. Experimental *Campylobacter jejuni* infection in humans. *J Infect Dis*. 1988;157:472.
70. Blaser MJ, Hardesty HL, Powers B, et al. Survival of *Campylobacter fetus* subsp. *jejuni* in biological milieus. *J Clin Microbiol*. 1980;11:309.
71. Doorduyn Y, Van Pelt W, Siezen CL, et al. Novel insight in the association between salmonellosis or campylobacteriosis and chronic illness, and the role of host genetics in susceptibility to these diseases. *Epidemiol Infect*. 2008;136:1225–1234.
72. Neal KR, Scott HM, Slack RC, et al. Omeprazole as a risk factor for *Campylobacter* gastroenteritis: case-control study. *BMJ*. 1996;312:414–415.
73. Sopwith W, Birtles A, Matthews M, et al. Identification of potential environmentally adapted *Campylobacter jejuni* strain, United Kingdom. *Emerg Infect Dis*. 2008;14:1769–1773.
74. King EO. The laboratory recognition of *Vibrio fetus* and a closely related vibrio isolated from cases of human vibriosis. *Ann N Y Acad Sci*. 1962;90:700.

75. Lambert ME, Schofield PF, Ironside AG, et al. *Campylobacter* colitis. *Br Med J*. 1979;1:857.
76. Van Spreeuwel JP, Duursma GC, Meijer CJLM, et al. *Campylobacter* colitis: histologic, immunohistochemical and ultrastructural findings. *Gut*. 1985;26:945–951.
77. Blaser MJ, Reller LB, Luechtefeld NW, et al. *Campylobacter* enteritis in Denver. *West J Med*. 1982;136:287–290.
78. Chen ML, Ge Z, Fox JG, et al. Disruption of tight junctions and induction of proinflammatory cytokine responses in colonic epithelial cells by *Campylobacter jejuni*. *Infect Immun*. 2006;74:6581–6589.
79. Backert S, Boehm M, Wessler S, et al. Transmigration route of *Campylobacter jejuni* across polarized intestinal epithelial cells: paracellular, transcellular or both? *Cell Commun Signal*. 2013;11:72.
80. Kopecko DJ, Hu L, Zaal KJM. *Campylobacter jejuni*–microtubule-dependent invasion. *Trends Microbiol*. 2001;9:389–396.
81. Oelschlaeger TA, Guerry P, Kopecko DJ. Unusual microtubule-dependent endocytosis mechanisms triggered by *Campylobacter jejuni* and *Citrobacter freundii*. *Proc Natl Acad Sci USA*. 1993;90:6884–6888.
82. Watson RO, Galan JE. *Campylobacter jejuni* survives within epithelial cells by avoiding delivery to lysosomes. *PLoS Pathog*. 2008;4:e14.
83. Grant CCR, Konkel ME, Cieplak W, et al. Role of flagella in adherence, internalization, and translocation of *Campylobacter jejuni* in nonpolarized and polarized epithelial cell cultures. *Infect Immun*. 1993;61:1764–1771.
84. Yao R, Burr DH, Doig P, et al. Isolation of motile and non-motile insertional mutants of *Campylobacter jejuni*: the role of motility in adherence and invasion of eukaryotic cells. *Mol Microbiol*. 1994;14:883–893.
85. Konkel ME, Kim BJ, Rivera-Amill V, et al. Bacterial secreted proteins are required for the internaliztion of *Campylobacter jejuni* into cultured mammalian cells. *Mol Microbiol*. 1999;32:691–701.
86. Konkel ME, Klena JD, Rivera-Amill V, et al. Secretion of virulence proteins from *Campylobacter jejuni* is dependent on a functional flagellar export apparatus. *J Bacteriol*. 2004;186:3296–3303.
87. Rivera-Amill V, Konkel ME. Secretion of *Campylobacter jejuni* Cia proteins is contact dependent. In: Paul PS, Francis DH, eds. *Mechanisms in the Pathogenesis of Enteric Diseases 2*. New York: Plenum; 1999:225–229.
88. Poly F, Ewing C, Goon S, et al. Heterogeneity of a *Campylobacter jejuni* protein that is secreted through the flagella filament. *Infect Immun*. 2007;75:3859–3867.
89. Faber E, Gripp E, Maurischat S, et al. Novel immunomodulatory flagellin-like protein FlaC in *Campylobacter jejuni* and other campylobacterales. *mSphere*. 2016;1. e00028–15.
90. de Zoete MR, Keestra AM, Wagenaar JA, et al. Reconstitution of a functional Toll-like receptor 5 binding site in *Campylobacter jejuni* flagellin. *J Biol Chem*. 2010;285:12149–12158.
91. Johnson WM, Lior H. A new heat-labile cytolethal distending toxin (CLDT) produced by *Escherichia coli* isolates from clinical material. *Microb Pathog*. 1988;4:103–113.
92. Mortensen NP, Schiellerup P, Boisen N, et al. The role of *Campylobacter jejuni* cytolethal distending toxin in gastroenteritis: toxin detection, antibody production, and clinical outcome. *APMIS*. 2011;119:626–634.
93. Whitehouse CA, Balbo PB, Pesci EC, et al. *Campylobacter jejuni* cytolethal distending toxin causes a G_2-phase cell cycle block. *Infect Immun*. 1998;66:1934–1940.
94. Lara-Tejero M, Galan JE. A bacterial toxin that controls cell cycle progression as a deoxyribonuclease I-like protein. *Science*. 2000;290:354–357.
95. Hickey TE, Majam G, Guerry P. Intracellular survival of *Campylobacter jejuni* in human monocytic cells and induction of apoptotic death by cytolethal distending toxin. *Infect Immun*. 2005;73:5194–5197.
96. Shenker BJ, Besack D, McKay T, et al. Induction of cell cycle arrest in lymphocytes by *Actinobacillus actinomycetemcomitans* cytolethal distending toxin requires three subunits for maximum activity. *J Immunol*. 2005;174:2228–2234.
97. Bacon DJ, Alm RA, Burr DH, et al. Involvement of a plasmid in virulence of *Campylobacter jejuni* 81-176. *Infect Immun*. 2000;68:4384–4390.
98. Bacon DJ, Alm RA, Hu L, et al. DNA sequence and mutational analyses of the pVir plasmid of *Campylobacter jejuni* 81-176. *Infect Immun*. 2002;70:6242–6250.
99. Louwen RP, van Belkum A, Wagenaar JA, et al. Lack of association between the presence of the pVir plasmid and bloody diarrhea in *Campylobacter jejuni* enteritis. *J Clin Microbiol*. 2006;44:1867–1868.
100. Tracz DM, Keelan M, Ahmed-Bentley J, et al. pVir and bloody diarrhea in *Campylobacter jejuni* enteritis. *Emerg Infect Dis*. 2005;11838-11843.
101. Kervella M, Pages J-M, Pei Z, et al. Isolation and characterization of two *Campylobacter* glycine-extracted proteins that bind to HeLa cell membranes. *Infect Immun*. 1993;61:3440–3448.
102. Pei Z, Ellison RT III, Blaser MJ. Identification, purification and characterization of major antigenic proteins of *Campylobacter jejuni*. *J Biol Chem*. 1991;266:16363–16369.
103. Pei Z, Blaser MJ. PEB1, the major cell-binding factor of *Campylobacter jejuni,* is a homolog of the binding component in gram-negative nutrient transport systems. *J Biol Chem*. 1993;267:18717–18725.
104. Pei Z, Burucoa C, Grignon B, et al. Mutation in the *peb1A* locus of *Campylobacter jejuni* reduces interactions with epithelial cells and intestinal colonization of mice. *Infect Immun*. 1998;66:938–943.
105. Jin S, Song YC, Emili A, et al. JlpA of *Campylobacter jejuni* interacts with surface-exposed heat shock protein 90 alpha and triggers signalling pathways leading to the activation of NF-kappaB and p38 MAP kinase in epithelial cells. *Cell Microbiol*. 2003;5:165–174.
106. Konkel ME, Garvis SG, Tipton SL, et al. Identification and molecular cloning of a gene encoding a fibronectin-binding protein (CadF) from *Campylobacter jejuni*. *Mol Microbiol*. 1997;24:953–963.
107. Naikare H, Palyada K, Panciera R, et al. Major role for FeoB in *Campylobacter jejuni* ferrous iron acquisition, gut colonization, and intracellular survival. *Infect Immun*. 2006;74:5433–5444.
108. Moran AP. Structure and conserved characteristics of *Campylobacter jejuni* lipopolysaccharides. *J Infect Dis*. 1997;176:S115–S121.
109. Penner JL, Aspinall GO. Diversity of lipopolysaccharide structures in *Campylobacter jejuni*. *J Infect Dis*. 1997;176:S135–S138.
110. Perez Perez GI, Blaser MJ. Lipopolysaccharide characteristics of pathogenic campylobacters. *Infect Immun*. 1985;47:353–359.
111. Ang CW, De Klerk MA, Endtz HP, et al. Guillain-Barre syndrome- and Miller Fisher syndrome-associated *Campylobacter jejuni* lipopolysaccharides induce anti-GM1 and anti-GQ1b Antibodies in rabbits. *Infect Immun*. 2001;69:2462–2469.
112. Aspinall GO, Fujimoto S, McDonald AG, et al. Lipopolysaccharides from *Campylobacter jejuni* associated with Guillain-Barré syndrome patients mimic human gangliosides in structure. *Infect Immun*. 1994;62:2122–2125.
113. Heikema AP, Jacobs BC, Horst-Kreft D, et al. Siglec-7 specifically recognizes *Campylobacter jejuni* strains associated with oculomotor weakness in Guillain-Barré syndrome and Miller Fisher syndrome. *Clin Microbiol Infect*. 2013;19:E106–E112.
114. Goodyear CS, O'Hanlon GM, Plomp JJ, et al. Monoclonal antibodies raised against Guillain-Barre syndrome-associated *Campylobacter jejuni* lipopolysaccharides react with neuronal gangliosides and paralyze muscle-nerve preparations. *J Clin Invest*. 1999;104:697–708.
115. Heikema AP, Islam Z, Horst-Kreft D, et al. *Campylobacter jejuni* capsular genotypes are related to Guillain-Barre syndrome. *Clin Microbiol Infect*. 2015;21:e1–e9.
116. Keo T, Collins J, Kunwar P, et al. *Campylobacter* capsule and lipooligosaccharide confer resistance to serum and cationic antimicrobials. *Virulence*. 2011;2:30–40.
117. Mortensen NP, Kuijf ML, Ang CW, et al. Sialylation of *Campylobacter jejuni* lipo-oligosaccharides is associated with severe gastro-enteritis and reactive arthritis. *Microbes Infect*. 2009;11:988–994.
118. Blaser MJ, Smith PF, Kohler PA. Susceptibility of *Campylobacter* isolates to the bactericidal activity in human serum. *J Infect Dis*. 1985;151:227.
119. Wagenaar JA, van Bergen MA, Blaser MJ, et al. *Campylobacter fetus* infections in humans: exposure and disease. *Clin Infect Dis*. 2014;58:1579–1586.
120. Escher R, Brunner C, von Steiger N, et al. Clinical and epidemiological analysis of *Campylobacter fetus* subsp. *fetus* infections in humans and comparative genetic analysis with strains isolated from cattle. *BMC Infect Dis*. 2016;16:198.
121. Blaser MJ, Smith PF, Hopkins JA, et al. Pathogenesis of *Campylobacter fetus* infections: serum resistance associated with high molecular weight surface proteins. *J Infect Dis*. 1987;155:696.
122. Dworkin J, Blaser MJ. Molecular mechanisms of *Campylobacter fetus* surface layer protein expression. *Mol Microbiol*. 1997;26:433–440.
123. Blaser MJ, Smith PF, Repine JE, et al. Pathogenesis of *Campylobacter fetus* infections: failure of C3b to bind explains serum and phagocytosis resistance. *J Clin Invest*. 1988;81:1434–1444.
124. Wang E, Garcia MM, Blake MS, et al. Shift in S-layer protein expression responsible for antigenic variation in *Campylobacter fetus*. *J Bacteriol*. 1993;175:4979–4984.
125. Tummuru MKR, Blaser MJ. Rearrangement of *sapA* homologs with conserved and variable regions in *Campylobacter fetus*. *Proc Natl Acad Sci USA*. 1993;90:7265–7269.
126. Tu Z-C, Gaudreau C, Blaser MJ. Mechanisms underlying *Campylobacter fetus* pathogenesis in humans: surface-layer protein variation in relapsing infections. *J Infect Dis*. 2005;191:2082–2089.
127. Kirkpatrick BD, Lyon CE, Porter CK, et al. Lack of homologous protection against *Campylobacter jejuni* CG8421 in a human challenge model. *Clin Infect Dis*. 2013;57:1106–1113.
128. Walker RI, Caldwell MB, Lee EC, et al. Pathophysiology of *Campylobacter* enteritis. *Microbiol Rev*. 1985;50: 81–94.
129. Black RF, Perlman D, Clements ML, et al. Human volunteer studies with *C. jejuni*. In: Nachamkin I, Blaser MJ, Tompkins LS, eds. *Campylobacter fetus: Current Status and Future Trends*. Washington, DC: American Society for Microbiology; 1992:207–215.
130. Cawthraw SA, Feldman RA, Sayers AR, et al. Long-term antibody responses following human infection with *Campylobacter jejuni*. *Clin Exp Immunol*. 2002;130:101–106.
131. Johnson RJ, Wang SP, Shelton WR, et al. Persistent *Campylobacter jejuni* infection in an immunocompromised host. *Ann Intern Med*. 1984;100:832–834.
132. Melamed I, Bujanover Y, Igra YS, et al. *Campylobacter* enteritis in normal and immunodeficient children. *Am J Dis Child*. 1983;137:752–753.
133. Coker AO, Isokpehi RD, Thomas BN, et al. Human campylobacteriosis in developing countries. *Emerg Infect Dis*. 2002;8:237–244.
134. Blaser MJ, Smith PF, Kohler PF. Susceptibility of *Campylobacter* isolates to the bactericidal activity of human serum. *J Infect Dis*. 1985;151:227–235.
135. Havelaar AH, van Pelt W, Ang CW, et al. Immunity to *Campylobacter*: its role in risk assessment and epidemiology. *Crit Rev Microbiol*. 2009;35:1–22.
136. Huizinga R, Easton AS, Donachie AM, et al. Sialylation of *Campylobacter jejuni* lipo-oligosaccharides: impact on phagocytosis and cytokine production in mice. *PLoS ONE*. 2012;7:e34416.
137. Dalpke A, Frank J, Peter M, et al. Activation of Toll-like receptor 9 by DNA from different bacterial species. *Infect Immun*. 2006;74:940–946.
138. Watson RO, Galan JE. Signal transduction in *Campylobacter jejuni*-induced cytokine production. *Cell Microbiol*. 2005;7:655–665.
139. Keo T, Collins J, Kunwar P, et al. *Campylobacter* capsule and lipooligosaccharide confer resistance to serum and cationic antimicrobials. *Virulence*. 2011;2:30–40.
140. Blaser MJ, Smith PF, Repine JE, et al. Pathogenesis of *Campylobacter fetus* infections. Failure of encapsulated *Campylobacter fetus* to bind C3b explains serum and phagocytosis resistance. *J Clin Invest*. 1988;81:1434–1444.
141. Gazaigne L, Legrand P, Renaud B, et al. *Campylobacter fetus* bloodstream infection: risk factors and clinical features. *Eur J Clin Microbiol Infect Dis*. 2008;27:185–189.
142. Guerrant RL, Lahita RG, Winn EC Jr, et al. Campylobacteriosis in man: pathogenic mechanisms and review of 91 bloodstream infections. *Am J Med*. 1978;65:484.
143. Blaser MJ, Berkowitz ID, LaForce FM, et al. *Campylobacter* enteritis: clinical and epidemiologic features. *Ann Intern Med*. 1979;91:179.
144. Skirrow MB. *Campylobacter* enteritis: a "new" disease. *Br Med J*. 1977;2:9.
145. Karmali MA, Fleming PC. *Campylobacter* enteritis in children. *J Pediatr*. 1979;94:527.
146. Riddle MS, Gutierrez RL, Verdu EF, et al. The chronic gastrointestinal consequences associated with campylobacter. *Curr Gastroenterol Rep*. 2012;14:395–405.
147. McKinley MJ, Taylor M, Sangree MH. Toxic megacolon with *Campylobacter* colitis. *Conn Med*. 1980;44:496.
148. Tee W, Anderson BN, Ross BC, et al. Atypical campylobacters associated with gastroenteritis. *J Clin Microbiol*. 1987;25:1248–1252.
149. Steele TW, Sangster N, Lanser JA. DNA relatedness and biochemical features of *Campylobacter* spp. isolated in central and south Australia. *J Clin Microbiol*. 1985;22:71–74.
150. Drake AA, Gilchrist MJ, Washington JA II, et al. Diarrhea due to *Campylobacter fetus* subspecies *jejuni:* a clinical review of 73 cases. *Mayo Clin Proc*. 1981;56:414.
151. Michalak DM, Perrault J, Gilchrist MJ, et al. *Campylobacter fetus* ss. *jejuni*: a cause of massive lower gastrointestinal hemorrhage. *Gastroenterology*. 1980;79:742.
152. Anders BJ, Lauer BA, Paisley JW. *Campylobacter* gastroenteritis in neonates. *Am J Dis Child*. 1981; 135:900.

153. Goossens H, Henocque G, Kremp L, et al. Nosocomial outbreak of *Campylobacter jejuni* meningitis in newborn infants. *Lancet*. 1986;2:146–149.
154. Wright EP, Seager J. Convulsions associated with *Campylobacter* enteritis. *Br Med J*. 1980;281:454.
155. Orlicek SL, Welch DF, Kuhls TL. Septicemia and meningitis caused by *Helicobacter cinaedi* in a neonate. *J Clin Microbiol*. 1993;31:569–571.
156. Nielsen H, Hansen KK, Gradel KO, et al. Bacteraemia as a result of *Campylobacter* species: a population-based study of epidemiology and clinical risk factors. *Clin Microbiol Infect*. 2010;16:57–61.
157. Blaser MJ, Perez GP, Smith PF, et al. Extraintestinal *Campylobacter jejuni* and *Campylobacter coli* infections: host factors and strain characteristics. *J Infect Dis*. 1986;153:552.
158. Bhattacharyya R, Flores EJ, Azar MM. Case 30-2017—A 65-year-old woman with altered mental status, bacteremia, and acute liver failure. *N Engl J Med*. 2017;377:1274–1282.
159. Gilbert GL, Davoren RA, Cole ME, et al. Midtrimester abortion associated with septicaemia caused by *Campylobacter jejuni*. *Med J Aust*. 1981;1:585.
160. Mertens A, DeSmet M. *Campylobacter* cholecystitis. *Lancet*. 1979;1:1092.
161. Ezpeleta C, Rojo de Ursua P, Obregon F, et al. Acute pancreatitis associated with *Campylobacter jejuni* bacteremia. *Clin Infect Dis*. 1992;15:1050.
162. Feder HM, Rasoulpour M, Rodriquez AJ. *Campylobacter* urinary tract infection: value of the urine Gram stain. *JAMA*. 1986;256:2389.
163. Kerstens PJ, Endtz HP, Meis JF, et al. Erysipelas-like skin lesions associated with *Campylobacter jejuni* septicemia in patients with hypogammaglobulinemia. *Eur J Clin Microbiol Infect Dis*. 1992;11:842–847.
164. Bloomfield SJ, et al. Long-term colonization by *Campylobacter jejuni* within a human host: evolution, antimicrobial resistance, and adaptation. *J Infect Dis*. 2018;217:103–111.
165. Baqar S, et al. Recrudescent *Campylobacter jejuni* in an immunocompetent adult following experimental infection with a well-characterized organism. *Clin Vaccine Immunol*. 2010;17:80–86.
166. Mishu B, Blaser MJ. The role of *Campylobacter jejuni* infection in the initiation of Guillain-Barré syndrome. *Clin Infect Dis*. 1993;17:104–108.
167. Rees JH, Soudain SE, Gregory NA, et al. *Campylobacter jejuni* infection and Guillain-Barré syndrome. *N Engl J Med*. 1995;333:1374–1379.
168. Kaldor J, Speed BR. Guillain-Barré syndrome and *Campylobacter jejuni*: a serological study. *Br Med J (Clin Res Ed)*. 1984;288:1867–1870.
169. Mishu B, Ilyas AA, Koski CL, et al. Serologic evidence of *Campylobacter jejuni* infection preceding Guillain-Barré syndrome. *Ann Intern Med*. 1993;118:947–953.
170. Kuroki S, Saida T, Nukina M, et al. *Campylobacter jejuni* strains from patients with Guillain-Barré syndrome belong mostly to Penner serogroup 19 and contain β-*N*-acetylglucosamine residues. *Ann Neurol*. 1993;33:243–247.
171. Fujimoto S, Allos BM, Misawa N, et al. Restriction fragment length polymorphism analysis and random amplified polymorphic DNA analysis of *Campylobacter jejuni* strains isolated from patients with Guillain-Barré syndrome. *J Infect Dis*. 1997;176:1105–1108.
172. Allos BM, Lippy FT, Carlsen A, et al. *Campylobacter jejuni* strains from patients with Guillain-Barré syndrome. *Emerg Infect Dis*. 1998;4:263–268.
173. Loshaj-Shala A, Regazzoni L, Daci A, et al. Guillain Barre syndrome (GBS): new insights in the molecular mimicry between *C. jejuni* and human peripheral nerve (HPN) proteins. *J Neuroimmunol*. 2015;289:168–176.
174. Aspinall GO, Fujimoto S, McDonald AG, et al. Lipopolysaccharides from *Campylobacter jejuni* associated with Guillain-Barré syndrome patients mimic human gangliosides in structure. *Infect Immun*. 1994;62:2122–2125.
175. Ang CW, De Klerk MA, Endtz HP, et al. Guillain-Barré syndrome- and Miller Fisher syndrome-associated *Campylobacter jejuni* lipopolysaccharides induce anti-GM1 and GQ1b antibodies in rabbits. *Infect Immun*. 2001;69:2462–2469.
176. Goodyear CS, O'Hanlon GM, Plomp JJ, et al. Monoclonal antibodies raised against Guillain-Barré syndrome-associated *Campylobacter jejuni* lipopolysaccharides react with neuronal gangliosides and paralyze muscle-nerve preparations. *J Clin Invest*. 1999;104:697–708.
177. Hannu T, Kauppi M, Tuomala M, et al. Reactive arthritis following an outbreak of *Campylobacter jejuni* infection. *J Rheumatol*. 2004;31:528–530.
178. Pope JE, Krizova A, Garg AX, et al. *Campylobacter* reactive arthritis: a systematic review. *Semin Arthritis Rheum*. 2007;37:48–55.
179. Bremell T, Bjelle A, Svedhem A. Rheumatic symptoms following an attack of *Campylobacter* enteritis: a five year follow up. *Ann Rheum Dis*. 1991;50:934–948.
180. Lecuit M, Abachin E, Martin A, et al. Immunoproliferative small intestinal disease associated with *Campylobacter jejuni*. *N Engl J Med*. 2004;350:239–248.
181. Uzoigwe C. *Campylobacter* infections of the pericardium and myocardium. *Clin Microbiol Infect*. 2005;11:253–255.
182. Kotilainen P, Lehtopolku M, Hakanen AJ. Myopericarditis in a patient with *Campylobacter* enteritis: a case-report and literature review. *Scand J Infect Dis*. 2006;38:549–552.
183. Humphrey KS. *Campylobacter* infection and hepatocellular injury. *Lancet*. 1993;341:49.
184. Monselise A, Blickstein D, Ostfeld I, et al. A case of cellulitis complicating *Campylobacter jejuni* subspecies *jejuni* bacteremia and review of the literature. *Eur J Clin Microbiol Infect Dis*. 2004;23:718–721.
185. Carter JE, Cimolai N. IgA nephropathy associated with *Campylobacter jejuni* enteritis. *Nephron*. 1991;58:101–102.
186. Pacanowski J, Lalande V, Lacombe K, et al. *Campylobacter* bacteremia: clinical features and factors associated with fatal outcome. *Clin Infect Dis*. 2008;47:790–796.
187. Gazaigne L, Legrand P, Renaud B, et al. *Campylobacter fetus* bloodstream infection: risk factors and clinical features. *Eur J Clin Microbiol Infect Dis*. 2008;27:185–189.
188. Neuzil KM, Wang E, Haas D, et al. Persistence of *Campylobacter fetus* bacteremia associated with absence of opsonizing antibodies. *J Clin Microbiol*. 1994;32:1718–1720.
189. Haruyama A, Toyoda S, Kikuchi M, et al. *Campylobacter fetus* as Cause of Prosthetic Valve Endocarditis. *Tex Heart Inst J*. 2011;38:584–587.
190. Farrugia DC, Eykyn SJ, Smyth EG. *Campylobacter fetus* endocarditis: two case reports and review. *Clin Infect Dis*. 1994;18:443–446.
191. Miki K, Maekura R, Hiraga T, et al. Infective tricuspid valve endocarditis with pulmonary emboli caused by *Campylobacter fetus* after tooth extraction. *Intern Med*. 2005;44:1055–1059.
192. Melendez BA, Hollis HW Jr, Rehring TF. Mycotic popliteal aneurysm rupture secondary to *Campylobacter fetus*. *Ann Vasc Surg*. 2015;29:122, e9–122.e11.
193. Fujihara N, Takakura S, Saito T, et al. A case of perinatal sepsis by *Campylobacter fetus* subsp. *fetus* infection successfully treated with carbapenem – case report and literature review. *J Infect*. 2006;53:e199–e202.
194. Herve J, Aissa N, Legrand P, et al. *Campylobacter fetus* meningitis in a diabetic adult cured by imipenem. *Eur J Clin Microbiol Infect Dis*. 2004;23:722–724.
195. Cone LA, Dreisbach PB, Hirschberg J, et al. Cellulitis and septic arthritis caused by *Campylobacter fetus* and *Campylobacter jejuni*: report of 2 cases and review of the literature. *J Clin Rheumatol*. 2003;9:362–369.
196. Lawrence R, Nibbe AF, Levin S. Lung abscess secondary to *Vibrio fetus* malabsorption syndrome and acquired agammaglobulinemia. *Chest*. 1971;60:191.
197. Meier PA, Dooley DP, Jorgensen JH, et al. Development of quinolone-resistant *Campylobacter fetus* bacteremia in human immunodeficiency virus-infected patients. *J Infect Dis*. 1998;177:951–954.
198. Wagenaar JA, van Bergen MAP, Blaser MJ, et al. *Campylobacter fetus* infections in humans: exposure and disease. *Clin Infect Dis*. 2014;58:1579–1586.
199. Wagenaar JA, van Bergen MA, Blaser MJ, et al. *Campylobacter fetus* infections in humans: exposure and disease. *Clin Infect Dis*. 2014;58:1579.
200. Tu Z, Zeitlin G, Gagner J-P, et al. *Campylobacter fetus* of reptile origin as a human pathogen. *J Clin Microbiol*. 2004;42:4405–4407.
201. Dingle KE, Blaser MJ, Tu ZC, et al. Genetic relationships among reptile and mammalian *Campylobacter fetus* by multilocus sequence typing. *J Clin Microbiol*. 2010;48:977–980.
202. Patrick ME, Gilbert MJ, Blaser MJ, et al. Human infections with new subspecies of *Campylobacter fetus*. *Emerg Infect Dis*. 2013;19:1678–1680.
203. Marchand-Senécal X, Bekal S, Pilon PA, et al. *Campylobacter fetus* cluster among men who have sex with men, Montreal, Quebec, Canada, 2014-2016. *Clin Infect Dis*. 2017;65:1751.
204. Benjamin JS, Leaper S, Owen RJ, et al. Description of *Campylobacter laridis*, a new species comprising the nalidixic acid resistant thermophilic *Campylobacter* (NARTC group). *Curr Microbiol*. 1983;8:231–238.
205. Simor AE, Wilcox L. Enteritis associated with *Campylobacter laridis*. *J Clin Microbiol*. 1987;25:10–12.
206. Quinn TC, Goodell SE, Fennell C, et al. Infections with *Campylobacter jejuni* and *Campylobacter*-like organisms in homosexual men. *Ann Intern Med*. 1984;101:187–192.
207. Saito S, Tsukahara M, Ohkusu K, et al. *Helicobacter fennelliae* bacteremia: three case reports and literature review. *Medicine (Baltimore)*. 2016;95:e3556.
208. Pérez-Santiago J, Ramirez-Gaona M, Holm-Kennedy R, et al. Bacteremia and skin infections in four patients caused by *Helicobacter*-like organisms. *Open Forum Infect Dis*. 2017;4:ofx074.
209. Kiehlbauch JA, Tauxe RV, Baker CN, et al. *Helicobacter cinaedi*–associated bacteremia and cellulitis in immunocompromised patients. *Ann Intern Med*. 1994;121:90–93.
210. Goosens H, Pot B, Vlaes L, et al. Characterization and description of "*Campylobacter upsaliensis*" isolated from human feces. *J Clin Microbiol*. 1990;28:1039–1046.
211. Nakamura I, Omori N, Umeda A, et al. First case report of fatal sepsis due to *Campylobacter upsaliensis*. *J Clin Microbiol*. 2015;53:713.
212. Gaudreau C, Lamothe F. *Campylobacter upsaliensis* isolated from a breast abscess. *J Clin Microbiol*. 1992;30:1354–1356.
213. Damborg P, Guardabassi L, Pedersen K, et al. Comparative analysis of human and canine *Campylobacter upsaliensis* isolates by amplified fragment length polymorphism. *J Clin Microbiol*. 2008;46:1504–1506.
214. Edmonds P, Patton CM, Griffin PM, et al. *Campylobacter hyointestinalis* associated with human gastrointestinal disease in the United States. *J Clin Microbiol*. 1987;25:685–691.
215. Vandamme P, Pugina P, Benzi G, et al. Outbreak of recurrent abdominal cramps associated with *Arcobacter butzleri* in an Italian school. *J Clin Microbiol*. 1992;30:2335–2337.
216. Sorlin P, van Damme P, Nortier J, et al. Recurrent "*Flexispira rappini*" bacteremia in an adult patient undergoing hemodialysis: case report. *J Clin Microbiol*. 1999;37:1319–1323.
217. Abbott SL, Waddington M, Lindquist D, et al. Description of *Campylobacter curvus* and *C. curvus*–like strains associated with sporadic episodes of bloody gastroenteritis and Brainerd's diarrhea. *J Clin Microbiol*. 2005;43:585–588.
218. Wetsch NM, Somani K, Tyrrell GJ, et al. *Campylobacter curvus*–associated hepatic abscess: a case report. *J Clin Microbiol*. 2006;44:1909–1911.
219. Chua K, Gurtler V, Montgomery J, et al. *Campylobacter insulaenigrae* causing septicaemia and enteritis. *J Med Microbiol*. 2007;56:1565–1567.
220. Nielsen HL, Engberg J, Ejlertsen T, et al. Short-term and medium-term clinical outcomes of *Campylobacter concisus* infection. *Clin Microbiol Infect*. 2012;18:E459–E465.
221. Kalischuk LD, Inglis GD. Comparative genotypic and pathogenic examination of *Campylobacter concisus* isolates from diarrheic and non-diarrheic humans. *BMC Microbiol*. 2011;11:53.
222. Man SM, Kaakoush NO, Leach ST, et al. Host attachment, invasion, and stimulation of proinflammatory cytokines by *Campylobacter concisus* and other non-*Campylobacter jejuni Campylobacter* species. *J Infect Dis*. 2010;202:1855–1865.
223. Ismail Y, Mahendran V, Octavia S, et al. Investigation of the enteric pathogenic potential of oral *Campylobacter concisus* strains isolated from patients with inflammatory bowel disease. *PLoS ONE*. 2012;7:e38217.
224. Man SM, Zhang L, Day AS, et al. *Campylobacter concisus* and other *Campylobacter* species in children with newly diagnosed Crohn's disease. *Inflamm Bowel Dis*. 2010;16:1008–1016.
225. Hess DL, Pettersson AM, Rijnsburger MC, et al. Gastroenteritis caused by *Campylobacter concisus*. *J Med Microbiol*. 2012;61:746–749.
226. Kovach Z, Kaakoush NO, Lamb S, et al. Immunoreactive proteins of *Campylobacter concisus*, an emergent intestinal pathogen. *FEMS Immunol Med Microbiol*. 2011;63:387–396.
227. Mahendran V, Riordan SM, Grimm MC, et al. Prevalence of *Campylobacter* species in adult Crohn's disease and the preferential colonization sites of *Campylobacter* species in the human intestine. *PLoS ONE*. 2011;6:e25417.
228. Burgos-Portugal JA, Kaakoush NO, Raftery MJ, et al. Pathogenic potential of *Campylobacter ureolyticus*. *Infect Immun*. 2012;80:883–890.
229. Man SM. The clinical importance of emerging *Campylobacter* species. *Nat Rev Gastroenterol Hepatol*. 2011;8:669–685.
230. Bullman S, Corcoran D, O'Leary J, et al. *Campylobacter ureolyticus*: an emerging gastrointestinal pathogen? *FEMS Immunol Med Microbiol*. 2011;61:228–230.
231. O'Donovan D, Corcoran GD, Lucey B, et al. *Campylobacter ureolyticus:* a portrait of the pathogen. *Virulence*. 2014;5:498–506.
232. Kaur T, Singh J, Huffman MA, et al. *Campylobacter troglodytis* sp. nov., isolated from feces of

human-habituated wild chimpanzees *(Pan troglodytes schweinfurthii)* in Tanzania. *Appl Environ Microbiol*. 2011;77:2366–2373.

233. Debruyne L, On SL, De Brandt E, et al. Novel *Campylobacter lari*–like bacteria from humans and molluscs: description of *Campylobacter peloridis* sp. nov., *Campylobacter lari* subsp. *concheus* subsp. nov. and *Campylobacter lari* subsp. *lari* subsp. nov. *Int J Syst Evol Microbiol*. 2009;59:1126–1132.
234. Shinha T. Fatal bacteremia caused by *Campylobacter gracilis*, United States. *Emerg Infect Dis*. 2015;21:1084–1085.
235. Fitzgerald C, Tu ZC, Patrick M, et al. *Campylobacter fetus* subsp. *testudinum* subsp. nov., isolated from humans and reptiles. *Int J Syst Evol Microbiol*. 2014;64:2944–2948.
236. Blaser MJ. *Campylobacter fetus*: emerging infection and model system for bacterial pathogenesis at mucosal surfaces. *Clin Infect Dis*. 1998;27:256–258.
237. Poppert S, Haas M, Yildiz T, et al. Identification of thermotolerant *Campylobacter* species by fluorescence in situ hybridization. *J Clin Microbiol*. 2008;46:2133–2136.
238. Paisley JW, Mirrett S, Lauer BA, et al. Darkfield microscopy of human feces for the presumptive diagnosis of *Campylobacter* enteritis. *J Clin Microbiol*. 1982;15:61.
239. Sazie ESM, Titus AE. Rapid diagnosis of *Campylobacter* enteritis. *Ann Intern Med*. 1982;96:62.
240. Ternhag A, Asikainen T, Giesecke J, et al. A meta-analysis on the effects of antibiotic treatment on duration of symptoms caused by infection with *Campylobacter* species. *Clin Infect Dis*. 2007;44:696.
241. Ruiz-Palacios GM. The health burden of *Campylobacter* infection and the impact of antimicrobial resistance: playing chicken. *Clin Infect Dis*. 2007;44:701.
242. Anders BJ, Lauer BA, Paisley JW, et al. Double-blind placebo controlled trial of erythromycin for treatment of *Campylobacter* enteritis. *Lancet*. 1982;1:131.
243. Vanhoof R, Gordts B, Dierickx R, et al. Bacteriostatic and bactericidal activities of 24 antimicrobial agents against *Campylobacter fetus* subsp. *jejuni*. *Antimicrob Agents Chemother*. 1980;18:118.
244. Shane AL, Mody RK, Crump JA, et al. 2017 Infectious Diseases Society of America clinical practice guidelines for the diagnosis and management of infectious diarrhea. *Clin Infect Dis*. 2017;65:e45.
245. Centers for Disease Control and Prevention. The pre-travel consultation: travelers' diarrhea. The Yellow Book: CDC health information for international travel, 2018. https://wwwnc.cdc.gov/travel/yellowbook/2018/the-pre-travel-consultation/travelers-diarrhea. Accessed September 5, 2017.
246. Riddle MS, Connor BA, Beeching NJ, et al. Guidelines for the prevention and treatment of travelers' diarrhea: a graded expert panel report. *J Travel Med*. 2017;24:S57–S74.
247. Committee on Infectious Diseases; American Academy of Pediatrics, Kimberlin David W, Brady Michael T., et al. Campylobacter infections. In: *Red Book. 2018-2021 Report of the Committee on Infectious Diseases*. 31st ed. Itasca, IL: American Academy of Pediatrics; 2018.
248. Lachance N, Gaudreau C, Lamothe F, et al. Susceptibilities of β-lactamase-positive and -negative strains of *Campylobacter coli* to β-lactam agents. *Antimicrob Agents Chemother*. 1993;37:1174–1176.
249. Aguilar-Company J, Los-Arcos I, Pigrau C, et al. Potential use of fosfomycin-tromethamine for treatment of recurrent *Campylobacter* species enteritis. *Antimicrob Agents Chemother*. 2016;60:4398.
250. Taylor DN, Blaser MJ, Echeverria P, et al. Erythromycin-resistant *Campylobacter* infections in Thailand. *Antimicrob Agents Chemother*. 1987;31:438–442.
251. Smith GS, Blaser MJ. Fatalities associated with *Campylobacter jejuni* infections. *JAMA*. 1985;253:2873.
252. Hammarström V, Smith CIE, Hammarström L. Oral immuno-globulin treatment in *Campylobacter jejuni* enteritis. *Lancet*. 1993;341:1036.
253. Francioli P, Herzstein J, Grob J-P, et al. *Campylobacter fetus* subspecies *fetus* bacteremia. *Arch Intern Med*. 1985;145:289–292.
254. Tremblay C, Gaudreau C, Lorange M. Epidemiology and antimicrobial susceptibilities of 111 *Campylobacter fetus* subsp. *fetus* strains isolated in Quebec, Canada from 1983 to 2000. *J Clin Microbiol*. 2003;41:463–466.
255. Velazquez JB, Jimenez A, Chomon B, et al. Incidence and transmission of antibiotic resistance in *Campylobacter jejuni* and *Campylobacter coli*. *J Antimicrob Chemother*. 1995;35:173–178.
256. Nirdnoy W, Mason CJ, Guerry P. Mosaic structure of a multiple-drug-resistant, conjugative plasmid from *Campylobacter jejuni*. *Antimicrob Agents Chemother*. 2005;49:2454–2459.
257. Lin J, Michel LO, Zhang Q. CmeABC functions as a multidrug efflux system in *Campylobacter jejuni*. *Antimicrob Agents Chemother*. 2002;46:2124–2131.
258. Gootz TD, Martin BA. Characterization of high-level quinolone resistance in *Campylobacter jejuni*. *Antimicrob Agents Chemother*. 1991;35:840–845.
259. Manavathu EK, Hiratsuka K, Taylor DE. Nucleotide sequence analysis and expression of a tetracycline-resistance gene from *Campylobacter jejuni*. *Gene*. 1988;62:17–26.
260. Gibreel A, Kos VN, Keelan M, et al. Macrolide resistance in *Campylobacter jejuni* and *Campylobacter coli:* molecular mechanism and stability of the resistance phenotype. *Antimicrob Agents Chemother*. 2005;49:2753–2759.
261. Mamelli L, Prouzet-Mauleon V, Pages JM, et al. Molecular basis of macrolide resistance in *Campylobacter:* role of efflux pumps and target mutations. *J Antimicrob Chemother*. 2005;56:491–497.
262. Gibreel A, Skold O. High-level resistance to trimethoprim in clinical isolates of *Campylobacter jejuni* by acquisition of foreign genes (*dfr1* and *dfr9*) expressing drug-insensitive dihydrofolate reductases. *Antimicrob Agents Chemother*. 1998;42:3059–3064.
263. Page WJ, Huyer G, Huyer M, et al. Characterization of the porins of *Campylobacter jejuni* and *Campylobacter coli* and implications for antibiotic susceptibility. *Antimicrob Agents Chemother*. 1989;33:297–303.
264. Pumbwe L, Randall LP, Woodward MJ, et al. Expression of the efflux pump genes *cmeB, cmeF* and the porin gene *porA* in multiple-antibiotic-resistant *Campylobacter jejuni*. *J Antimicrob Chemother*. 2004;54:341–347.
265. Jeon B, Muraoka W, Scupham A, et al. Roles of lipooligosaccharide and capsular polysaccharide in antimicrobial resistance and natural transformation of *Campylobacter jejuni*. *J Antimicrob Chemother*. 2009;63:462–468.
266. Pinto-Alphandary H, Mabilat C, Courvalin P. Emergence of aminoglycoside resistance genes *aadA* and *aadE* in the genus *Campylobacter*. *Antimicrob Agents Chemother*. 1990;34:1294–1296.
267. Alfredson DA, Korolik V. Isolation and expression of a novel molecular class D beta-lactamase, OXA-61, from *Campylobacter jejuni*. *Antimicrob Agents Chemother*. 2005;49:2515–2518.
268. Griggs DJ, Peake L, Johnson MM, et al. Beta-lactamase-mediated beta-lactam resistance in *Campylobacter species:* prevalence of Cj0299 (bla OXA-61) and evidence for a novel beta-lactamase in *C. jejuni*. *Antimicrob Agents Chemother*. 2009;53:3357–3364.
269. Lachance N, Gaudreau C, Lamothe F, et al. Role of the beta-lactamase of *Campylobacter jejuni* in resistance to beta-lactam agents. *Antimicrob Agents Chemother*. 1991;35:813–818.
270. Lin J, Sahin O, Michel LO, et al. Critical role of multidrug efflux pump CmeABC in bile resistance and in vivo colonization of *Campylobacter jejuni*. *Infect Immun*. 2003;71:4250–4259.
271. Pumbwe L, Piddock LJ. Identification and molecular characterisation of CmeB, a *Campylobacter jejuni* multidrug efflux pump. *FEMS Microbiol Lett*. 2002;206:185–189.
272. Akiba M, Lin J, Barton YW, et al. Interaction of CmeABC and CmeDEF in conferring antimicrobial resistance and maintaining cell viability in *Campylobacter jejuni*. *J Antimicrob Chemother*. 2006;57:52–60.
273. Jeon B, Wang Y, Hao H, et al. Contribution of CmeG to antibiotic and oxidative stress resistance in *Campylobacter jejuni*. *J Antimicrob Chemother*. 2011;66:79–85.
274. Guerrant RL, Van Gilder T, Steiner TS, et al. Practice guidelines for the management of infectious diarrhea. *Clin Infect Dis*. 2001;32:331–351.
275. European Food Safety Authority. The European Union Summary Report on antimicrobial resistance in zoonotic and indicator bacteria from humans, animals and food in 260 2010. *EFSA J*. 2012;10:2598–2830.
276. Gupta A, Nelson JM, Barrett TJ, et al. Antimicrobial resistance among *Campylobacter strains,* United States, 1997-2001. *Emerg Infect Dis*. 2004;10:1102–1109.
277. US Department of Health and Human Services, Centers for Disease Control and Prevention. Antibiotic resistance threats in the United States, 2013. https://www.cdc.gov/drugresistance/pdf/ar-threats-2013-508.pdf.
278. Centers for Disease Control and Prevention (CDC). *NARMS Now: Human Data*. Atlanta: CDC; 2018.
279. Post A, Martiny D, van Waterschoot N, et al. Antibiotic susceptibility profiles among Campylobacter isolates obtained from international travelers between 2007 and 2014. *Eur J Clin Microbiol Infect Dis*. 2017;36:2101–2107.
280. Davidson D In the matter of enrofloxacin for poultry: withdrawal of approval of Bayer Corporation's new animal drug application 1 (NADA) 140-828 (Baytril). FDA Docket No. 00N-1571, 2004.
281. Bester LA, Essack SY. Observational study of the prevalence and antibiotic resistance of *Campylobacter* spp. from different poultry production systems in KwaZulu-Natal, South Africa. *J Food Prot*. 2012;75:154–159.
282. Smith SI, Sansa TI, Coker AO. Antibiotic susceptibility patterns and beta-lactamase production of animal and human isolates of *Campylobacter* in Lagos, Nigeria. *Z Naturforsch [C]*. 1999;54:583–586.
283. Drlica K, Zhao X. DNA gyrase, topoisomerase IV, and the 4-quinolones. *Microbiol Mol Biol Rev*. 1997;61:377–392.
284. Willmott CJ, Critchlow SE, Eperon IC, et al. The complex of DNA gyrase and quinolone drugs with DNA forms a barrier to transcription by RNA polymerase. *J Mol Biol*. 1994;242:351–363.
285. Ellis-Pegler RB, Hyman LK, Ingram RJ, et al. A placebo controlled evaluation of lomefloxacin in the treatment of bacterial diarrhoea in the community. *J Antimicrob Chemother*. 1995;36:259–263.
286. Luo N, Pereira S, Sahin O, et al. Enhanced in vivo fitness of fluoroquinolone-resistant *Campylobacter jejuni* in the absence of antibiotic selection pressure. *Proc Natl Acad Sci USA*. 2005;102:541–546.
287. Segreti J, Gootz TD, Goodman LJ, et al. High-level quinolone resistance in clinical isolates of *Campylobacter jejuni*. *J Infect Dis*. 1992;165:667–670.
288. Wretlind B, Stromberg A, Ostlund L, et al. Rapid emergence of quinolone resistance in *Campylobacter jejuni* in patients treated with norfloxacin. *Scand J Infect Dis*. 1992;24:685–686.
289. Wang Y, Huang WM, Taylor DE. Cloning and nucleotide sequence of the *Campylobacter jejuni gyrA* gene and characterization of quinolone resistance mutations. *Antimicrob Agents Chemother*. 1993;37:457–463.
290. Ge B, McDermott PF, White DG, et al. Role of efflux pumps and topoisomerase mutations in fluoroquinolone resistance in *Campylobacter jejuni* and *Campylobacter coli*. *Antimicrob Agents Chemother*. 2005;49:3347–3354.
291. Luo N, Sahin O, Lin J, et al. In vivo selection of *Campylobacter* isolates with high levels of fluoroquinolone resistance associated with *gyrA* mutations and the function of the CmeABC efflux pump. *Antimicrob Agents Chemother*. 2003;47:390–394.
292. Yan M, Sahin O, Lin J, et al. Role of the CmeABC efflux pump in the emergence of fluoroquinolone-resistant *Campylobacter* under selection pressure. *J Antimicrob Chemother*. 2006;58:1154–1159.
293. Piddock LJ, Ricci V, Pumbwe L, et al. Fluoroquinolone resistance in *Campylobacter* species from man and animals: detection of mutations in topoisomerase genes. *J Antimicrob Chemother*. 2003;51:19–26.
294. Cagliero C, Mouline C, Cloeckaert A, et al. Synergy between efflux pump CmeABC and modifications in ribosomal proteins L4 and L22 in conferring macrolide resistance in *Campylobacter jejuni* and *Campylobacter coli*. *Antimicrob Agents Chemother*. 2006;50:3893–3896.
295. Lin J, Yan M, Sahin O, et al. Effect of macrolide usage on emergence of erythromycin-resistant *Campylobacter* isolates in chickens. *Antimicrob Agents Chemother*. 2007;51:1678–1686.
296. Gibreel A, Taylor DE. Macrolide resistance in *Campylobacter jejuni* and *Campylobacter coli*. *J Antimicrob Chemother*. 2006;58:243–255.
297. Lehtopolku M, Kotilainen P, Haanpera-Heikkinen M, et al. Ribosomal mutations as the main cause of macrolide resistance in *Campylobacter jejuni* and *Campylobacter coli*. *Antimicrob Agents Chemother*. 2011;55:5939–5941.
298. Perez-Boto D, Lopez-Portoles JA, Simon C, et al. Study of the molecular mechanisms involved in high-level macrolide resistance of Spanish *Campylobacter jejuni* and *Campylobacter coli* strains. *J Antimicrob Chemother*. 2010;65:2083–2088.
299. Gibreel A, Wetsch NM, Taylor DE. Contribution of the CmeABC efflux pump to macrolide and tetracycline resistance in *Campylobacter jejuni*. *Antimicrob Agents Chemother*. 2007;51:3212–3216.
300. Payot S, Avrain L, Magras C, et al. Relative contribution of target gene mutation and efflux to fluoroquinolone and erythromycin resistance, in French poultry and pig isolates of *Campylobacter coli*. *Int J Antimicrob Agents*. 2004;23:468–472.
301. Logue CM, Danzeisen GT, Sherwood JS, et al. Repeated therapeutic dosing selects macrolide-resistant *Campylobacter* spp. in a turkey facility. *J Appl Microbiol*. 2010;109:1379–1388.
302. Almofti YA, Dai M, Sun Y, et al. Impact of erythromycin resistance on the virulence properties and fitness of *Campylobacter jejuni*. *Microb Pathog*. 2011;50:336–342.
303. Han F, Pu S, Wang F, et al. Fitness cost of macrolide resistance in *Campylobacter jejuni*. *Int J Antimicrob Agents*. 2009;34:462–466.

304. Hao H, Dai M, Wang Y, et al. 23S rRNA mutation A2074C conferring high-level macrolide resistance and fitness cost in *Campylobacter jejuni*. *Microb Drug Resist*. 2009;15:239–244.
305. Zeitouni S, Collin O, Andraud M, et al. Fitness of macrolide resistant *Campylobacter coli* and *Campylobacter jejuni*. *Microb Drug Resist*. 2012;18:101–108.
306. Corcoran D, Quinn T, Cotter L, et al. An investigation of the molecular mechanisms contributing to high-level erythromycin resistance in *Campylobacter*. *Int J Antimicrob Agents*. 2006;27:40–45.
307. Jeon B, Zhang Q. Sensitization of *Campylobacter jejuni* to fluoroquinolone and macrolide antibiotics by antisense inhibition of the CmeABC multidrug efflux transporter. *J Antimicrob Chemother*. 2009;63:946–948.
308. Caldwell DB, Wang Y, Lin J. Development, stability, and molecular mechanisms of macrolide resistance in *Campylobacter jejuni*. *Antimicrob Agents Chemother*. 2008;52:3947–3954.
309. Kurincic M, Botteldoorn N, Herman L, et al. Mechanisms of erythromycin resistance of *Campylobacter* spp. isolated from food, animals and humans. *Int J Food Microbiol*. 2007;120:186–190.

217 *Helicobacter pylori* and Other Gastric *Helicobacter* Species

Timothy L. Cover and Martin J. Blaser

SHORT VIEW SUMMARY

Definition
- *Helicobacter pylori* is a gram-negative bacterium that persistently colonizes the human stomach.
- Most *H. pylori*–colonized persons remain asymptomatic, but the presence of *H. pylori* is associated with an increased risk of peptic ulceration and gastric cancer.
- Increasing evidence shows benefits associated with gastric *H. pylori* colonization in addition to the known disease risks.

Epidemiology
- *H. pylori* is present in humans throughout the world, but the prevalence is variable and depends on age and geography.
- The prevalence of *H. pylori* in developed countries has been declining over the past several decades.

Microbiology
- *H. pylori* is a urease-positive, gram-negative, spiral-shaped bacterium.

Diagnosis
- Noninvasive diagnostic approaches include serology, urea breath test, and stool antigen test.
- Invasive diagnostic approaches are endoscopy and analysis of gastric tissue.

Therapy
- Antimicrobial treatment is indicated in patients with peptic ulceration or patients considered to have a high risk of gastric cancer.
- Treatment consists of a combination of antimicrobials and often includes an agent to diminish gastric acidity.
- An increasing proportion of strains show resistance to antibiotics. Knowledge of antimicrobial susceptibility allows optimization of treatment regimens.

Prevention
- Prevention of *H. pylori* acquisition by immunization is not routinely practiced. An important focus is on the prevention of *H. pylori*–associated illnesses, including gastric adenocarcinoma.

Helicobacter pylori (formerly known as *Campylobacter pylori* or *pyloridis*) was first isolated from humans in 1982.[1,2] This highly motile, curved, gram-negative rod lives within the mucus layer overlying the gastric mucosal epithelium.[3] *H. pylori* is commonly found in the human stomach, and when present, it is usually the single dominant species. Essentially all persons colonized with *H. pylori* have a cellular infiltrate in the lamina propria of the gastric antrum and fundus.[4,5] The first isolation of *H. pylori* in pure culture and recognition of its association with gastritis and peptic ulcer disease led to the awarding of the Nobel Prize in Medicine in 2005 to Barry Marshall and Robin Warren, two physicians in Australia. The presence of *H. pylori* increases the risk of peptic ulcer disease[6–8] and gastric cancer,[9–13] but also may be associated with health benefits, including a decreased risk of esophageal reflux and its consequences and protection against childhood asthma and related disorders.[9,14–18] With the development of effective therapies to eradicate *H. pylori,* physicians are faced with the challenge of determining which patients will benefit from therapy and which may be harmed.

MICROBIOLOGY

H. pylori organisms are small (0.5–1.0 μm in width and 2.5–4.0 μm in length), curved, microaerophilic, gram-negative rods.[2,19] Because they closely resemble members of the genus *Campylobacter,* they were initially considered to belong to that genus. However, multiple genotypic and phenotypic characteristics are different from those of campylobacters, and a new genus, *Helicobacter,* was established.[19] Most mammals studied to date are colonized with one or more distinct *Helicobacter* species. Examples include *Helicobacter mustelae* in ferrets,[19,20] *Helicobacter felis* in dogs and cats,[20] *Helicobacter muridarum* in mice, *Helicobacter nemestrinae* in nonhuman primates, and *Helicobacter acinonychis* in cheetahs.[21] Individual *Helicobacter* species preferentially colonize either the stomach or the intestinal tract, and typically are adapted for colonization of specific mammalian hosts.

H. pylori is the most important human-associated species from a medical perspective and may be considered the prototype for this group of gastric organisms. Related gastric bacteria that colonize the human stomach are considered in a separate section of this chapter. *Helicobacter fennelliae* and *Helicobacter cinaedi* are intestinal organisms that cause diarrheal illnesses, and *H. cinaedi* also can cause bacteremia and cellulitis, especially in immunocompromised patients.[22] Because the clinical features of these infections resemble those of *Campylobacter* spp., they are discussed in Chapter 216. *Helicobacter* spp., such as *Helicobacter hepaticus, Helicobacter bilis,* and *Helicobacter rappini,* have been identified in the colon and biliary tract of rodents,[23] and there is human carriage as well. Preliminary evidence suggests that these species might colonize the diseased human biliary tract,[24,25] but whether they contribute to biliary pathology is uncertain.[26] Thus there are both gastric and intestinal residential *Helicobacter* species in humans. In contrast to *H. pylori,* the residential intestinal species are not known to cause disease in nonimmunocompromised humans.

H. pylori cells are highly motile, with a rapid corkscrew motion, and have multiple, polar, sheathed flagella.[19] Although these cells are classically curved or spiral in fresh cultures, spherical (coccoid) forms are present in older cultures. The major biochemical properties of *H. pylori* and several related bacteria are shown in Table 217.1. A prominent biochemical characteristic of gastric helicobacters is production of urease. *H. pylori* urease is a hexadimer consisting of 61- and 28-kDa subunits, both of which are essential for activity.[27]

The complete genome sequences of hundreds of different *H. pylori* strains have been determined,[28–30] and the transcriptome of one strain has been analyzed in detail.[31] Comparison of these genome sequences has permitted the definition of a core genome,[32–34] and numerous strain-specific genes have been identified, including genes that encode restriction-modification enzymes[35] and others that encode cell surface components.[36,37] In comparison to many other bacterial species, *H. pylori*

TABLE 217.1 Biochemical Characteristics of *Helicobacter pylori* and Related Bacteria

CHARACTERISTIC	*H. PYLORI*	*HELICOBACTER MUSTELAE*	*HELICOBACTER FELIS*	*CAMPYLOBACTER JEJUNI*
Urease	+	+	+	–
Catalase	+	+	+	+
Oxidase	+	+	+	+
H_2S production	–	–	–	+
Guanosine plus cytosine content (%)	35–38	36	42.5	33–36
Hippurate hydrolysis	–	–	–	+
Nitrate reduction	–	+	+	+
Resistance to nalidixic acid (30-µg disk)	+	–	+	–
Cephalothin (30-µg disk)	–	+	–	+
Growth at 42°C	–	+	+	+
Growth at 37°C	+	+	+	+
Growth at 25°C	–	–	–	–

Data from Goodwin et al.,[19] Paster et al.,[20] Eaton et al.,[21] and Kiehlbauch et al.[22]

has few two-component regulatory systems. Frameshift mutations occur commonly within open reading frames encoding certain *H. pylori* proteins, which suggests that *H. pylori* may use mutation to control phenotype, with the host selecting for the "most fit" organism within a particular environmental niche.[38,39] Plasmids are present in most *H. pylori* isolates; they vary in size, and their functional properties are mostly unknown at present. Bacteriophages also have been detected in *H. pylori*.[40]

Although *H. pylori* is highly homogeneous in the biochemical characteristics analyzed in clinical microbiology, including urease, oxidase, and catalase positivity, there is substantial genetic variation among strains, both in gene content and in nucleotide sequences of individual genes.[41,42] Humans may be simultaneously colonized with more than one strain of *H. pylori,* and as many as five different strains have been identified in an individual subject.[43–45] *H. pylori* strains are naturally competent (i.e., able to take up heterologous DNA) and frequently undergo intergenomic and intragenomic recombination.[42,45,46] The strain-specific restriction-modification systems diminish recombination and may facilitate colonization of a host by several different strains simultaneously.[35,47,48] Because of point mutations and recombination, *H. pylori* are among the most varied of all species in the human biosphere.[49–52]

Distinct clonal lineages of *H. pylori* are not readily identifiable when comparing strains from unrelated humans,[45] but multilocus sequence typing has revealed the existence of multiple *H. pylori* populations with distinct geographic distributions.[53] Classification of strains by this approach is useful for anthropologic studies, and may be relevant to differences in pathogenicity of strains in hosts of different ethnicities, as described in recent studies from South America.[54] The most important dichotomy among *H. pylori* strains is the presence or absence of the cytotoxin-associated genes (*cag*) pathogenicity island, a 35- to 40-kilobase chromosomal region encoding the secreted protein CagA and genes encoding components of a type IV secretion system that mediates delivery of CagA into host cells.[55–59] Both $cagA^+$ and $cagA^-$ strains are present in *H. pylori* populations in most parts of the world,[58] which suggests that this chromosomal region was acquired in the distant past. The *cag* status of an *H. pylori* strain is relevant to the risk of a number of clinical outcomes (as discussed later). Another heterogeneous locus that provides a basis for typing strains is *vacA,* a conserved gene that encodes a secreted protein (vacuolating cytotoxin) that interacts with epithelial cells.[60] Three regions of *vacA* have major polymorphisms: the *s* region (with allelic types s1 and s2), the *m* region (with allelic types m1 and m2), and the *i* region (with allelic types i1 and i2).[61–63] Type s1 *vacA* genotypes are strongly linked to the presence of *cagA,* and therefore $cagA^+$ and *vacA* s1–positive genotypes are each associated with similar clinical outcomes.[64]

EPIDEMIOLOGY

H. pylori has been isolated from persons in all parts of the world.[53,65,66] Similar organisms have been isolated from primates, but other animal sources for *H. pylori* have not been identified, nor have reservoirs been found in food, soil, or water. It now appears likely that humans are the major, if not sole, reservoir for *H. pylori.* Analyses of genetic heterogeneity among strains indicate that *H. pylori* has been present in humans for at least 100,000 years, if not longer, and the current geographic distribution of *H. pylori* alleles reflects ancient migrations of human populations.[53,66–68] The recent finding of *H. pylori* DNA in a 5300-year-old corpse, frozen in ice, provides further evidence for the long-standing association of *H. pylori* with humans.[69] These data support the notion that *H. pylori* is indigenous to humans, as its relatives are to other mammals.[70] Prolonged coevolution of *H. pylori* with human populations may be linked to reduced pathogenicity. For example, a study from Colombia found that African-ancestry *H. pylori* was relatively benign in humans of African ancestry but was associated with higher risk of gastric cancer in individuals of Amerindian ancestry.[54]

The high prevalence of colonization among persons in settings where sanitary conditions are suboptimal, including institutions for individuals with intellectual disabilities and orphanages, suggests that horizontal fecal-oral transmission or oral-oral transmission occurs.[65,71] *H. pylori* has occasionally been isolated from feces, especially from children.[72] *H. pylori* has also been isolated from dental plaque,[43] and *H. pylori* DNA may be detected in saliva by the polymerase chain reaction assay,[73] which raises the possibility of oral-oral transmission as well. Studies of persons attending clinics for either sexually transmitted diseases or infertility indicate that sexual transmission does not occur very frequently, if at all.[74] The relative contribution of fecal-oral, oral-oral, or vomitus-oral[75] transmission of *H. pylori* is not known. *H. pylori* infection clusters in families,[76] and the presence of the organism in a child is highly associated with large family size and older siblings.[77,78] On occasion, transmission occurs from person to person via improperly cleaned endoscopes.[79]

In most populations, *H. pylori* appears to be almost universally acquired during childhood,[65,80] but not in the first year of life.[81] Once acquired, *H. pylori* can persistently colonize the stomach for decades or for an entire lifetime. *H. pylori* has also been found to transiently colonize children and nonhuman primates, a phenomenon that may be associated with less pathogenic strains,[82] or the selective bottleneck of transmission to a new host, with failure to sufficiently adapt to sustain colonization. The prevalence of *H. pylori* colonization varies considerably among different populations around the world[83] and is dependent on age[5,71] and socioeconomic development (Fig. 217.1). In developing countries, by age 10 years, more than 70% of persons carry *H. pylori.* Among non-Hispanic whites in the United States, little colonization is

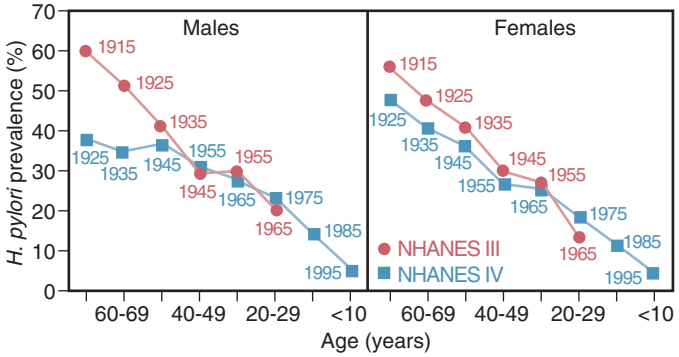

FIG. 217.1 Age-specific prevalence of *Helicobacter pylori* positivity among men and women in the National Health and Nutrition Examination Survey (NHANES) III, phase I (1988–1991; *red circles*) and NHANES IV, phase I (1999–2000; *blue squares*), by age and year of birth. Year of birth is shown at the midpoint of decade birth categories. Prevalence data for participants 19 years of age or younger were not available in NHANES III. *(From Chen Y, Blaser MJ.* Helicobacter pylori *colonization is inversely associated with childhood asthma.* J Infect Dis. *2008;198:553–560.)*

currently occurring during childhood, and the relatively high prevalence rates in adults reflect acquisition many decades ago. Among blacks and Hispanics, a high prevalence is seen at all ages.[84–86] The annual incidence of acquisition has ranged from 0.5% among epidemiologists in the United States to 7.4% among persons at an institution for individuals with intellectual disabilities in Australia.[87] The incidence of *H. pylori* has been progressively declining in the United States and other developed countries,[87–89] probably as a result of smaller family sizes, decreased crowding, improved sanitation, and more than 70 years of widespread antibiotic use.[70,90,91] Thus the age-related increase in prevalence reflects a birth cohort phenomenon (with persons born earlier having higher acquisition rates in childhood), continuing exposure and low-level new colonization into young adulthood, and continuing loss of *H. pylori* because of collateral effects from antibiotic use. The birth cohort effect predominates. The prevalence of *H. pylori* is increased among immigrants and persons of lower socioeconomic status.[5,84,86] In total, in less than 1 century in the United States, colonization has gone from being ubiquitous to being present in 5% of children[16]; this is a change in human microecology of major proportion.

PATHOLOGY AND PATHOGENESIS

H. pylori is able to survive and multiply in the acidic gastric environment, which is hostile to the growth of most bacteria. When intraluminal acidity diminishes as a result of gastric atrophy, *H. pylori* is less able to colonize the stomach, possibly because of competing organisms. *H. pylori* characteristics that permit gastric colonization include microaerophilism for survival within the mucous gel, spiral shape, flagella for motility within this viscous layer, and urease activity, which generates ammonium ions that buffer gastric acidity.[92,93] Although most organisms appear to be free-living in the mucous layer, smaller numbers appear to be adherent to the mucosal epithelial cells. Organisms may be found at the luminal surface of the gastric mucosa and also deeper within gastric glands.[94] *H. pylori* localizes almost exclusively in association with gastric-type epithelium. Affected gastric epithelium may be in the gastric antrum or fundus[95] or may be ectopic in the duodenum or in the esophagus.[4,96–98] In contrast, *H. pylori* does not colonize intestinal epithelium, even when present in the stomach.[4] Several important *H. pylori* adhesins have been identified, including the outer membrane proteins BabA (which binds to fucosylated Lewis b receptor on gastric epithelial cells), SabA (which binds to sialyl Lewis X receptors), HopQ (which binds to carcinoembryonic antigen-related cell adhesion molecules), AlpA, and AlpB.[99–104]

The gastric tissue colonized by *H. pylori* almost always exhibits an inflammatory infiltrate.[5] The lamina propria most commonly contains mononuclear cells, including lymphocytes, monocytes, and plasma cells. Neutrophils and, to a lesser extent, eosinophils may be present in the lamina propria and epithelium. The epithelial glands have a more complex architecture and less mucus when *H. pylori* is present than when it is absent.[4] In children, a follicular lymphoid pattern is common. The presence of *H. pylori* induces these changes, and the bacterium is not just a secondary colonizer.

Persons colonized with *H. pylori* have different gastric secretory physiology than do those who are not colonized. On average, colonized persons have higher gastrin levels, which are reduced by eradication of the organism.[105,106] The mechanism for increased gastrin production appears to be related to low gastric somatostatin levels,[107,108] which may reflect cytokine production in the colonized antrum.[109,110] Increased gastrin may contribute to the increase in parietal cell mass observed in many patients with duodenal ulceration. *H. pylori* products may directly affect parietal cells,[111,112] which may diminish acid production. Effects of *H. pylori* on both the fundus (the site of acid production) and the antrum (which produces factors regulating acid production) may in part be responsible for the multiplicity of outcomes of colonization.[97,109,113]

H. pylori does not appear to invade tissues, except as an incidental finding. Thus *H. pylori*–induced alterations in host tissue are likely a response to extracellular bacterial products or to cellular alterations induced by contact with the organism. Many of the alterations in gastric epithelial cells caused by *H. pylori* are attributable to CagA, which is secreted and translocated into cells through a type IV secretion system.[59,114–118] The 3′ region of *cagA* contains DNA repeats flanking sites that encode tyrosine phosphorylation motifs.[114] *H. pylori* populations include individual cells with zero, one, two, or more tyrosine phosphorylation motifs in CagA, which arise through intragenomic recombination.[119] Once injected into the epithelium by the type IV secretion system, Src and Abl kinases phosphorylate these tyrosine residues,[120,121] and phospho-CagA interacts with multiple intracellular regulatory molecules, including SHP-2[122,123] and PAR1b/MARK2,[124,125] that affect mitogen-activated protein kinases and the actin cytoskeleton; these pathways affect cell shape, cell cycle events, and cytokine production.[126–128] Nonphosphorylated CagA also can interact with cellular proteins, resulting in altered signaling events.[129] Experiments in animal models indicate that CagA and the type IV secretion system contribute to gastric inflammation and, in some cases, can promote histologic changes that are linked to gastric cancer.[118,130–132] The *cag* type IV secretion system contributes to the entry of heptose-1,7-bisphosphate (an intermediary metabolite of *H. pylori* lipopolysaccharide biosynthesis) into epithelial cells, which stimulates proinflammatory signaling.[133,134] The *cag* island is metastable, and isolates in individual patients may vary in the presence of the island, specific genes or regions, or subgenic sequences, as in the case of the 3′ region of *cagA*.[119] This instability creates a population of variants that can interact with the host in myriad ways.

Besides CagA, several other secreted or released *H. pylori* proteins are relevant. The secreted pore-forming protein VacA can cause several alterations in gastric epithelial cells (including alterations in endosomal compartments and cell death), and can also target several types of immune cells (T cells, B cells, mast cells, and macrophages).[60,135–138] Certain forms of VacA (type s1) are active in cellular assays in vitro, whereas other forms of VacA (type s2) are inactive in these assays.[61,139,140] T-cell activity is downregulated by both VacA and *H. pylori* γ-glutamyl transpeptidase; this downregulation may contribute to *H. pylori* persistence, as well as protection against allergic and asthmatic conditions.[141–144] HtrA, a secreted protease, facilitates access of *H. pylori* to the basolateral surface of gastric epithelial cells.[145] Urease may be shed by *H. pylori* cells, has been observed in affected tissues, and is a chemoattractant and activator of host phagocytic cells.[146,147]

The presence of *H. pylori* overlying the gastric mucosa activates epithelial cells to produce proinflammatory cytokines[148–151] and activates mononuclear and polymorphonuclear cells to produce cytokines, superoxide, and other proinflammatory molecules.[147,152,153] Although *H. pylori* colonization of the stomach is consistently accompanied by an inflammatory gastric host response, several factors serve to downregulate host responses.[38,109] Bacterial lipopolysaccharide usually has proinflammatory activities, but *H. pylori* lipopolysaccharide has remarkably little.[154] Similarly, *H. pylori* flagellin is modified so that it is poorly recognized by Toll-like receptor 5 (TLR5).[155] *H. pylori* lipopolysaccharide may express type II Lewis antigens (Le^x, Le^y, neither, or both of these antigens),[36,37,156] as well as type I antigens (Le^a, Le^b).[37] This observation

is significant because these antigens are present on gastric epithelial cells, and there is evidence that the host Lewis phenotype selects for the particular Lewis expression of the *H. pylori* population.[157,158] This may represent a form of molecular mimicry that promotes *H. pylori* persistence. T-regulatory cells in the gastric mucosa may downregulate local inflammatory responses.[159,160] Finally, *H. pylori* glucosylation of host-derived cholesterol contributes to immune evasion.[161]

Both bacterial and host factors may be determinants of outcome.[97,162] Virtually all patients with duodenal ulceration are colonized by strains possessing *cagA* (and thus the *cag* pathogenicity island).[6,163,164] Similarly, *cagA*$^+$ strains have been associated with a higher risk of stomach cancer than *cagA*$^-$ strains.[10,97,165,166] In East Asia, most *H. pylori* strains are *cagA*$^+$, which may account for the relatively high incidence of gastric cancer in this part of the world. Strains from patients with ulcers or stomach cancer more commonly contain type s1 *vacA* and more frequently express the BabA adhesin compared with strains from patients without these diseases.[60,61,167,168] Differences among colonized hosts in cell-mediated immunity and cytokine responses to *H. pylori* are other possible determinants of outcome variability.[169,170] Humans are polymorphic in the genetic loci involved in regulating proinflammatory cytokine production, and allelic variation in genetic determinants of interleukin-1β and interleukin-10 production affects risk of gastric cancer in *H. pylori*–positive persons.[168,171,172]

Findings similar to those observed in humans develop in several animal models of infection, including nonhuman primates and Mongolian gerbils.[173–180] The development of experimental *H. pylori* infections in these models and in human volunteers has allowed new avenues for exploring host-microbe interactions.[181,182]

CLINICAL CONSEQUENCES ASSOCIATED WITH *H. PYLORI* COLONIZATION

In the majority of *H. pylori*–colonized persons, the presence of this organism is not associated with any readily identifiable clinical consequences. Nevertheless, colonization is associated with certain types of upper gastrointestinal pathology, and appears to protect against other diseases (Table 217.2). From a clinical standpoint, the major consequences of *H. pylori* colonization are as follows.

Gastrointestinal Diseases

Acute Acquisition

Natural, voluntary, or accidental *H. pylori* acquisition may cause an acute upper gastrointestinal illness with nausea and upper abdominal pain.[1,79,183–185] Vomiting, burping, and fever may also be present. Symptoms last from 3 to 14 days, with most illnesses persisting less than 1 week. A diagnosis of food poisoning may be made in persons seeking medical attention. For many individuals, the acquisition of *H. pylori* is clinically silent.[184] Most data suggesting symptomatic acquisition relate to adults, but worldwide, most acquisition actually occurs in children[81]; the relative proportion of symptomatic and asymptomatic acute acquisition at any age is not known. In the weeks after acquisition, intense gastritis develops; hypochlorhydria ensues and may persist for as long as 1 year. In children, there is a transient increase in serum pepsinogen I levels.[81] One adult volunteer who ingested *H. pylori* seemed to have had an acute self-limited infection[185]; the frequency of this phenomenon is not known.

Persistent Colonization

It now is clear that, after acquisition, *H. pylori* persists for years, if not decades, in most persons (Fig. 217.2).[184,186] Not every exposure to *H. pylori* leads to persistent colonization, because of either lack of adaptation to the particular host[187] or coincident or proximate use of antibiotics. Tissue and serologic responses to colonization develop in essentially all persistently colonized persons.[5] The acute *H. pylori*–induced upper gastrointestinal symptoms resolve and do not return in most persons; most with persistent *H. pylori* colonization are asymptomatic. However, studies of patients with nonulcer dyspepsia indicate that *H. pylori* may be slightly more common in such patients than in age-matched control subjects[188] and that *H. pylori* colonization may be one of the causes of this common but poorly defined and heterogeneous group of disorders. Supporting this hypothesis are the results of some studies indicating that some patients with nonulcer dyspepsia who are colonized with *H. pylori* show better responses to antimicrobial therapy than treatment with placebo,[188] an effect not seen in patients with nonulcer dyspepsia who do not have *H. pylori* colonization.[189] However, in other studies, no difference between *H. pylori* treatment and placebo was found.[190,191] In total, *H. pylori* is probably responsible for fewer than 10% of cases of nonulcer dyspepsia.[192,193] Even if such an association exists, no markers are available to identify those patients with nonulcer dyspepsia in whom antibiotics would have a beneficial effect. Better definition of nonulcer dyspepsia and ascertainment of both *H. pylori* and host genotypes in individual patients should lead to a better understanding of whether *H. pylori* persistence is associated with symptoms in particular patients in the absence of ulceration or neoplasia.

TABLE 217.2 Association of *Helicobacter pylori* with Common Pathologic Lesions of the Upper Gastrointestinal Tract and with Nongastrointestinal Diseases

LESION	ASSOCIATION WITH *H. PYLORI*
Chronic diffuse superficial gastritis	Nearly always associated[4,5]
Type A (pernicious anemia) gastritis	Negative association[194,306]
NSAID gastropathy	Negative or no association[307]
Acute erosive gastritis (e.g., alcohol, aspirin)	No association[4]
Gastric ulceration	Commonly observed in patients who are not ingesting NSAIDs or aspirin[6–8,194]
Duodenal ulceration	Usually associated with idiopathic lesions (non–drug induced, non–Zollinger-Ellison syndrome)[6,8,98,308]
Gastric adenocarcinoma	Positively associated with (noncardia) cancers of the gastric body and antrum[10–13,97,212]
Gastric lymphoma	Strongly associated with MALT-type B-cell lymphomas[231,232]
Idiopathic thrombocytopenic purpura	Often associated[261–263]
Nonulcer dyspepsia	Little or no association[188–192]
Gastroesophageal reflux disease	Presence of *cag*$^+$ strains has protective association[230,237]
Barrett's esophagus	May colonize distalmost gastric epithelium in patients with gastric colonization[4]; presence of *cag*$^+$ strains has protective association[230]
Adenocarcinoma of the esophagus	Presence of *cag*$^+$ strains has protective association[97,226,227]
Childhood asthma and related allergic disorders (allergic rhinitis, eczema, and skin sensitization)	Presence of *cag*$^+$ strains has protective association[15–18]

MALT, Mucosa-associated lymphoid tumor; *NSAID,* nonsteroidal antiinflammatory drug.

Duodenal Ulceration

More than 90% of patients with "idiopathic" duodenal ulceration (i.e., not associated with nonsteroidal antiinflammatory drug use or Zollinger-Ellison syndrome) carry *H. pylori,* a prevalence significantly higher than in age-matched control subjects.[7,194] Previous *H. pylori* colonization is associated with a three- to fourfold increased risk of development of duodenal ulceration,[7] and the risk is even higher in patients colonized with *cagA*$^+$ strains.[6]

H. pylori may colonize the duodenum but only overlies metaplastic islands of gastric-type epithelium (gastric metaplasia).[4,98,195] The occurrence of *H. pylori* colonization and gastric metaplasia is highly associated with active duodenitis, a precursor lesion to ulceration,[195] and the presence of *H. pylori* in the duodenum is associated with a markedly increased risk of duodenal ulceration.[98] One model proposes that *H. pylori*–induced

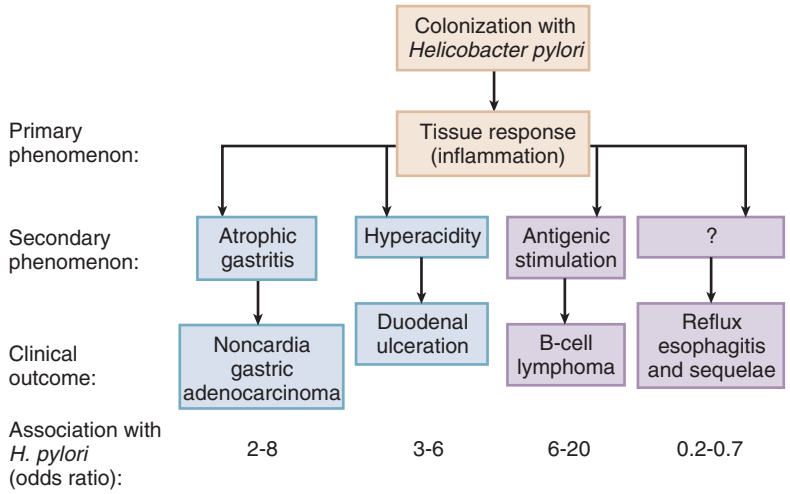

FIG. 217.2 Association of *Helicobacter pylori* colonization and disease states. After *H. pylori* acquisition, virtually all persons develop persistent colonization that lasts for life. Colonization induces tissue responses termed *chronic gastritis.* This process affects gastric physiology, including glandular structure, acid secretion, and antigen processing, which in turn affect disease risk. Colonization with *H. pylori* increases the risk of certain diseases (duodenal ulcer, gastric ulcer, noncardia gastric adenocarcinoma, B-cell lymphomas, and possibly idiopathic thrombocytopenic purpura) but decreases the risk of gastroesophageal reflux disease and its complications, including Barrett's esophagus, adenocarcinoma of the esophagus or gastric cardia, and possibly childhood asthma and related allergic disorders.

hypergastrinemia results in increased acid production, which leads to the development of gastric metaplasia in the duodenum; *H. pylori* colonization of the metaplastic gastric tissue then results in duodenitis and ulceration.

Although a significant body of evidence associating *H. pylori* colonization with idiopathic duodenal ulceration has accumulated, a causative role of *H. pylori* in ulcer disease is unproven; none of the experimental human studies has shown progression to ulceration, and why peptic ulcer disease has a remitting and relapsing course in the face of persistent colonization has never been resolved.[70] However, a large number of treatment studies using antimicrobial agents have helped define the natural history of ulcer disease. First, the use of antimicrobial agents (in the absence of acid-suppressive therapy) can heal duodenal ulcers at a rate similar to that observed with acid-suppressive therapy alone.[196,197] Second, after ulcer healing, eradication of *H. pylori* is associated with significantly lower recurrence rates than if the organism remains present.[196–198] When antimicrobial therapy that eradicates *H. pylori* is added to short-term acid-suppressive therapy, long-term ulcer relapse rates are markedly reduced, although not completely eliminated.[198,199] Altogether, these findings implicate *H. pylori* as playing a role in duodenal ulcer pathogenesis and demonstrate that antimicrobial therapy rather than long-term acid-suppressive therapy is indicated for most patients[8] because it changes the natural history of idiopathic duodenal ulcer disease. Recent studies have provided evidence that, after *H. pylori* eradication, the incidence of reflux esophagitis is doubled compared with the reflux incidence associated with failed eradication.[200,201] Thus, removal of *H. pylori* from the stomach of patients with duodenal ulceration has both benefits and costs.

Gastric Ulceration

In comparison to the high proportion of duodenal ulcer patients colonized with *H. pylori,* a smaller (50% to 80%) proportion of patients with benign gastric ulcers are colonized by *H. pylori.* The major reason is that a much higher proportion of gastric ulcers are due to nonsteroidal antiinflammatory drug or aspirin use. When such use is excluded, most of the remaining patients with benign gastric ulcers are colonized with *H. pylori,* which corresponds to a prevalence significantly higher than in age-matched control subjects.[7,194] The results of treatment of gastric ulceration with antimicrobial agents parallel the results of treatment of duodenal ulceration, changing the natural history of these diseases.[198,202]

Gastric Carcinoma

Because *H. pylori* colonization induces a tissue response (termed *chronic gastritis*) and because chronic gastritis is well known as a risk factor for the development of gastric carcinoma,[203] a role for this organism in carcinogenesis has been advanced.[97] The decreasing incidence of noncardia gastric carcinoma in developed countries is consistent with an older age of *H. pylori* acquisition[204,205] and decreasing frequency of *H. pylori* acquisition as industrialization has proceeded.[16,206] The epidemiologic characteristics of *H. pylori* colonization—including increasing prevalence at an older age; higher prevalence in blacks, Hispanics, and Asians; association with lower socioeconomic status; and early-life crowding—are all similar to the characteristics associated with noncardia gastric cancer.[65] In addition, the development of intestinal metaplasia and atrophic gastritis, two pathologic entities that are risk factors for gastric cancer, is associated with *H. pylori.*[207,208] Thus a direct role of this organism in gastric cancer is biologically plausible, and in several parts of the world there is a correlation between regions with high *H. pylori* prevalence rates and high gastric cancer rates.[209,210] Prospective studies of gastric cancer conducted in Hawaii, California, and England[211,212] and several retrospective and prospective studies each indicate that *H. pylori* is a risk factor for noncardia gastric cancer.[97] This association involves adenocarcinoma of the antrum and body of the stomach, and both the intestinal and diffuse histologic types of gastric cancer.[11–13,212] Odds ratios range from approximately 2.7 to more than 20, and the proportion of gastric cancers attributable to *H. pylori* infection ranges from about 60% to more than 90%.[12,13,210]

The presumed mechanism for adenocarcinoma involves the chronicity of *H. pylori*–induced tissue responses (inflammation), with progression to atrophic and subsequently intestinal metaplastic histology as important steps in pathogenesis. However, this process probably requires decades on average, and *H. pylori* colonization is neither necessary nor sufficient for oncogenesis. *H. pylori* alteration of signal transduction and cell-cycle kinetics in epithelial cells or gastric stem cells may predispose to neoplasia,[94,213,214] and the relationship between epithelial cell proliferation and apoptosis is probably also important.[213,215,216] *H. pylori*–induced damage to host DNA, including double-strand breaks, also may contribute to carcinogenesis.[217] Experiments in animal models suggest that CagA and the *cag* type IV secretion system (encoded by the *cag* pathogenicity island) have an important role in the pathogenesis of gastric cancer.[131,132]

There are numerous bacterial, host, and environmental factors that influence the risk of gastric cancer.[97,162] For example, the risk of gastric cancer is higher among persons colonized with *cagA*$^+$ strains than among persons colonized with *cagA*$^-$ strains[97,166] and is elevated among hosts carrying specific genetic determinants of interleukin-1β production.[168] Consumption of a high-salt diet or a diet low in fruits and vegetables has also been linked to increased gastric cancer risk. Definition of

additional host and bacterial factors that increase the risk of cancer development is an important research priority.

Gastric inflammation resolves following *H. pylori* treatment,[218] so there has been great interest in the possibility that treatment could result in regression of premalignant gastric histologic parameters or a reduced risk of gastric cancer. *H. pylori* eradication does not influence the presence of intestinal metaplasia, but several studies reported that atrophic gastritis is reversible, especially if intestinal metaplasia is not present.[219,220] Similarly, there is evidence that eradication can prevent the development of gastric atrophy.[201] Several trials reported that treatment regimens designed to eradicate *H. pylori* can reduce the risk of gastric cancer, especially in East Asian countries.[201,212,221–225]

No positive association has been observed between the presence of *H. pylori* and cancers of the gastric cardia, gastroesophageal junction, or esophagus,[11–13,211] and increasing data indicate that *H. pylori* carriage may have a protective effect against development of these types of cancer.[226,227] In parallel with the decline in *H. pylori* acquisition in developed countries, the incidence of adenocarcinomas of the gastric antrum and body has decreased over time in these countries.[228] In contrast, the incidence of several types of adenocarcinoma of the gastric cardia, gastroesophageal junction, and lower portion of the esophagus has risen dramatically.[228,229] The observed temporal relationships suggest that *H. pylori* loss may play a role in the increase of these cancers.[226,227,230] One recently recognized type of gastric cancer that occurs in the gastric corpus, primarily in women less than 50 years of age,[229] has been given the name "CYF" cancer, for gastric *c*orpus predominant, *y*oung age (<50 years old) dominant, and *f*emale dominant (rate of increase double compared with men). This type of cancer has been increasing in incidence in both Sweden and the United States, with the rise apparently beginning with the cohort born in the early 1950s. One hypothesis to explain the increasing incidence of this disease is a two-hit model, involving the loss of *H. pylori* as well as antibiotic alteration of the microbiome. Further work is needed to understand the pathogenesis of these cancers.

Gastric Lymphoma

Most gastric lymphomas arise from B lymphocytes and are termed *mucosa-associated lymphoid tumors* (MALTs or MALTomas]). *H. pylori* colonization is strongly associated with these tumors,[231,232] and eradication of *H. pylori* often leads to improvement in tumor histology.[233,234] Whether *H. pylori* eradication leads to regression of true malignancies, which are rare,[235] in contrast to the more common and more benign monoclonal lymphoid proliferation associated with *H. pylori,* is unknown. The pathogenesis of these disorders may involve chronic antigenic stimulation by *H. pylori* and subsequent induction of a polyclonal lymphoid response, a single clone of which proliferates and then undergoes neoplastic transformation.

Esophageal Diseases

Much evidence has accumulated that the incidence of *H. pylori* colonization, especially *cagA*$^+$ strains,[88] has progressively decreased in developed countries during the 20th century.[206] During this time, the occurrence of three related diseases—gastroesophageal reflux disease, Barrett's esophagus, and adenocarcinoma of the esophagus—has been increasing dramatically.[228,236] It is generally believed that Barrett's esophagus will develop in a proportion of patients with gastroesophageal reflux disease and that esophageal adenocarcinoma will develop in some of these patients. An important question is whether the loss of *H. pylori* from populations in developed countries may in some way predispose to this pathogenic sequence. Patients with gastroesophageal reflux disease are less likely to be colonized with *H. pylori* (especially *cagA*$^+$ strains) than are control subjects,[230,237] and eradication of *H. pylori* in patients with duodenal ulceration doubled the rate of gastroesophageal reflux disease development.[200] The presence of *cagA*$^+$ *H. pylori* strains is inversely associated with Barrett's esophagus and esophageal adenocarcinomas.[230,238–240] There is a clear and consistent inverse relationship between colonization with *H. pylori* (especially *cagA*$^+$ strains) and each of the disorders in the pathway (gastroesophageal reflux disease, Barrett's esophagus, and esophageal adenocarcinoma).[227] These findings suggest that *H. pylori* in the stomach is protective against the development of these esophageal disorders. The presence of decreased gastric acidity induced by long-term *H. pylori* persistence may be partially responsible.[240–243] Other potential mechanisms include *H. pylori* effects on gastric hormones, such as leptin,[244] or on the microbiota colonizing the stomach and esophagus.[245,246] In addition to the inverse relationship between *H. pylori* and the esophageal diseases described here, an inverse relationship between *H. pylori* and eosinophilic esophagitis has been reported.[247]

Asthma and Related Disorders

In recent years, asthma and related allergic disorders, including allergic rhinitis, eczema (atopic dermatitis), and skin allergies, have become markedly more prevalent, just as *H. pylori* colonization has become less common; today, less than 6% of US children are colonized with *H. pylori,*[16] a progressive decrease from the presumed near-universal presence of *H. pylori* a century ago. The results of epidemiologic studies suggest that there are specific inverse relationships between *H. pylori* and these disorders.[17] Several points have emerged about the inverse relationships. First, these associations are observed primarily with childhood-onset asthma.[15,16] Second, the effects for allergic rhinitis, eczema, and skin sensitization parallel those for asthma.[15–18] Third, the relationships have been observed not only in the United States but also in other developed countries.[18] Fourth, the strongest inverse associations are with the *cagA*$^+$ strains.[15]

These findings are consistent with the hypothesis that *H. pylori* protects against asthma and related disorders, or alternatively, *H. pylori* may be a marker for other changes in microecology that actually are the protective factors. Recent experiments have shown that *H. pylori* infection of mouse models can lead to protection against allergic asthma,[144,248] which supports the former possibility. The *H. pylori*–positive stomach includes a rich compartment of T-regulatory cells that is much better developed than in *H. pylori*–negative persons,[159,160] which could influence systemic immunity. The work in this area is in the early stages, but if *H. pylori* contributes to the protection against childhood asthma, this would influence clinical decisions related to *H. pylori* eradication.

Metabolic Disorders

In recent years, it has become clear that the stomach produces leptin, a hormone related to satiety and energy homeostasis,[249,250] and, to a greater extent, ghrelin, which has opposing effects.[251] A growing body of studies indicates that *H. pylori* status affects the levels of these hormones,[252–254] and, in particular, *H. pylori* eradication leads to an increase in ghrelin and diminution of the physiologic feeding-related decrease in serum ghrelin.[251] The data indicate that *H. pylori* colonization, by its effects on epithelial and immune cells, is involved in the regulation of these hormones.[252] It is now also clear that a generation of children in developed countries is growing up without *H. pylori* in their stomachs.[255] The full consequences of this change are unknown, but at the least, *H. pylori*–positive persons are taller.[256] Large cross-sectional studies of healthy adults have found no relationships of *H. pylori* with body mass index,[257,258] but several focused studies have observed such relationships.[259] In developed countries, there appears to be an inverse relationship between a country's *H. pylori* prevalence and its obesity prevalence.[260] This will be an important area to consider in future years.

Idiopathic Thrombocytopenic Purpura

Over the past 2 decades, there have been an increasing number of reports, chiefly from East Asia, showing an epidemiologic association between the presence of *H. pylori* and the diagnosis of idiopathic thrombocytopenic purpura (ITP).[261–265] *H. pylori* eradication has been attempted as a therapy for ITP, and although not conclusive, the results suggest that eradication can have beneficial effects in some patients.[263,264] At present, for patients diagnosed with ITP, physicians should assess *H. pylori* status, and, if positive, should consider eradicating colonization of *H. pylori* as one therapeutic approach.[263,265]

Other Inverse Associations

Several studies have reported inverse associations between the presence of *H. pylori* and inflammatory bowel disease.[266] In addition, some studies have suggested that *H. pylori* has a protective effect against diarrheal diseases and tuberculosis.[267–270]

Overview of Clinical Consequences

In summary, the presence of *H. pylori* is associated with mixed effects on human health, some favorable and some unfavorable. Although the full range of *H. pylori* effects on human health and disease is not yet completely understood, a recent study concluded that *H. pylori* status was not related to overall all-cause mortality in persons older than 40 years.[271]

DIAGNOSIS

Ascertainment of *H. pylori* colonization can be made either invasively by endoscopy and biopsy, or noninvasively by serologic analysis, breath test, or fecal antigen analysis. Properly done, each of these methodologies has a diagnostic accuracy exceeding 95%; each has advantages and disadvantages (Table 217.3).

Endoscopy with biopsy involves the most expense and is an invasive procedure, but it can yield a great deal of information, including assessment of structural lesions, such as ulcers, masses, and strictures. Biopsy specimens may be cultured for *H. pylori* on antibiotic-containing media (to diminish overgrowth by any competing flora), such as Skirrow medium, as well as with a nonselective medium, such as chocolate agar.[272] Use of two media increases the yield. Plates should be incubated for 2 to 5 days at 35° to 37°C in a moist microaerobic atmosphere (5% oxygen). Comma- or S-shaped motile organisms with catalase, oxidase, and urease activity are identified as *H. pylori*.[272] Culture enables a determination of antimicrobial susceptibilities, which may be increasingly important as antimicrobial resistance broadens. Alternatively, the organisms may be visualized on histologic sections prepared with Gram, silver, Giemsa, or acridine orange stain or by immunofluorescence or immunoperoxidase methods.[272] DNA probe and polymerase chain reaction methodologies have been developed as well, but they have no current clinical justification unless genotyping of strains or antimicrobial susceptibility testing becomes clinically important. For rapid detection of *H. pylori,* biopsy samples may be incubated at 37°C to examine for preformed urease activity. After incubation for 1 hour, the assay has a sensitivity of approximately 60%, and by 24 hours, more than 90%; bacterial overgrowth of the stomach may reduce the test's specificity, especially for longer incubations and in older patients.

High-titer, stable serum immunoglobulin G responses develop nearly universally in *H. pylori*–colonized persons.[273,274] Because serology essentially samples the entire stomach, whereas biopsy samples only a small region and the inflammatory process may be patchy, serologic analysis may be more sensitive than diagnostic methods involving biopsies.[274] With successful antimicrobial therapy, antibody levels decrease, although 3 to 6 months may be required for a noticeable effect; after ineffective therapy, high antibody levels persist.[275,276] Soon after the initial acquisition of *H. pylori,* immunoglobulin M seroconversion is noted, but levels return to baseline; with recurrence after inadequate therapy, immunoglobulin M seroconversion may again be observed.[186] A number of testing services and kits are commercially available that allow physicians to detect *H. pylori* in individual patients; the serologic assays have generally been standardized for adults, so interpretation of results in children requires caution.[277] Office-based rapid serologic tests have lower sensitivity and specificity for both adults and children.

H. pylori–positive persons also shed *H. pylori* antigens in their stools. Stool antigen assay is a relatively noninvasive means to detect positivity and to monitor therapeutic responses as early as 1 month after ending treatment. Notably, the addition of some preservatives (such as formaldehyde) to stool can cause false-negative assay results.

The high urease activity of *H. pylori* has facilitated the development of urea breath tests. Subjects fast and are then given a meal containing carbon-13 (^{13}C)-urea or carbon-14 (^{14}C)-urea; over the next hour, their breath is examined for urea-derived carbon dioxide–13 ($^{13}CO_2$) or carbon dioxide–14 ($^{14}CO_2$).[278,279] Results of these assays correlate with numbers of urease-producing *H. pylori* organisms, and can be falsely negative after therapy that suppresses but does not eradicate the organism. A negative test result 1 to 3 months after cessation of therapy usually indicates eradication of the organism.

TABLE 217.3 Modalities for *Helicobacter pylori* Diagnosis

MODALITY	ADVANTAGES	DISADVANTAGES
Endoscopy with biopsy	Permits inspection of pathology; allows detection of ulcers, neoplasms	Invasive, expensive, time consuming
Culture	Permits determination of antimicrobial susceptibilities and pathogenic features of isolates	Not optimally sensitive in most laboratories. Requires several days for results.
Histology	Generally more sensitive than culture. Allows direct visualization of organism and extent and nature of tissue involvement.	Gastritis may be patchy, and biopsy may be performed on wrong area. Insensitivity to detect small numbers of organisms. Requires several days for results.
Urease detection	Rapid; most positive results seen within 2 hr	Increased sensitivity requires longer incubation. May have false-positive results with bacterial overgrowth.
Serology	Noninvasive, rapid, quantitative, inexpensive	No determination of lesions or pathology, no antimicrobial susceptibility. Not rapidly responsive to therapy.
Urea breath tests	Relatively noninvasive, relatively rapid, quantitative, rapidly responsive to therapy. Most valuable for assessing response to eradication therapy after 4–8 wk.	Involves expensive instrumentation or administration of radioisotopes. More invasive and less convenient than serology. No determination of lesions or pathology, no antimicrobial susceptibility.
Stool antigen tests	Relatively noninvasive, relatively rapid, rapidly responsive to therapy. Most valuable for assessing response to eradication therapy after 6–8 wk.	Not quantitative. Requires stool specimen, relatively expensive for developing countries. No determination of lesions or pathology, no antimicrobial susceptibility.

THERAPY

Indications for Treatment

At present, several indications have emerged for therapy directed against *H. pylori.* For patients with peptic ulceration who are colonized with *H. pylori,* antimicrobial therapies that eradicate *H. pylori* are associated with substantially lower ulcer recurrence rates than are short-course therapies directed exclusively against gastric acidity.[198,199] Thus antimicrobial therapy now is included in the primary therapy for essentially all cases of duodenal ulceration.[8] Gastric ulcers associated with *H. pylori* can be treated in the same manner as for duodenal ulceration.[202] In patients with gastric MALTomas, antimicrobial therapy directed against *H. pylori* seems to cause tumor regression in most patients.[232] Eradication therapy also should be considered for patients with ITP who are *H. pylori* positive,[263,264] for patients who have undergone resection for early-stage gastric cancer,[220] and for patients with a high risk of gastric cancer. Data concerning the efficacy of antimicrobial therapy for *H. pylori*–associated nonulcer dyspepsia are not clear-cut; only a small proportion of patients with nonulcer dyspepsia have improvement in symptoms following eradication of *H. pylori.*[188,191,192]

Treatment Regimens

Most *H. pylori* isolates are susceptible in vitro to a variety of antimicrobial agents, including bismuth salts, amoxicillin, nitrofurans, tetracyclines, and aminoglycosides.[280,281] Primary resistance to imidazoles (such as metronidazole and tinidazole) occurs in 20% to 40% of isolates[280,282] and is most common in young women who may have received these agents for gynecologic infections or in persons from developing countries treated for parasitic infections. However, primary resistance can be present in isolates from both men and women in all age groups[283] and is associated with previous exposure to a nitroimidazole, even decades earlier.[284] Resistance to clarithromycin, other macrolides, and fluoroquinolones is increasingly common,[280,285,286] and resistance to amoxicillin and tetracycline also has been reported.[281]

Several principles of chemotherapy have emerged. First, treatment with a single agent results in apparent eradication of organisms in only a minority of cases. The cumulative effects of multiple monotherapy antibiotic courses may have hastened the disappearance of *H. pylori* from the general population, but because of its unreliability for eradication, all current treatments use combination therapy.[287] Second, some agents that are effective in vitro may be ineffective in vivo, even in combination with other agents. Erythromycin is a good example of this phenomenon.[288] The ineffectiveness of many antibiotics at an acidic pH may be responsible for the lack of activity in vivo. Third, acquired resistance frequently develops after therapy with some agents but not others. To date, resistance to bismuth salts has not been reported, and resistance to amoxicillin or tetracycline develops infrequently. In contrast, acquired resistance to fluoroquinolones is so frequent that it appears to preclude their general use. Secondary resistance to imidazoles occurs in 10% to 30% of cases, even when used in combination with other agents,[289] and the development of resistance to macrolides and rifampin can also occur. Fourth, to determine true eradication of the organism and not just temporary suppression, the patient must be shown to be free of the organism at least 1 month after the cessation of therapy if biopsy, breath test, or stool antigen test is used, and at least 6 months if serologic examination is used. Better definition of these end points is currently under investigation.

Multiple antibiotic regimens can be used successfully for eradicating *H. pylori,* but none of the regimens is 100% effective (i.e., failure rates are typically >10%) and the optimal therapeutic regimen has not been defined. Suboptimal patient compliance, antibiotic resistance, and suboptimal penetration or activity of antibiotics in the gastric environment are all factors that contribute to treatment failure. Examples of commonly used treatment regimens are shown in Table 217.4.[281,290] Proton pump inhibitors, such as omeprazole and lansoprazole, are commonly included in treatment regimens (see Table 217.4).[281,291] Proton pump inhibitors are directly inhibitory to *H. pylori,*[292] inhibit urease activity,[293] and may improve antimicrobial efficacy by inducing gastric pH neutrality. Regional antibiotic resistance patterns should potentially be considered when choosing an antibiotic regimen.[286] Tailoring therapy based on antibiotic resistance testing (e.g., analyzing 23S ribosomal RNA sequences to detect mutations associated with clarithromycin resistance) may lead to improved eradication rates.[294]

In patients from whom *H. pylori* is isolated after therapy, the organisms usually are identical to the initial isolates, thus indicating that recurrence reflects relapse rather than the acquisition of a new organism. When imidazoles or macrolides are used and fail, virtually all of the recurrent organisms are resistant. In cases of treatment failure, patients should receive antimicrobial therapy with a different regimen (see Table 217.4).[281,290,295] *H. pylori* culture and susceptibility testing should be considered before implementing second-line therapies.

TABLE 217.4 Treatment Regimens for *Helicobacter pylori*

Regimens for Initial Treatment
Bismuth quadruple therapy: PPI (standard dose twice daily) + metronidazole (500 mg three or four times daily) + tetracycline (500 mg four times daily) + bismuth four times daily (dose depends on preparation) for 14 days
Non-bismuth quadruple therapy: PPI (standard dose twice daily) + amoxicillin (1 g twice daily) + metronidazole (500 mg twice times daily) + clarithromycin (500 mg twice daily) for 14 days
PPI triple therapy[a]: PPI (standard dose twice daily) + amoxicillin (1 g twice daily) + clarithromycin (500 mg twice daily) for 14 days
Regimens Appropriate for Use After Prior Treatment Failure
Bismuth quadruple therapy: PPI (standard dose twice daily) + metronidazole (500 mg three or four times daily) + tetracycline (500 mg four times daily) + bismuth four times daily (dose depends on preparation) for 14 days
Levofloxacin triple therapy: PPI (standard dose twice daily) + amoxicillin (1 g twice daily) + levofloxacin (500 mg once daily) for 14 days

[a]Restricted to areas with low clarithromycin resistance or high eradication rates.
PPI, Proton pump inhibitor.
Modified from Fallone et al.[281] *and Malfertheiner et al.*[290]

Benefits and Risks of Antibiotic Therapy

Eradication of *H. pylori* is beneficial in patients with peptic ulcer disease, gastric MALT lymphoma, and several other conditions, and can potentially reduce the risk of gastric cancer (see "Indications for Treatment" earlier). Physicians must balance the potential benefits of antibiotic treatment with possible adverse consequences, which include development of antimicrobial resistance in *H. pylori* and other commensal bacteria,[296] medication-induced upper gastrointestinal symptoms, and less common complications, such as antibiotic-associated colitis and candidiasis. Several studies have provided evidence that, after *H. pylori* eradication, the incidence of reflux esophagitis is doubled compared with the reflux incidence associated with failed eradication,[200,201] and an increased incidence of several types of cancer involving the cardia, gastroesophageal junction, esophagus, or corpus has been linked to the absence of *H. pylori.*[228,229] If *H. pylori* protects against diarrheal diseases, as some evidence suggests,[268–270] eradication of *H. pylori* among children in developing countries could increase the risk of morbidity and mortality. Finally, if the data showing an inverse association of *H. pylori* with childhood asthma and allergic conditions and possible metabolic syndromes reflect an actual protective effect, then there should be great caution about treating young children to eradicate *H. pylori.*[255] Thus removal of *H. pylori* from the stomach has both benefits and costs. As more studies become available, physicians will need to develop criteria for treatment to optimize the therapeutic-to-toxic ratio.

Prevention of *H. pylori* Acquisition

In regions of the world with a high incidence of gastric cancer, there has been interest in the possibility that immunization might prevent *H. pylori* acquisition and *H. pylori*–associated gastric cancer.[181,182] One recent phase III study in China reported that administration of an oral recombinant vaccine to children reduced the incidence of *H. pylori* acquisition.[297] Further studies are needed to assess the long-term protective effects and benefits of vaccines.

OTHER GASTRIC HELICOBACTERS

In addition to *H. pylori,* other spiral organisms may occasionally be present in the human stomach.[298–305] These organisms are spirochetal in morphology and strongly urease positive.[298–305] Non–*H. pylori* species are much less commonly observed in the gastric mucosa than *H. pylori* and occur in perhaps 1% of persons.[300] However, these 0.5- to 1.0-µm by 4- to 8-µm spirochetes are easily visualized in specimens from colonized persons. Acquisition of some organisms (e.g., *H. felis, H. salomonis,* or *H. bizzozeronii*) probably results from contact with animals.[303] Other organisms, originally called *Gastrospirillum hominis* and more recently known as *Helicobacter heilmannii* sensu lato, have not been cultivated in vitro.[298–305] Both *H. pylori* and *H. heilmannii* may be present in the same person. The role of non–*H. pylori* species in gastric disease in humans is uncertain.

Key References

The complete reference list is available online at Expert Consult.

2. Marshall BJ, Warren JR. Unidentified curved bacilli in the stomach of patients with gastritis and peptic ulceration. *Lancet.* 1984;1:1311–1313.
5. Dooley CP, Fitzgibbons PL, Cohen H, et al. Prevalence of *Helicobacter pylori* infection and histologic gastritis in asymptomatic persons. *N Engl J Med.* 1989;321:1562–1566.
7. Nomura A, Stemmerman GN, Chyou PH, et al. *Helicobacter pylori* infection and the risk for duodenal and gastric ulceration. *Ann Intern Med.* 1994;120:977–981.
8. NIH Consensus Conference. *Helicobacter pylori* in peptic ulcer disease. NIH consensus development panel on *Helicobacter pylori* in peptic ulcer disease. *JAMA.* 1994;272:65–69.
10. Nomura AMY, Lee J, Stemmerman G, et al. *Helicobacter pylori cagA* seropositivity and gastric carcinoma risk in a Japanese American population. *J Infect Dis.* 2002;186:1138–1144.
12. Nomura A, Stemmerman GN, Chyou P-H, et al. *Helicobacter pylori* infection and gastric carcinoma in a population of Japanese-Americans in Hawaii. *N Engl J Med.* 1991;325:1132–1136.
13. Parsonnet J, Friedman GD, Vandersteen DP, et al. *Helicobacter pylori* infection and the risk of gastric carcinoma. *N Engl J Med.* 1991;325:1127–1131.

15. Chen Y, Blaser MJ. Inverse associations of *Helicobacter pylori* with asthma and allergy. *Arch Intern Med*. 2007;167:821–827.
16. Chen Y, Blaser MJ. *Helicobacter pylori* colonization is inversely associated with childhood asthma. *J Infect Dis*. 2008;198:553–560.
28. Tomb J-F, White O, Kerlavage AR, et al. The complete genome sequence of the gastric pathogen *Helicobacter pylori*. *Nature*. 1997;388:539–547.
53. Falush D, Wirth T, Linz B, et al. Traces of human migration in *Helicobacter pylori* populations. *Science*. 2003;299:1582–1585.
54. Kodaman N, Pazos A, Schneider BG, et al. Human and *Helicobacter pylori* coevolution shapes the risk of gastric disease. *Proc Natl Acad Sci USA*. 2014;111:1455–1460.
60. Cover TL, Blanke SR. *Helicobacter pylori* VacA, a paradigm for toxin multifunctionality. *Nat Rev Microbiol*. 2005;3:320–332.
61. Atherton J, Cao P, Peek RM, et al. Mosaicism in vacuolating cytotoxin alleles of *Helicobacter pylori*: association of specific *vacA* types with cytotoxin production and peptic ulceration. *J Biol Chem*. 1995;270:1771–1777.
66. Linz B, Balloux F, Moodley Y, et al. An African origin for the intimate association between humans and *Helicobacter pylori*. *Nature*. 2007;445:915–918.
68. Moodley Y, Linz B, Bond RP, et al. Age of the association between *Helicobacter pylori* and man. *PLoS Pathog*. 2012;8:e1002693.
69. Maixner F, Krause-Kyora B, Turaev D, et al. The 5300-year-old *Helicobacter pylori* genome of the iceman. *Science*. 2016;351:162–165.
70. Blaser MJ. Helicobacters are indigenous to the human stomach: duodenal ulceration is due to changes in gastric microecology in the modern era. *Gut*. 1998;43:721–727.
75. Parsonnet J, Shmuely H, Haggerty T. Fecal and oral shedding of *Helicobacter pylori* from healthy infected adults. *JAMA*. 1999;282:2240–2245.
82. O'Ryan ML, Lucero Y, Rabello M, et al. Persistent and transient *Helicobacter pylori* infections in early childhood. *Clin Infect Dis*. 2015;61:211–218.
83. Hooi JKY, Lai WY, Ng WK, et al. Global prevalence of *Helicobacter pylori* infection: systematic review and meta-analysis. *Gastroenterology*. 2017;153:420–429.
114. Odenbreit S, Puls J, Sedlmaier B, et al. Translocation of *Helicobacter pylori* CagA into gastric epithelial cells by type IV secretion. *Science*. 2000;287:1497–1500.
118. Hatakeyama M. *Helicobacter pylori* CagA and gastric cancer: a paradigm for hit-and-run carcinogenesis. *Cell Host Microbe*. 2014;15:306–316.
166. Blaser MJ, Pérez-Pérez GI, Kleanthous H, et al. Infection with *Helicobacter pylori* strains possessing *cagA* is associated with an increased risk of developing adenocarcinoma of the stomach. *Cancer Res*. 1995;55:2111–2115.
168. Figueiredo C, Machado JC, Pharoah P, et al. *Helicobacter pylori* and interleukin 1 genotyping: an opportunity to identify high-risk individuals for gastric carcinoma. *J Natl Cancer Inst*. 2002;94:1680–1687.
171. El-Omar EM, Carrington M, Chow WH, et al. Interleukin-1 polymorphisms associated with increased risk of gastric cancer. *Nature*. 2000;404:398–402.
188. McColl K, Murray L, El-Omar E, et al. Symptomatic benefit from eradicating *Helicobacter pylori* infection in patients with nonulcer dyspepsia. *N Engl J Med*. 1998;339:1869–1874.
190. Talley NJ, Vakil N, Ballard ED 2nd, et al. Absence of benefit of eradicating *Helicobacter pylori* in patients with nonulcer dyspepsia. *N Engl J Med*. 1999;341:1106–1111.
191. Blum AL, Talley NJ, O'Moráin C, et al. Lack of effect of treating *Helicobacter pylori* infection in patients with nonulcer dyspepsia. *N Engl J Med*. 1998;339:1875–1881.
197. Marshall BJ, Goodwin CS, Warren JR, et al. Prospective double-blind trial of duodenal ulcer relapse after eradication of *Campylobacter pylori*. *Lancet*. 1988;2:1437–1445.
199. Hentschel E, Brandstatter G, Dragoisics B, et al. Effect of ranitidine and amoxicillin plus metronidazole on the eradication of *Helicobacter pylori* and the recurrence of duodenal ulcer. *N Engl J Med*. 1993;328:308–312.
200. Labenz J, Blum AL, Bayerdörffer E, et al. Curing *Helicobacter pylori* infection in patients with duodenal ulcer may provoke reflux esophagitis. *Gastroenterology*. 1997;112:1442–1447.
202. Sung JJ, Chung SC, Ling TK, et al. Antibacterial treatment of gastric ulcers associated with *Helicobacter pylori*. *N Engl J Med*. 1995;332:139–142.
203. Correa P. Human gastric carcinogenesis: a multistep and multifactorial process—first American Cancer Society award lecture on cancer epidemiology and prevention. *Cancer Res*. 1992;52:6735–6740.
204. Blaser MJ, Chyou PH, Nomura A. Age at establishment of *Helicobacter pylori* infection and gastric carcinoma, gastric ulcer, and duodenal ulcer risk. *Cancer Res*. 1995;55:562–565.
205. Blaser MJ, Nomura A, Lee J, et al. Early life family structure and microbially-induced cancer risk. *PLoS Med*. 2007;4:53–58.
211. Forman D, Newell DG, Fullerton F, et al. Association between infection with *Helicobacter pylori* and risk of gastric cancer: evidence from a prospective investigation. *Br Med J*. 1991;302:1302–1305.
212. Uemura N, Okamoto S, Yamamoto S, et al. *Helicobacter pylori* infection and the development of gastric cancer. *N Engl J Med*. 2001;345:784–789.
221. Wong BC, Lam SK, Wong WM, et al. *Helicobacter pylori* eradication to prevent gastric cancer in a high-risk region of China: a randomized controlled trial. *JAMA*. 2004;291:187–194.
226. Chow W-H, Blaser MJ, Blot WJ, et al. An inverse relation between *cagA*$^+$ strains of *Helicobacter pylori* infection and risk of esophageal and gastric cardia adenocarcinoma. *Cancer Res*. 1998;58:588–590.
227. Islami F, Kamangar F. *Helicobacter pylori* and esophageal cancer risk: a meta-analysis. *Cancer Prev Res (Phila)*. 2008;1:329–338.
229. Anderson WF, Rabkin CS, Turner N, et al. The changing face of noncardia gastric cancer incidence among US non-Hispanic whites. *J Natl Cancer Inst*. 2018;110:608–615.
231. Parsonnet J, Hansen S, Rodriguez L, et al. *Helicobacter pylori* infection and gastric lymphoma. *N Engl J Med*. 1994;330:1267–1271.
248. Arnold IC, Dehzad N, Reuter S, et al. *Helicobacter pylori* infection prevents allergic asthma in mouse models through the induction of regulatory T cells. *J Clin Investig*. 2011;121:3088–3093.
260. Lender N, Talley NJ, Enck P, et al. Review article: associations between *Helicobacter pylori* and obesity—an ecological study. *Aliment Pharmacol Ther*. 2014;40:24–31.
268. Rothenbacher D, Blaser MJ, Bode G, et al. An inverse relationship between gastric colonization by *Helicobacter pylori* and diarrheal illnesses in children: results of a population-based cross-sectional study. *Infect Dis*. 2000;182:1446–1449.
271. Chen Y, Segers S, Blaser MJ. Association between *Helicobacter pylori* and mortality in the NHANES III study. *Gut*. 2013;62:1262–1269.
281. Fallone CA, Chiba N, van Zanten SV, et al. The Toronto consensus for the treatment of *Helicobacter pylori* infection in adults. *Gastroenterology*. 2016;151:51–69.
290. Malfertheiner P, Megraud F, O'Morain CA, et al. Management of *Helicobacter pylori* infection—the Maastricht V/Florence consensus report. *Gut*. 2017;66:6–30.
291. McColl KE. Clinical practice: *Helicobacter pylori* infection. *N Engl J Med*. 2010;362:1597–1604.
294. Lee HJ, Kim JI, Cheung DY, et al. Eradication of *Helicobacter pylori* according to 23S ribosomal RNA point mutations associated with clarithromycin resistance. *J Infect Dis*. 2013;208:1123–1130.

References

1. Marshall B. *Helicobacter* connections. *ChemMedChem*. 2006;1:783–802.
2. Marshall BJ, Warren JR. Unidentified curved bacilli in the stomach of patients with gastritis and peptic ulceration. *Lancet*. 1984;1:1311–1313.
3. Hazell SL, Lee A, Brady L, et al. *Campylobacter pyloridis* and gastritis: association with intracellular spaces and adaptation to an environment of mucus as important factors in colonization of the gastric epithelium. *J Infect Dis*. 1986;153:658–663.
4. Tham KT, Peek RM, Atherton JC, et al. *Helicobacter pylori* genotypes, host factors, and gastric mucosal histopathology in peptic ulcer disease. *Hum Pathol*. 2001;32:264–273.
5. Dooley CP, Fitzgibbons PL, Cohen H, et al. Prevalence of *Helicobacter pylori* infection and histologic gastritis in asymptomatic persons. *N Engl J Med*. 1989;321:1562–1566.
6. Nomura AMY, Pérez-Pérez GI, Lee J, et al. Relationship between *H. pylori cagA* status and risk of peptic ulcer disease. *Am J Epidemiol*. 2002;155:1054–1059.
7. Nomura A, Stemmerman GN, Chyou PH, et al. *Helicobacter pylori* infection and the risk for duodenal and gastric ulceration. *Ann Intern Med*. 1994;120:977–981.
8. NIH Consensus Conference. *Helicobacter pylori* in peptic ulcer disease. NIH consensus development panel on *Helicobacter pylori* in peptic ulcer disease. *JAMA*. 1994;272:65–69.
9. Blaser MJ. The changing relationships of *Helicobacter pylori* and humans: implications for health and disease. *J Infect Dis*. 1999;179:1523–1530.
10. Nomura AMY, Lee J, Stemmerman G, et al. *Helicobacter pylori cagA* seropositivity and gastric carcinoma risk in a Japanese American population. *J Infect Dis*. 2002;186:1138–1144.
11. *Helicobacter* and Cancer Collaborative Group. Gastric cancer and *Helicobacter pylori:* a combined analysis of twelve case-control studies nested within prospective cohorts. *Gut*. 2001;49:347–353.
12. Nomura A, Stemmerman GN, Chyou P-H, et al. *Helicobacter pylori* infection and gastric carcinoma in a population of Japanese-Americans in Hawaii. *N Engl J Med*. 1991;325:1132–1136.
13. Parsonnet J, Friedman GD, Vandersteen DP, et al. *Helicobacter pylori* infection and the risk of gastric carcinoma. *N Engl J Med*. 1991;325:1127–1131.
14. Cover TL, Blaser MJ. *Helicobacter pylori* in health and disease. *Gastroenterology*. 2009;136:1863–1873.
15. Chen Y, Blaser MJ. Inverse associations of *Helicobacter pylori* with asthma and allergy. *Arch Intern Med*. 2007;167:821–827.
16. Chen Y, Blaser MJ. *Helicobacter pylori* colonization is inversely associated with childhood asthma. *J Infect Dis*. 2008;198:553–560.
17. Reibman J, Marmor M, Filner J, et al. Asthma is inversely associated with *Helicobacter pylori* status in an urban population. *PLoS ONE*. 2008;3:e4060.
18. Blaser MJ, Chen Y, Reibman J. Does *Helicobacter pylori* protect against asthma and allergy? *Gut*. 2008;57:561–567.
19. Goodwin CS, Armstrong JA, Chilvers T, et al. Transfer of *Campylobacter pylori* and *Campylobacter mustelae* to *Helicobacter* gen. nov. as *Helicobacter pylori* comb. nov. and *Helicobacter mustelae* comb. nov., respectively. *Int J Syst Bacteriol*. 1989;39:397–405.
20. Paster BJ, Lee A, Fox JG, et al. Phylogeny of *Helicobacter felis* sp. nov., *Helicobacter mustelae,* and related bacteria. *Int J Syst Bacteriol*. 1991;41:31–38.
21. Eaton KA, Dewhirst FE, Radin MJ, et al. *Helicobacter acinonyx* sp. nov., isolated from cheetahs with gastritis. *Int J Syst Bacteriol*. 1993;43:99–106.
22. Kiehlbauch JA, Tauxe RV, Baker CN, et al. *Helicobacter cinaedi*-associated bacteremia and cellulitis in immunocompromised patients. *Ann Intern Med*. 1994;121:90–93.
23. Fox JG, Dewhirst FE, Tully JG, et al. *Helicobacter hepaticus* sp. nov., a microaerophilic bacterium isolated from livers and intestinal mucosal scrapings from mice. *J Clin Microbiol*. 1994;32:1238–1245.
24. Fox JG, Dewhirst FE, Shen Z, et al. Hepatic *Helicobacter* species identified in bile and gallbladder tissue from Chileans with chronic cholecystitis. *Gastroenterology*. 1998;114:755–763.
25. Matsukura N, Yokomuro S, Yamada S, et al. Association between *Helicobacter bilis* in bile and biliary tract malignancies: *H. bilis* in bile from Japanese and Thai patients with benign and malignant diseases in the biliary tract. *J Cancer Res*. 2002;93:842–847.
26. Blaser MJ. Helicobacters and biliary tract disease. *Gastroenterology*. 1998;114:840–842.
27. Dunn BE, Campbell GP, Pérez-Pérez GI, et al. Purification and characterization of *Helicobacter pylori* urease. *J Biol Chem*. 1990;265:9464–9469.
28. Tomb J-F, White O, Kerlavage AR, et al. The complete genome sequence of the gastric pathogen *Helicobacter pylori*. *Nature*. 1997;388:539–547.
29. Alm RA, Ling LL, Moir DT, et al. Genomic-sequence comparison of two unrelated isolates of the human gastric pathogen *Helicobacter pylori*. *Nature*. 1999;397:176–180.
30. Oh JD, Kling-Bäckhed H, Giannakis M, et al. The complete genome sequence of a chronic atrophic gastritis *Helicobacter pylori* strain: evolution during disease progression. *Proc Natl Acad Sci USA*. 2006;103:9999–10004.
31. Sharma CM, Hoffmann S, Darfeuille F, et al. The primary transcriptome of the major human pathogen *Helicobacter pylori*. *Nature*. 2010;464:250–255.
32. McClain MS, Shaffer CL, Israel DA, et al. Genome sequence analysis of *Helicobacter pylori* strains associated with gastric ulceration and gastric cancer. *BMC Genomics*. 2009;10:3.
33. de Reuse H, Bereswill S. Ten years after the first *Helicobacter pylori* genome: comparative and functional genomics provide new insights in the variability and adaptability of a persistent pathogen. *FEMS Immunol Med Microbiol*. 2007;50:165–176.
34. Gressmann H, Linz B, Ghai R, et al. Gain and loss of multiple genes during the evolution of *Helicobacter pylori*. *PLoS Genet*. 2005;1:e43.
35. Xu Q, Morgan RD, Roberts RJ, et al. Identification of type II restriction and modification systems in *Helicobacter pylori* reveals their substantial diversity among strains. *Proc Natl Acad Sci USA*. 2000;97:9671–9676.
36. Appelmelk BJ, Martino MC, Veenhof E, et al. Phase variation in H type I and Lewis a epitopes of *Helicobacter pylori* lipopolysaccharide. *Infect Immun*. 2000;68:5928–5932.
37. Wang G, Ge Z, Rasko DA, et al. Lewis antigens in *Helicobacter pylori*: biosynthesis and phase variation. *Mol Microbiol*. 2000;36:1187–1196.
38. Blaser MJ, Atherton JC. *Helicobacter pylori* persistence: biology and disease. *J Clin Invest*. 2004;113:321–333.
39. Webb GF, Blaser MJ. Dynamics of bacterial phenotype selection in a colonized host. *Proc Natl Acad Sci USA*. 2002;99:3135–3140.
40. Lehours P, Vale FF, Bjursell MK, et al. Genome sequencing reveals a phage in *Helicobacter pylori*. *MBio*. 2011;2:e00239–11.
41. Akopyanz N, Bukanov NO, Westblom TU, et al. DNA diversity among clinical isolates of *Helicobacter pylori* detected by PCR-based RAPD fingerprinting. *Nucleic Acids Res*. 1992;20:5137–5142.
42. Achtman M, Azuma T, Berg DE, et al. Recombination and clonal groupings within *Helicobacter pylori* from different geographical regions. *Mol Microbiol*. 1999;32:459–470.
43. Shames B, Krajden S, Fuksa M, et al. Evidence for the occurrence of the same strain of *Campylobacter pylori* in the stomach and dental plaque. *J Clin Microbiol*. 1989;27:2849–2850.
44. Kersulyte D, Chalkauskas H, Berg DE. Emergence of recombinant strains of *Helicobacter pylori* during human infection. *Mol Microbiol*. 1999;31:31–43.
45. Suerbaum S, Smith JM, Bapumia K, et al. Free recombination with *Helicobacter pylori*. *Proc Natl Acad Sci USA*. 1998;95:12619–12624.
46. Falush D, Kraft C, Taylor NS, et al. Recombination and mutation during long-term gastric colonization by *Helicobacter pylori:* estimates of clock rates, recombination size, and minimal age. *Proc Natl Acad Sci USA*. 2001;98:15056–15061.
47. Ando T, Xu Q, Torres M, et al. Restriction-modification system differences in *Helicobacter pylori* are a barrier to interstrain plasmid transfer. *Mol Microbiol*. 2000;7:1052–1065.
48. Aras RA, Takata T, Ando T, et al. Regulation of the HpyII restriction-modification system of *Helicobacter pylori* by gene deletion and horizontal reconstitution. *Mol Microbiol*. 2001;42:369–382.
49. Bjorkholm B, Sjolund M, Falk PG, et al. Mutation frequency and biological cost of antibiotic resistance in *Helicobacter pylori*. *Proc Natl Acad Sci USA*. 2001;98:14607–14612.
50. Israel DA, Salama N, Krishna U, et al. *Helicobacter pylori* genetic diversity within the gastric niche of a single human host. *Proc Natl Acad Sci USA*. 2001;98:14625–14630.
51. Aras RA, Kang J, Tschumi A, et al. Extensive repetitive DNA facilitates prokaryotic genome plasticity. *Proc Natl Acad Sci USA*. 2003;100:13579–13584.
52. Aras RA, Fischer W, Pérez-Pérez GI, et al. Plasticity of repetitive DNA sequences within a bacterial (type IV) secretion system component. *J Exp Med*. 2003;198:1349–1360.
53. Falush D, Wirth T, Linz B, et al. Traces of human migration in *Helicobacter pylori* populations. *Science*. 2003;299:1582–1585.
54. Kodaman N, Pazos A, Schneider BG, et al. Human and *Helicobacter pylori* coevolution shapes the risk of gastric disease. *Proc Natl Acad Sci USA*. 2014;111:1455–1460.
55. Censini S, Lange C, Xiang J, et al. *cag,* a pathogenicity island of *Helicobacter pylori* encodes type I-specific and disease-associated virulence factors. *Proc Natl Acad Sci USA*. 1996;93:14648–14653.
56. Akopyanz NS, Clifton SW, Kersulyte D, et al. Analyses of the *cag* pathogenicity island of *Helicobacter pylori*. *Mol Microbiol*. 1998;28:37–53.
57. Tummuru MKR, Sharma SA, Blaser MJ. *Helicobacter pylori picB,* a homologue of the *Bordetella pertussis* toxin secretion protein, is required for induction of IL-8 in gastric epithelial cells. *Mol Microbiol*. 1995;18:867–876.
58. Olbermann P, Josenhans C, Moodley Y, et al. A global overview of the genetic and functional diversity in the *Helicobacter pylori cag* pathogenicity island. *PLoS Genet*. 2010;6:e1001069.
59. Frick-Cheng AE, Pyburn TM, Voss BJ, et al. Molecular and structural analysis of the *Helicobacter pylori cag* type IV secretion system core complex. *MBio*. 2016;7:e2001–e2015.
60. Cover TL, Blanke SR. *Helicobacter pylori* VacA, a paradigm for toxin multifunctionality. *Nat Rev Microbiol*. 2005;3:320–332.
61. Atherton J, Cao P, Peek RM, et al. Mosaicism in vacuolating cytotoxin alleles of *Helicobacter pylori:* association of specific *vacA* types with cytotoxin production and peptic ulceration. *J Biol Chem*. 1995;270:1771–1777.
62. van Doorn L-J, Figueiredo C, Sanna R, et al. Expanding allelic diversity of *Helicobacter pylori vacA*. *J Clin Microbiol*. 1998;36:2597–2603.
63. Rhead JL, Letley DP, Mohammadi M, et al. A new *Helicobacter pylori* vacuolating cytotoxin determinant, the intermediate region, is associated with gastric cancer. *Gastroenterology*. 2007;133:926–936.
64. Atherton JC, Peek RM, Tham KT, et al. The clinical and pathological importance of heterogeneity in *vacA,* encoding the vacuolating cytotoxin of *Helicobacter pylori*. *Gastroenterology*. 1997;112:92–99.
65. Taylor DN, Blaser MJ. The epidemiology of *Helicobacter pylori* infections. *Epidemiol Rev*. 1991;13:42–59.
66. Linz B, Balloux F, Moodley Y, et al. An African origin for the intimate association between humans and *Helicobacter pylori*. *Nature*. 2007;445:915–918.
67. Ghose C, Pérez-Pérez GI, Dominguez-Bello MG, et al. East Asian genotypes of *Helicobacter pylori:* strains in Amerindians provide evidence for its ancient human carriage. *Proc Natl Acad Sci USA*. 2002;99:15107–15111.
68. Moodley Y, Linz B, Bond RP, et al. Age of the association between *Helicobacter pylori* and man. *PLoS Pathog*. 2012;8:e1002693.
69. Maixner F, Krause-Kyora B, Turaev D, et al. The 5300-year-old *Helicobacter pylori* genome of the iceman. *Science*. 2016;351:162–165.
70. Blaser MJ. Helicobacters are indigenous to the human stomach: duodenal ulceration is due to changes in gastric microecology in the modern era. *Gut*. 1998;43:721–727.
71. Pérez-Pérez GI, Bodhidatta L, Wongsrichanalai J, et al. Seroprevalence of *Helicobacter pylori* infections in Thailand. *J Infect Dis*. 1990;161:1237–1241.
72. Thomas JE, Gibson GR, Darboe MK, et al. Isolation of *Helicobacter pylori* from human faeces. *Lancet*. 1992;340:1194–1195.
73. Kabir S. Detection of *Helicobacter pylori* DNA in feces and saliva by polymerase chain reaction: a review. *Helicobacter*. 2004;9:115–123.
74. Polish LB, Douglas JM, Davidson AJ, et al. Characterization of risk factors for *Helicobacter pylori* infection among men attending an STD clinic: lack of evidence for sexual transmission. *J Clin Microbiol*. 1991;29:2139–2143.
75. Parsonnet J, Shmuely H, Haggerty T. Fecal and oral shedding of *Helicobacter pylori* from healthy infected adults. *JAMA*. 1999;282:2240–2245.
76. Drumm B, Pérez-Pérez GI, Blaser MJ, et al. Intrafamilial clustering of *Helicobacter pylori* infection. *N Engl J Med*. 1990;322:359–363.
77. Goodman KJ, Correa P, Tengana Aux HJ, et al. *Helicobacter pylori* infection in the Colombian Andes: a population-based study of transmission pathways. *Am J Epidemiol*. 1996;144:290–299.
78. Goodman KJ, Correa P. Transmission of *Helicobacter pylori* among siblings. *Lancet*. 2000;355:358–362.
79. Graham DY, Alpert LC, Smith JL, et al. Iatrogenic *Campylobacter pylori* infection is a cause of epidemic achlorhydria. *Am J Gastroenterol*. 1988;83:974–980.

80. Mitchell HM, Li YY, Hu PJ, et al. Epidemiology of *Helicobacter pylori* in southern China: identification of early childhood as the critical period for acquisition. *J Infect Dis*. 1992;166:149–153.
81. Pérez-Pérez GI, Sack RB, Reid R, et al. Transient and persistent *Helicobacter pylori* colonization in Native American children. *J Clin Microbiol*. 2003;41:2401–2407.
82. O'Ryan ML, Lucero Y, Rabello M, et al. Persistent and transient *Helicobacter pylori* infections in early childhood. *Clin Infect Dis*. 2015;61:211–218.
83. Hooi JKY, Lai WY, Ng WK, et al. Global prevalence of *Helicobacter pylori* infection: systematic review and meta-analysis. *Gastroenterology*. 2017;153:420–429.
84. Graham DY, Malaty HM, Evans DG, et al. Epidemiology of *Helicobacter pylori* in an asymptomatic population in the United States: effect of age, race and socioeconomic status. *Gastroenterology*. 1991;100:1495–1501.
85. Dehesa M, Dooley CP, Cohen HA, et al. High prevalence of *Helicobacter pylori* in an asymptomatic Hispanic population. *J Clin Microbiol*. 1991;29:1128–1131.
86. Everhart JE, Kruszon-Moran D, Pérez-Pérez GI, et al. Seroprevalence and ethnic differences in *Helicobacter pylori* infection among adults in the United States. *J Infect Dis*. 2000;181:1359–1363.
87. Parsonnet J. The incidence of *Helicobacter pylori* infection. *Aliment Pharmacol Ther*. 1995;9:45–51.
88. Pérez-Pérez GI, Salomaa A, Kosunen TU, et al. Evidence that *cagA*$^+$ *Helicobacter pylori* strains are disappearing more rapidly than *cagA*$^-$ strains. *Gut*. 2002;50:295–298.
89. Banatvala N, Mayo K, Megraud F, et al. The cohort effect and *Helicobacter pylori*. *J Infect Dis*. 1993;168:219–221.
90. Mendall MA, Googin PM, Molineaux N, et al. Childhood living conditions and *Helicobacter pylori* seropositivity in adult life. *Lancet*. 1992;339:896.
91. Sitas F, Forman D, Yarnell JWG, et al. *Helicobacter pylori* infection rates in relation to age and social class in a Welsh male population. *Gut*. 1991;32:25–28.
92. Scott DR, Weeks D, Hong C, et al. The role of internal urease in acid resistance of *Helicobacter pylori*. *Gastroenterology*. 1998;114:58–70.
93. McGowan CC, Necheva AS, Forsyth MH, et al. Promoter analysis of *Helicobacter pylori* genes whose expression is enhanced at low pH. *Mol Microbiol*. 2003;48:1225–1239.
94. Sigal M, Rothenberg ME, Logan CY, et al. *Helicobacter pylori* activates and expands Lgr5(+) stem cells through direct colonization of the gastric glands. *Gastroenterology*. 2015;148:1392–1404.
95. Morris A, Maher K, Thomsen L, et al. Distribution of *Campylobacter pylori* in the human stomach obtained at postmortem. *Scand J Gastroenterol*. 1988;23:257–264.
96. Price AB. Histological aspects of *Campylobacter pylori* colonization and infection of gastric and duodenal mucosa. *Scand J Gastroenterol*. 1988;23:21–24.
97. Peek RM, Blaser MJ. *Helicobacter pylori* and gastrointestinal tract adenocarcinomas. *Nat Rev Cancer*. 2002;2:28–37.
98. Carrick J, Lee A, Hazell S, et al. *Campylobacter pylori,* duodenal ulcer and gastric metaplasia: possible role of functional heterotrophic tissue in ulcerogenesis. *Gut*. 1989;30:790–797.
99. Ilver D, Arnqvist A, Ogren J, et al. *Helicobacter pylori* adhesion binding fucosylated histo-blood group antigens revealed by retagging. *Science*. 1998;279:373–377.
100. Mahdavi J, Sonden B, Hurtig M, et al. *Helicobacter pylori* SabA adhesin in persistent infection and chronic inflammation. *Science*. 2002;297:573–578.
101. Pride DT, Blaser MJ. Concerted evolution between duplicated genetic elements in *Helicobacter pylori*. *J Mol Biol*. 2002;316:627–640.
102. Odenbreit S, Till M, Hofreuter D, et al. Genetic and functional characterization of the *alpAB* gene locus essential for the adhesion of *Helicobacter pylori* to human gastric tissue. *Mol Microbiol*. 1999;31:1537–1548.
103. Königer V, Holsten L, Harrison U, et al. *Helicobacter pylori* exploits human CEACAMs via HopQ for adherence and translocation of CagA. *Nat Microbiol*. 2016;2:16188.
104. Javaheri A, Kruse T, Moonens K, et al. *Helicobacter pylori* adhesin HopQ engages in a virulence-enhancing interaction with human CEACAMs. *Nat Microbiol*. 2016;2:16189.
105. Smith JTL, Pounder RF, Nwokolo CU, et al. Inappropriate hypergastrinaemia in asymptomatic healthy subjects with *Helicobacter pylori*. *Gut*. 1990;31:522–525.
106. McColl KEL, Fullarton GM, Nujumi AM, et al. Lowered gastrin and gastric activity after eradication of *Campylobacter pylori* in duodenal ulcer. *Lancet*. 1989;2:499–500.
107. Moss SF, Legon S, Bishop AE, et al. Effect of *Helicobacter pylori* on gastric somatostatin in duodenal ulcer disease. *Lancet*. 1992;340:930–932.
108. Tham TCK, Chen L, Dennison N, et al. Effect of *Helicobacter pylori* eradication on antral somatostatin cell density in humans. *Eur J Gastroenterol Hepatol*. 1998;10:289–291.
109. Blaser MJ. Hypotheses on the pathogenesis and natural history of *Helicobacter pylori*-induced inflammation. *Gastroenterology*. 1992;102:720–727.
110. Yamamoto S, Kaneko H, Konagaya T, et al. Interactions among gastric somatostatin, interleukin-8, and mucosal inflammation in *Helicobacter pylori*-positive peptic ulcer patients. *Helicobacter*. 2001;6:136–145.
111. Cave DR, Vargas M. Effect of a *Campylobacter pylori* protein on acid secretion by parietal cells. *Lancet*. 1989;2:187–189.
112. Wang F, Xia P, Wu F, et al. *Helicobacter pylori* VacA disrupts apical membrane-cytoskeletal interactions in gastric parietal cells. *J Biol Chem*. 2008;283:26714–26725.
113. Blaser MJ. Ecology of *Helicobacter pylori* in the human stomach. *J Clin Invest*. 1997;100:759–762.
114. Odenbreit S, Puls J, Sedlmaier B, et al. Translocation of *Helicobacter pylori* CagA into gastric epithelial cells by type IV secretion. *Science*. 2000;287:1497–1500.
115. Segal ED, Cha J, Lo J, et al. Altered states: involvement of phosphorylated CagA in the induction of host cellular growth changes by *Helicobacter pylori*. *Proc Natl Acad Sci USA*. 1999;96:14559–14564.
116. Kwok T, Zabler D, Urman S, et al. *Helicobacter* exploits integrin for type IV secretion and kinase activation. *Nature*. 2007;449:862–866.
117. Shaffer CL, Gaddy JA, Loh JT, et al. *Helicobacter pylori* exploits a unique repertoire of type IV secretion system components for pilus assembly at the bacteria-host cell interface. *PLoS Pathog*. 2011;7:e1002237.
118. Hatakeyama M. *Helicobacter pylori* CagA and gastric cancer: a paradigm for hit-and-run carcinogenesis. *Cell Host Microbe*. 2014;15:306–316.
119. Aras RA, Lee Y, Kim S-K, et al. Natural variation in populations of persistently colonizing bacteria affects human host cell phenotype. *J Infect Dis*. 2003;188:486–496.
120. Tammer I, Brandt S, Hartig R, et al. Activation of abl by *Helicobacter pylori:* a novel kinase for CagA and crucial mediator of host cell scattering. *Gastroenterology*. 2007;132:1309–1319.
121. Selbach M, Moese S, Hauck CR, et al. Src is the kinase of *Helicobacter pylori* CagA protein in vitro and in vivo. *J Biol Chem*. 2002;277:6775–6778.
122. Higashi H, Tsutsumi R, Muto S, et al. SHP-2 tyrosine phosphatase as an intracellular target of *Helicobacter pylori* CagA protein. *Science*. 2002;295:683–686.
123. Yamazaki S, Yamakawa A, Yoshiuki I, et al. The CagA protein of *Helicobacter pylori* is translocated into epithelial cells and binds to SHP-2 in human gastric mucosa. *J Infect Dis*. 2003;187:334–337.
124. Saadat I, Higashi H, Obuse C, et al. *Helicobacter pylori* CagA targets PAR1/MARK kinase to disrupt epithelial cell polarity. *Nature*. 2007;447:330–333.
125. Zeaiter Z, Cohen D, Müsch A, et al. Analysis of detergent-resistant membranes of *Helicobacter pylori* infected gastric adenocarcinoma cells reveals a role for MARK2/Par1b in CagA-mediated disruption of cellular polarity. *Cell Microbiol*. 2008;10:781–794.
126. Tsutsumi R, Higashi H, Higuchi M, et al. Attenuation of *Helicobacter pylori* CagA - SHP-2 signaling by interaction between CagA and C-terminal src kinase. *J Biol Chem*. 2003;278:3664–3670.
127. Selbach M, Moese S, Hurwitz R, et al. The *Helicobacter pylori* CagA protein induces cortactin dephosphorylation and actin rearrangement by c-Src inactivation. *EMBO J*. 2003;22:515–528.
128. Amieva MR, Vogelmann R, Covacci A, et al. Disruption of the epithelial apical-junctional complex by *Helicobacter pylori* CagA. *Science*. 2003;300:1430–1434.
129. Mimuro H, Suzuki T, Tanaka J, et al. Grb2 is a key mediator of *Helicobacter pylori* CagA protein activities. *Mol Cell*. 2002;10:745–755.
130. Ohnishi N, Yuasa H, Tanaka S, et al. Transgenic expression of *Helicobacter pylori* CagA induces gastrointestinal and hematopoietic neoplasms in mouse. *Proc Natl Acad Sci USA*. 2008;105:1003–1008.
131. Ogura K, Maeda S, Nakao M, et al. Virulence factors of *Helicobacter pylori* responsible for gastric diseases in Mongolian gerbil. *J Exp Med*. 2000;192:1601–1610.
132. Rieder G, Merchant JL, Haas R. *Helicobacter pylori* cag-type IV secretion system facilitates corpus colonization to induce precancerous conditions in Mongolian gerbils. *Gastroenterology*. 2005;128:1229–1242.
133. Viala J, Chaput C, Boneca IG, et al. Nod1 responds to peptidoglycan delivered by the *Helicobacter pylori* cag pathogenicity island. *Nat Immunol*. 2004;5:1166–1174.
134. Stein SC, Faber E, Bats SH, et al. *Helicobacter pylori* modulates host cell responses by CagT4SS-dependent translocation of an intermediate metabolite of LPS inner core heptose biosynthesis. *PLoS Pathog*. 2017;13: e1006514.
135. Papini E, Satin B, Norais N, et al. Selective increase of the permeability of polarized epithelial cell monolayers by *Helicobacter pylori* vacuolating toxin. *J Clin Invest*. 1998;102:813–820.
136. Iwamoto H, Czajkowsky DM, Cover TL, et al. VacA from *Helicobacter pylori:* a hexameric chloride channel. *FEBS Lett*. 1999;450:101–104.
137. Molinari M, Salio M, Galli C, et al. Selective inhibition of Ii-dependent antigen presentation by *Helicobacter pylori* toxin VacA. *J Exp Med*. 1998;187:135–140.
138. Zheng PY, Jones NL. *Helicobacter pylori* strains expressing the vacuolating cytotoxin interrupt phagosome maturation in macrophages by recruiting and retaining TACO (coronin 1) protein. *Cell Microbiol*. 2003;5:25–40.
139. Letley DP, Rhead JL, Twells RJ, et al. Determinants of non-toxicity in the gastric pathogen *Helicobacter pylori*. *J Biol Chem*. 2003;278:26734–26741.
140. McClain MS, Cao P, Iwamoto H, et al. A 12-amino-acid segment, present in type s2 but not type s1 *Helicobacter pylori* VacA proteins, abolishes cytotoxin activity and alters membrane channel formation. *J Bacteriol*. 2001;183:6499–6508.
141. Gebert B, Fischer W, Weiss E, et al. *Helicobacter pylori* vacuolating cytotoxin inhibits T lymphocyte activation. *Science*. 2003;301:1099–1102.
142. Sundrud MS, Torres VJ, Unutmaz D, et al. Inhibition of primary human T cell proliferation by *Helicobacter pylori* vacuolating toxin (VacA) is independent of VacA effects on IL-2 secretion. *Proc Natl Acad Sci USA*. 2004;101:7727–7732.
143. Schmees C, Prinz C, Treptau T, et al. Inhibition of T-cell proliferation by *Helicobacter pylori* gamma-glutamyl peptidase. *Gastroenterology*. 2007;132:1820–1833.
144. Oertli M, Noben M, Engler DB, et al. *Helicobacter pylori* γ-glutamyl transpeptidase and vacuolating cytotoxin promote gastric persistence and immune tolerance. *Proc Natl Acad Sci USA*. 2013;110:3047–3052.
145. Tegtmeyer N, Wessler S, Necchi V, et al. *Helicobacter pylori* employs a unique basolateral type IV secretion mechanism for CagA delivery. *Cell Host Microbe*. 2017;22:552–560.
146. Mai UE, Pérez-Pérez GI, Allen JB, et al. Surface proteins from *Helicobacter pylori* exhibit chemotactic activity for human leukocytes and are present in gastric mucosa. *J Exp Med*. 1992;175:517–525.
147. Mai UEH, Pérez-Pérez GI, Wahl LM, et al. Soluble surface proteins from *Helicobacter pylori* activate monocytes/macrophages by lipopolysaccharide-independent mechanism. *J Clin Invest*. 1991;87:894–900.
148. Sharma SA, Tummuru MKR, Miller GG, et al. Interleukin-8 response of gastric epithelial cell lines to *Helicobacter pylori* stimulation in vitro. *Infect Immun*. 1995;63:1681–1687.
149. Isomoto H, Miyazaki M, Mizuta Y, et al. Expression of nuclear factor kappa B in *Helicobacter pylori*-infected gastric mucosa detected with Southwestern histochemistry. *Scand J Gastroenterol*. 2000;35:247–254.
150. Su B, Ceponis PJ, Lebel S, et al. *Helicobacter pylori* activates Toll-like receptor 4 expression in gastrointestinal epithelial cells. *Infect Immun*. 2003;71:3496–3502.
151. Backhed F, Rokbi B, Torstensson E, et al. Gastric mucosal recognition of *Helicobacter pylori* is independent of Toll-like receptor 4. *J Infect Dis*. 2003;187:829–836.
152. Foryst-Ludwig A, Naumann M. p21-activated kinase 1 activates the nuclear factor kappa B (NF-kappa B)-inducing kinase-I kappa B kinases NF-kappa B pathway and proinflammatory cytokines in *Helicobacter pylori* infection. *J Biol Chem*. 2000;275:39779–39785.
153. Crabtree JE, Shallcross T, Wyatt JI, et al. Tumour necrosis factor alpha secretion by *Helicobacter pylori* colonized gastric mucosa. *Gut*. 1991;32:1473–1477.
154. Pérez-Pérez GI, Shepherd VL, Morrow JD, et al. Activation of human THP-1 and rat bone marrow-derived macrophages by *Helicobacter pylori* lipopolysaccharide. *Infect Immun*. 1995;63:1183–1187.
155. Galkin VE, Yu X, Bielnicki J, et al. Divergence of quaternary structures among bacterial flagellar filaments. *Science*. 2008;320:382–385.
156. Aspinall GO, Monteiro MA, Pang H, et al. Lipopolysaccharide of the *Helicobacter pylori* type strain NCTC 11637 (ATCC 43504): structure of the O antigen and core oligosaccharide regions. *Biochemistry*. 1996;35:2489–2497.
157. Wirth H-P, Yang M, Peek RM, et al. Phenotypic diversity in Lewis expression of *Helicobacter pylori* isolates from the same host. *J Lab Clin Med*. 1999;133:488–500.
158. Wirth HP, Yang M, Peek RM, et al. *Helicobacter pylori* Lewis expression is related to the host Lewis phenotype. *Gastroenterology*. 1997;113:1091–1098.
159. Lundgren A, Stromberg E, Sjoling A, et al. Mucosal FOXP3-expressing CD4+ CD25 high regulatory T cells in *Helicobacter pylori*-infected patients. *Infect Immun*. 2005;73:523–531.

160. Robinson K, Kenefeck R, Pidgeon EL, et al. *Helicobacter pylori*–induced peptic ulcer disease is associated with inadequate regulatory T cell responses. *Gut*. 2008;57:1375–1385.
161. Wunder C, Churin Y, Winau F, et al. Cholesterol glucosylation promotes immune evasion by *Helicobacter pylori*. *Nat Med*. 2006;12:1030–1038.
162. Cover TL. *Helicobacter pylori* diversity and gastric cancer risk. *MBio*. 2016;7:e01869–15.
163. Blaser MJ, Crabtree JE. CagA and the outcome of *Helicobacter pylori* infection. *Am J Clin Pathol*. 1996;106:565–567.
164. Cover TL, Glupczynski Y, Lage AP, et al. Serologic detection of infection with *cagA*$^+$ *Helicobacter pylori* strains. *J Clin Microbiol*. 1995;33:1496–1500.
165. Atherton JC. The pathogenesis of *Helicobacter pylori*-induced gastroduodenal disease. *Annu Rev Pathol Mech Dis*. 2006;1:63–96.
166. Blaser MJ, Pérez-Pérez GI, Kleanthous H, et al. Infection with *Helicobacter pylori* strains possessing *cagA* is associated with an increased risk of developing adenocarcinoma of the stomach. *Cancer Res*. 1995;55:2111–2115.
167. Gerhard M, Lehn N, Neumayer N, et al. Clinical relevance of the *Helicobacter pylori* gene for blood-group antigen-binding adhesin. *Proc Natl Acad Sci USA*. 1999;96:12778–12783.
168. Figueiredo C, Machado JC, Pharoah P, et al. *Helicobacter pylori* and interleukin 1 genotyping: an opportunity to identify high-risk individuals for gastric carcinoma. *J Natl Cancer Inst*. 2002;94:1680–1687.
169. Karttunen R. Blood lymphocyte proliferation, cytokine secretion and appearance of T cells with activation surface markers in cultures with *Helicobacter pylori:* comparison of the responses of subjects with and without antibodies to *H. pylori*. *Clin Exp Immunol*. 1991;83:396–400.
170. Machado JC, Pharoah P, Sousa S, et al. Interleukin 1β and interleukin 1RN polymorphisms are associated with increased risk of gastric carcinoma. *Gastroenterology*. 2001;121:823–829.
171. El-Omar EM, Carrington M, Chow WH, et al. Interleukin-1 polymorphisms associated with increased risk of gastric cancer. *Nature*. 2000;404:398–402.
172. El-Omar EM, Rabkin CS, Gammon MD, et al. Increased risk of noncardia gastric cancer associated with proinflammatory cytokine gene polymorphisms. *Gastroenterology*. 2003;124:1193–1201.
173. Hazell SL, Eichberg JW, Lee DR, et al. Selection of the chimpanzee over the baboon as a model for *Helicobacter pylori* infection. *Gastroenterology*. 1992;103:848–854.
174. Dubois A, Berg DE, Incecik ET, et al. Transient and persistent experimental infection of non-human primates with *Helicobacter pylori:* implications for human disease. *Infect Immun*. 1996;64:2885–2891.
175. Watanabe T, Tada M, Nagai H, et al. *Helicobacter pylori* infection induces gastric cancer in mongolian gerbils. *Gastroenterology*. 1998;115:642–648.
176. Aebischer T, Bumann D, Epple HJ, et al. Correlation of T cell response and bacterial clearance in human volunteers challenged with *Helicobacter pylori* revealed by randomised controlled vaccination with Ty21a-based *Salmonella* vaccines. *Gut*. 2008;57:1065–1072.
177. Marchetti M, Arico B, Burroni D, et al. Development of a mouse model of *Helicobacter pylori* infection that mimics human disease. *Science*. 1995;267:1655–1658.
178. Wirth H-P, Beins MH, Yang M, et al. Experimental infection of Mongolian gerbils with wild-type and mutant *Helicobacter pylori* strains. *Infect Immun*. 1998;66:4856–4866.
179. Sakagami T, Dixon M, O'Rourke J, et al. Atrophic gastric changes in both *Helicobacter felis* and *Helicobacter pylori* infected mice are host dependent and separate from antral gastritis. *Gut*. 1996;39:639–648.
180. Peek RM Jr, Wirth HP, Moss SF, et al. *Helicobacter pylori* alters gastric epithelial cell cycle events and gastrin secretion in Mongolian gerbils. *Gastroenterology*. 2000;118:48–59.
181. Aebischer T, Bumann D, Epple HJ, et al. Correlation of T cell response and bacterial clearance in human volunteers challenged with *Helicobacter pylori* revealed by randomised controlled vaccination with Ty21a-based *Salmonella* vaccines. *Gut*. 2008;57:1065–1072.
182. Malfertheiner P, Selgrad M, Wex T, et al. Efficacy, immunogenicity, and safety of a parenteral vaccine against *Helicobacter pylori* in healthy volunteers challenged with a Cag-positive strain: a randomised, placebo-controlled phase 1/2 study. *Lancet Gastroenterol Hepatol*. 2018;pii: S2468-1253(18)30125-0.
183. Morris A, Nicholson G. Ingestion of *Campylobacter pyloridis* causes gastritis and raised fasting gastric pH. *Am J Gastroenterol*. 1987;82:192–199.
184. Harford WV, Barnett C, Lee E, et al. Acute gastritis with hypochlorhydria: report of 35 cases with long-term follow-up. *Gut*. 2000;47:467–472.
185. Marshall B. Attempt to fulfil Koch's postulates for pyloric Campylobacter. *Med J Australia*. 1985;142:436–439.
186. Morris AJ, Ali MR, Nicholson GI, et al. Long term follow-up of voluntary ingestion of *Helicobacter pylori*. *Ann Intern Med*. 1991;114:662–663.
187. Kang J, Blaser MJ. Bacterial populations as perfect gases: genomic integrity and diversification tensions in *Helicobacter pylori*. *Nat Rev Microbiol*. 2006;4:826–836.
188. McColl K, Murray L, El-Omar E, et al. Symptomatic benefit from eradicating *Helicobacter pylori* infection in patients with nonulcer dyspepsia. *N Engl J Med*. 1998;339:1869–1874.
189. Kang JY, Tay HH, Wee A, et al. Effect of colloidal bismuth subcitrate on symptoms and gastric histology in non-ulcer dyspepsia: a double blind placebo controlled study. *Gut*. 1990;31:476–480.
190. Talley NJ, Vakil N, Ballard ED 2nd, et al. Absence of benefit of eradicating *Helicobacter pylori* in patients with nonulcer dyspepsia. *N Engl J Med*. 1999;341:1106–1111.
191. Blum AL, Talley NJ, O'Moráin C, et al. Lack of effect of treating *Helicobacter pylori* infection in patients with nonulcer dyspepsia. *N Engl J Med*. 1998;339:1875–1881.
192. Talley NJ, Ford AC. Functional dyspepsia. *N Engl J Med*. 2015;373:1853–1863.
193. Moayyedi P, Forman D, Braunholtz D, et al. The proportion of upper gastrointestinal symptoms in the community associated with *Helicobacter pylori*, lifestyle factors, and nonsteroidal anti-inflammatory drugs. Leeds HELP Study Group. *Am J Gastroenterol*. 2000;95:1448–1455.
194. Blaser MJ, Pérez-Pérez GI, Lindenbaum J, et al. Association of infection due to *Helicobacter pylori* with specific upper gastrointestinal pathology. *Rev Infect Dis*. 1991;13(suppl):S704–S708.
195. Wyatt JI, Rathbone BJ, Dixon MF, et al. *Campylobacter pyloridis* and acid-induced gastric metaplasia in the pathogenesis of duodenitis. *J Clin Pathol*. 1987;40:841–848.
196. Coghlan JG, Gilligan D, Humphreys H, et al. *Campylobacter pylori* and recurrence of duodenal ulcers—a 12-month follow-up study. *Lancet*. 1987;2:1109–1111.
197. Marshall BJ, Goodwin CS, Warren JR, et al. Prospective double-blind trial of duodenal ulcer relapse after eradication of *Campylobacter pylori*. *Lancet*. 1988;2:1437–1445.
198. Graham DY, Lew GM, Klein PD, et al. Effect of treatment of *Helicobacter pylori* infection on the long-term recurrence of gastric or duodenal ulcer: a randomized, controlled study. *Ann Intern Med*. 1992;116:705–708.
199. Hentschel E, Brandstatter G, Dragoisics B, et al. Effect of ranitidine and amoxicillin plus metronidazole on the eradication of *Helicobacter pylori* and the recurrence of duodenal ulcer. *N Engl J Med*. 1993;328:308–312.
200. Labenz J, Blum AL, Bayerdörffer E, et al. Curing *Helicobacter pylori* infection in patients with duodenal ulcer may provoke reflux esophagitis. *Gastroenterology*. 1997;112:1442–1447.
201. Lee YC, Chen TH, Chiu HM, et al. The benefit of mass eradication of *Helicobacter pylori* infection: a community-based study of gastric cancer prevention. *Gut*. 2013;62:676–682.
202. Sung JJ, Chung SC, Ling TK, et al. Antibacterial treatment of gastric ulcers associated with *Helicobacter pylori*. *N Engl J Med*. 1995;332:139–142.
203. Correa P. Human gastric carcinogenesis: a multistep and multifactorial process—first American Cancer Society award lecture on cancer epidemiology and prevention. *Cancer Res*. 1992;52:6735–6740.
204. Blaser MJ, Chyou PH, Nomura A. Age at establishment of *Helicobacter pylori* infection and gastric carcinoma, gastric ulcer, and duodenal ulcer risk. *Cancer Res*. 1995;55:562–565.
205. Blaser MJ, Nomura A, Lee J, et al. Early life family structure and microbially-induced cancer risk. *PLoS Med*. 2007;4:53–58.
206. Kosunen TU, Aromaa A, Knekt P, et al. *Helicobacter* antibodies in 1973 and 1994 in the adult population of Vammala, Finland. *Epidemiol Infect*. 1997;119:29–34.
207. Craanen ME, Dekker W, Blok P, et al. Intestinal metaplasia and *Helicobacter pylori:* an endoscopic bioptic study of the gastric antrum. *Gut*. 1992;33:16–20.
208. Kuipers EJ, Uyterlinde AM, Pena AS, et al. Long-term sequelae to *Helicobacter pylori* gastritis. *Lancet*. 1995;345:1525–1528.
209. Forman D, Sitas F, Newell DG, et al. Geographic association of *Helicobacter pylori* antibody prevalence and gastric cancer mortality in rural China. *Int J Cancer*. 1990;46:608–611.
210. The Eurogast Study Group. An international association between *Helicobacter pylori* infection and gastric cancer. *Lancet*. 1993;341:1359–1362.
211. Forman D, Newell DG, Fullerton F, et al. Association between infection with *Helicobacter pylori* and risk of gastric cancer: evidence from a prospective investigation. *Br Med J*. 1991;302:1302–1305.
212. Uemura N, Okamoto S, Yamamoto S, et al. *Helicobacter pylori* infection and the development of gastric cancer. *N Engl J Med*. 2001;345:784–789.
213. Peek RM Jr, Moss SF, Tham KT, et al. *Helicobacter pylori* cagA+ strains and dissociation of gastric epithelial cell proliferation from apoptosis. *J Natl Cancer Inst*. 1997;89:863–868.
214. Maeda S, Yoshida H, Mitsuno Y, et al. Analysis of apoptotic and antiapoptotic signaling pathways induced by *Helicobacter pylori*. *Mol Pathol*. 2002;55:286–293.
215. Moss SF, Calam J, Agarwal B, et al. Induction of gastric epithelial apoptosis by *Helicobacter pylori*. *Gut*. 1996;38:498–501.
216. Hoshi T, Sasano H, Kato K, et al. Cell damage and proliferation in human gastric mucosa infected by *Helicobacter pylori*—a comparison before and after *H. pylori* eradication in non-atrophic gastritis. *Hum Pathol*. 1999;30:1412–1417.
217. Toller IM, Neelsen KJ, Steger M, et al. Carcinogenic bacterial pathogen *Helicobacter pylori* triggers DNA double-strand breaks and a DNA damage response in its host cells. *Proc Natl Acad Sci USA*. 2011;108:14944–14949.
218. Rauws EA, Langenberg W, Houthoff HJ, et al. *Campylobacter pyloridis*-associated chronic active antral gastritis. A prospective study of its prevalence and the effects of antibacterial and antiulcer treatment. *Gastroenterology*. 1988;94:33–40.
219. Kong YJ, Yi HG, Dai JC, et al. Histological changes of gastric mucosa after *Helicobacter pylori* eradication: a systematic review and meta-analysis. *World J Gastroenterol*. 2014;20:5903–5911.
220. Choi IJ, Kook MC, Kim YI, et al. *Helicobacter pylori* therapy for the prevention of metachronous gastric cancer. *N Engl J Med*. 2018;378:1085–1095.
221. Wong BC, Lam SK, Wong WM, et al. *Helicobacter pylori* eradication to prevent gastric cancer in a high-risk region of China: a randomized controlled trial. *JAMA*. 2004;291:187–194.
222. Fukase K, Kato M, Kikuchi S, et al. Effect of eradication of *Helicobacter pylori* on incidence of metachronous gastric carcinoma after endoscopic resection of early gastric cancer: an open-label, randomised controlled trial. *Lancet*. 2008;372:392–397.
223. Ma JL, Zhang L, Brown LM, et al. Fifteen-year effects of *Helicobacter pylori,* garlic and vitamin treatments on gastric cancer incidence and mortality. *J Natl Cancer Inst*. 2012;104:488–492.
224. Li WQ, Ma JL, Zhang L, et al. Effects of *Helicobacter pylori* treatment on gastric cancer incidence and mortality in subgroups. *J Natl Cancer Inst*. 2014;106:pii: dju116.
225. Ford AC, Forman D, Hunt RH, et al. *Helicobacter pylori* eradication therapy to prevent gastric cancer in healthy asymptomatic infected individuals: systematic review and meta-analysis of randomised controlled trials. *BMJ*. 2014;348:g3174.
226. Chow W-H, Blaser MJ, Blot WJ, et al. An inverse relation between *cagA*$^+$ strains of *Helicobacter pylori* infection and risk of esophageal and gastric cardia adenocarcinoma. *Cancer Res*. 1998;58:588–590.
227. Islami F, Kamangar F. *Helicobacter pylori* and esophageal cancer risk: a meta-analysis. *Cancer Prev Res (Phila)*. 2008;1:329–338.
228. Devesa SS, Blot WJ, Fraumeni JF Jr. Changing patterns in the incidence of esophageal and gastric carcinoma in the United States. *Cancer*. 1998;83:2049–2053.
229. Anderson WF, Rabkin CS, Turner N, et al. The changing face of noncardia gastric cancer incidence among US non-Hispanic whites. *J Natl Cancer Inst*. 2018;110:608–615.
230. Vicari JJ, Peek RM, Falk GW, et al. The seroprevalence of *cagA*-positive *Helicobacter pylori* strains in the spectrum of gastroesophageal reflux disease. *Gastroenterology*. 1998;115:50–57.
231. Parsonnet J, Hansen S, Rodriguez L, et al. *Helicobacter pylori* infection and gastric lymphoma. *N Engl J Med*. 1994;330:1267–1271.
232. Wotherspoon AC, Ortiz Hidalgo C, Falzon MR, et al. *Helicobacter pylori*-associated gastritis and primary B-cell gastric lymphoma. *Lancet*. 1991;338:1175–1176.
233. Neubauer A, Thiede C, Morgner A, et al. Cure of *Helicobacter pylori* infection and duration of remission of low-grade gastric mucosa-associated lymphoid tissue lymphoma. *J Natl Cancer Inst*. 1997;89:1350–1353.

234. Okame M, Takaya S, Sato H, et al. Complete regression of early-stage gastric diffuse large B-cell lymphoma in an HIV-1-infected patient following *Helicobacter pylori* eradication therapy. *Clin Infect Dis*. 2014;58:1490–1492.
235. Pinotti G, Zucca E, Roggero E, et al. Clinical features, treatment and outcome in a series of 93 patients with low-grade gastric MALT lymphoma. *Leuk Lymphoma*. 1997;26:527–537.
236. El-Serag HB, Sonnenberg A. Opposing time trends of peptic ulcer and reflux disease. *Gut*. 1998;43:327–333.
237. Loffeld RJ, Werdmuller BFM, Kusters JG, et al. Colonization with *cagA*-positive *H. pylori* strains inversely associated with reflux oesophagitis and Barrett's oesophagitis. *Digestion*. 2000;62:95–99.
238. Vaezi MF, Falk GW, Peek RM, et al. CagA-positive strains of *Helicobacter pylori* may protect against Barrett's esophagus. *Am J Gastroenterol*. 2000;95:2206–2211.
239. Warburton-Timms VJ, Charlett A, Valori RM, et al. The significance of cagA(+) *Helicobacter pylori* in reflux oesophagitis. *Gut*. 2001;49:341–346.
240. Roulton-Jones J, Logan R. An inverse relation between *cagA*-positive strains of *Helicobacter pylori* infection and risk of esophageal and gastric cardia adenocarcinoma. *Helicobacter*. 1999;4:281–283.
241. Yamaji Y, Mitsushima T, Ikuma H, et al. Inverse background of *Helicobacter pylori* antibody and pepsinogen in reflux oesophagitis compared with gastric cancer: analysis of 5732 Japanese subjects. *Gut*. 2001;49:335–340.
242. Quieroz DMM, Rocha GA, de Oliveira CA, et al. Role of corpus gastritis and *cagA*-positive *Helicobacter pylori* infection in reflux esophagitis. *J Clin Microbiol*. 2002;40:2849–2853.
243. Koike T, Ohara S, Sekine H, et al. *Helicobacter pylori* infection prevents erosive reflux oesophagitis by decreasing gastric acid secretion. *Gut*. 2001;49:330–334.
244. Francois F, Roper J, Goodman AJ, et al. The association of gastric leptin with oesophageal inflammation and metaplasia. *Gut*. 2008;57:16–24.
245. Bik EM, Eckburg PB, Gill SR, et al. Molecular analysis of the bacterial microbiota in the human stomach. *Proc Natl Acad Sci USA*. 2006;103:732–737.
246. Pei Z, Bini EJ, Yang L, et al. Bacterial biota in the human distal esophagus. *Proc Natl Acad Sci USA*. 2004;101:4250–4255.
247. Dellon ES, Peery AF, Shaheen NJ, et al. Inverse association of esophageal eosinophilia with *Helicobacter pylori* based on analysis of a US pathology database. *Gastroenterology*. 2011;141:1586–1592.
248. Arnold IC, Dehzad N, Reuter S, et al. *Helicobacter pylori* infection prevents allergic asthma in mouse models through the induction of regulatory T cells. *J Clin Investig*. 2011;121:3088–3093.
249. Breidert M, Miehlke S, Glasow A, et al. Leptin and its receptor in normal human gastric mucosa and in *Helicobacter pylori*-associated gastritis. *Scand J Gastroenterol*. 1999;34:954–961.
250. Sobhani I, Bado A, Vissuzaine C, et al. Leptin secretion and leptin receptor in the human stomach. *Gut*. 2000;47:178–183.
251. Nwokolo CU, Freshwater DA, O'Hare P, et al. Plasma ghrelin following cure of *Helicobacter pylori*. *Gut*. 2003;52:637–640.
252. Roper J, Francois F, Shue PL, et al. Leptin and ghrelin in relation to *Helicobacter pylori* status in adult males. *J Clin Endocrinol Metab*. 2008;93:2350–2357.
253. Azuma T, Suto H, Ito Y, et al. Gastric leptin and *Helicobacter pylori* infection. *Gut*. 2001;49:324–329.
254. Konturek JW, Konturek SJ, Kwiecien N, et al. Leptin in the control of gastric secretion and gut hormones in humans infected with *Helicobacter pylori*. *Scand J Gastroenterol*. 2001;36:1148–1154.
255. Blaser MJ. Who are we? Indigenous microbes and the ecology of human diseases. *EMBO Rep*. 2006;7: 956–960.
256. Richter T, Richter T, List S, et al. Five- to 7-year-old children with *Helicobacter pylori* infection are smaller than *Helicobacter*-negative children: a cross-sectional population-based study of 3,315 children. *J Pediatr Gastroenterol Nutr*. 2001;33:472–475.
257. Ioannou GN, Weiss NS, Kearney DJ. Is *Helicobacter pylori* sero-positivity related to body mass index in the United States? *Aliment Pharmacol Ther*. 2005;21: 765–772.
258. Cho I, Blaser MJ, Francois F, et al. *Helicobacter pylori* and overweight status in the United States: data from the Third National Health and Nutrition Examination Survey. *Am J Epidemiol*. 2005;162:579–584.
259. Wu MS, Lee WJ, Wang HH, et al. A case-control study of association of *Helicobacter pylori* infection with morbid obesity in Taiwan. *Arch Intern Med*. 2005;165:1552–1555.
260. Lender N, Talley NJ, Enck P, et al. Review article: associations between *Helicobacter pylori* and obesity—an ecological study. *Aliment Pharmacol Ther*. 2014;40:24–31.
261. Franchini M, Veneri D. *Helicobacter pylori* infection and immune thrombocytopenic purpura: an update. *Helicobacter*. 2004;9:342–346.
262. Rostami N, Keshtkar-Jahromi M, Rahnavardi M, et al. Effect of eradication of *Helicobacter pylori* on platelet recovery in patients with chronic idiopathic thrombocytopenic purpura: a controlled trial. *Am J Hematol*. 2008;83:376–381.
263. Satake M, Nishikawa J, Fukagawa Y, et al. The long-term efficacy of *Helicobacter pylori* eradication therapy in patients with idiopathic thrombocytopenic purpura. *J Gastroenterol Hepatol*. 2007;22:2233–2237.
264. Stasi R, Sarpatwari A, Segal JB, et al. Effects of eradication of *Helicobacter pylori* infection in patients with immune thrombocytopenic purpura: a systematic review. *Blood*. 2009;113:1231–1240.
265. Asahi A, Nishimoto T, Okazaki Y, et al. *Helicobacter pylori* eradication shifts monocyte fcgamma receptor balance toward inhibitory FcgammaRIIB in immune thrombocytopenic purpura patients. *J Clin Investig*. 2008;118:2939–2949.
266. Kyburz A, Muller A. *Helicobacter pylori* and extragastric diseases. *Curr Topics Micro Immunol*. 2017;400:325–347.
267. Perry S, de Jong BC, Solnick JV, et al. Infection with *Helicobacter pylori* is associated with protection against tuberculosis. *PLoS ONE*. 2010;5:e8804.
268. Rothenbacher D, Blaser MJ, Bode G, et al. An inverse relationship between gastric colonization by *Helicobacter pylori* and diarrheal illnesses in children: results of a population-based cross-sectional study. *Infect Dis*. 2000;182:1446–1449.
269. Putsep K, Branden CI, Boman HG, et al. Antibacterial peptide from *H. pylori*. *Nature*. 1999;398:671–672.
270. Mattsson A, Lonroth H, Quiding-Jarbrink M, et al. Induction of B cell responses in the stomach of *Helicobacter pylori*-infected subjects after oral cholera vaccination. *J Clin Invest*. 1998;102:51–56.
271. Chen Y, Segers S, Blaser MJ. Association between *Helicobacter pylori* and mortality in the NHANES III study. *Gut*. 2013;62:1262–1269.
272. Dunn BE, Cohen H, Blaser MJ. *Helicobacter pylori*. *Clin Microbiol Rev*. 1997;10:720–741.
273. Evans DJ Jr, Evans DG, Graham DY, et al. A sensitive and specific serologic test for detection of *Campylobacter pylori* infection. *Gastroenterology*. 1989;96:1004–1008.
274. Romero-Gallo J, Pérez-Pérez GI, Novick RP, et al. Responses to *Helicobacter pylori* whole cell and CagA antigens amongst Ladakh patients undergoing endoscopy. *Clin Diagn Lab Immunol*. 2002;9:1313–1317.
275. Kosunen TU, Seppala K, Sarna S, et al. Diagnostic value of decreased IgG, IgA, and IgM antibody titres after eradication of *Helicobacter pylori*. *Lancet*. 1992;339: 893–895.
276. Pérez-Pérez GI, Cutler AF, Blaser MJ. Value of serology as a non-invasive method to evaluate the efficacy of treatment in *Helicobacter pylori* infection. *Clin Infect Dis*. 1997;25:1038–1043.
277. Khanna B, Cutler A, Israel NR, et al. Use caution with serologic testing for *Helicobacter pylori* infection in children. *J Infect Dis*. 1998;178:460–465.
278. Graham DY, Evans DJ, Alpert LC, et al. *Campylobacter pylori* detected non-invasively by the 13C-urea breath test. *Lancet*. 1987;1:1174–1177.
279. Marshall BJ, Surveyor I. Carbon-14 urea breath test for the diagnosis of *Campylobacter pyloridis*-associated gastritis. *J Nucl Med*. 1988;29:11–16.
280. Megraud F. Resistance of *Helicobacter pylori* to antibiotics. *Aliment Pharmacol Ther*. 1997;11:43–53.
281. Fallone CA, Chiba N, van Zanten SV, et al. The Toronto consensus for the treatment of *Helicobacter pylori* infection in adults. *Gastroenterology*. 2016;151:51–69.
282. Xia HX, Daw MA, Beattie S, et al. Prevalence of metronidazole-resistant *Helicobacter pylori* in dyspeptic patients. *Ir J Med Sci*. 1993;162:91–94.
283. Rautelin H, Seppala K, Renkonen OV, et al. Role of metronidazole resistance in therapy of *Helicobacter pylori* infections. *Antimicrob Agents Chemother*. 1992;36:163–166.
284. European Study Group on Antibiotic Susceptibility of *Helicobacter pylori*. Results of a multicentre European survey in 1991 of metronidazole in *Helicobacter pylori*. *Eur J Clin Microbiol Infect Dis*. 1992;11:777–781.
285. Taconnelli E, Carrara E, Savoldi A, et al. Discovery, research, and development of new antibiotics: the WHO priority list of antibiotic-resistant bacteria and tuberculosis. *Lancet Infect Dis*. 2018;18:318–327.
286. Savoldi A, Carrara E, Graham DY, et al. Prevalence of antibiotic resistance in *Helicobacter pylori*: a systematic review and meta-analysis in World Health Organization regions. *Gastroenterology*. 2018;155:1372–1382, e17.
287. Pavicic MJ, Namavar F, Verboom T, et al. In vitro susceptibility of *Helicobacter pylori* to several antimicrobial combinations. *Antimicrob Agents Chemother*. 1993;37:1184–1186.
288. McNulty CAM, Gearty JC, Crump B, et al. *Campylobacter pyloridis* and associated gastritis: investigator blind, placebo controlled trial of bismuth salicylate and erythromycin ethylsuccinate. *Br Med J*. 1986;293: 645–649.
289. Logan RPH, Gummett PA, Misiewicz JJ, et al. One week eradication regimen for *Helicobacter pylori*. *Lancet*. 1991;338:1249–1252.
290. Malfertheiner P, Megraud F, O'Morain CA, et al. Management of *Helicobacter pylori* infection—the Maastricht V/Florence consensus report. *Gut*. 2017;66:6–30.
291. McColl KE. Clinical practice: *Helicobacter pylori* infection. *N Engl J Med*. 2010;362:1597–1604.
292. Iwahi T, Satoh H, Nakao M, et al. Lansoprazole, a novel benzimidazole proton pump inhibitor, and its related compounds have selective activity against *Helicobacter pylori*. *Antimicrob Agents Chemother*. 1991;35:490–496.
293. Nagata K, Satoh H, Iwahi T, et al. Potent inhibitory action of the gastric proton pump inhibitor lansoprazole against urease activity of *Helicobacter pylori:* unique action selective for *H. pylori* cells. *Antimicrob Agents Chemother*. 1993;37:769–774.
294. Lee HJ, Kim JI, Cheung DY, et al. Eradication of *Helicobacter pylori* according to 23S ribosomal RNA point mutations associated with clarithromycin resistance. *J Infect Dis*. 2013;208:1123–1130.
295. Vakil N, Megraud F. Eradication therapy for *Helicobacter pylori*. *Gastroenterology*. 2007;133:985–1001.
296. Sjolund M, Wreiber K, Andersson DI, et al. Long-term persistence of resistant *Enterococcus* species after antibiotics to eradicate *Helicobacter pylori*. *Ann Intern Med*. 2003;139:483–487.
297. Zeng M, Mao XH, Li JX, et al. Efficacy, safety, and immunogenicity of an oral recombinant *Helicobacter pylori* vaccine in children in China: a randomised, double-blind, placebo-controlled, phase 3 trial. *Lancet*. 2015;386:1457–1464.
298. Dent JC, McNulty CAM, Ulff JS, et al. Spiral organisms in the gastric antrum. *Lancet*. 1987;2:96.
299. McNulty CAM, Dent JC, Curry A, et al. New spiral bacterium in gastric mucosa. *J Clin Pathol*. 1989;42: 585–591.
300. Heilmann KL, Borchard F. Gastritis due to spiral shaped bacteria other than *Helicobacter pylori:* clinical, histological, and ultrastructural findings. *Gut*. 1991;32:137–140.
301. Solnick JV, O'Rourke J, Lee A, et al. An uncultured gastric spiral organism is a newly identified *Helicobacter* in humans. *J Infect Dis*. 1993;168:379–385.
302. O'Rourke JL, Solnick JV, Neilan BA, et al. Description of *'Candidatus Helicobacter heilmannii'* based on DNA sequence analysis of 16S rRNA and urease genes. *Int J Syst Evol Microbiol*. 2004;54(Pt 6):2203–2211.
303. Haesebrouck F, Pasmans F, Flahou B, et al. Gastric helicobacters in domestic animals and nonhuman primates and their significance for human health. *Clin Microbiol Rev*. 2009;22:202–223.
304. DeGroote D, van Doorn LJ, van den Bulck K, et al. Detection of non-pylori *Helicobacter* species in *"Helicobacter heilmannii"*-infected humans. *Helicobacter*. 2005;10:398–406.
305. Bento-Miranda M, Figueiredo C. *Helicobacter heilmannii* sensu lato: an overview of the infection in humans. *World J Gastroenterol*. 2014;20:17779–17787.
306. Fong T-L, Dooley CP, Dehesa M, et al. *Helicobacter pylori* infection in pernicious anemia: a prospective controlled study. *Gastroenterology*. 1991;100:328–332.
307. Inglehart LW, Edlow DW, Mills L, et al. The presence of *Campylobacter pylori* in nonsteroidal antiinflammatory drug associated gastritis. *J Rheumatol*. 1989;16:599–603.
308. Johnston BJ, Reed PI, Ali MH. *Campylobacter*-like organisms in duodenal and antral endoscopic biopsies: relationship to inflammation. *Gut*. 1986;27:1132–1137.

218 Enterobacteriaceae

George E. Nelson and Matthew H. Greene

SHORT VIEW SUMMARY

Definition
- The Enterobacteriaceae are a family of gram-negative, non–spore-forming facultative anaerobes that ferment glucose, reduce nitrate to nitrite, and produce catalase.

Epidemiology
- Enterobacteriaceae are the most common gram-negative pathogens isolated in microbiology laboratories and are capable of causing both community-acquired and nosocomial infection in virtually every organ system.
- They are frequently implicated in outbreak settings, especially in hospital settings.
- These organisms have numerous mechanisms of antimicrobial resistance, with increasing global spread.

Microbiology
- The family includes clinically important genera such as *Escherichia coli*, *Klebsiella*, and *Enterobacter*, among others.
- *Salmonella*, *Shigella*, and *Yersinia* are also members but are discussed in separate chapters.

Diagnosis
- Methods of diagnosis vary by pathogen but include standard culture technique as well as immunoassays, polymerase chain reaction assays, or DNA probes specific to a particular virulence factor.
- Multiplex gastrointestinal molecular assays—termed culture-independent diagnostic tests (CIDTs)—are now commonly used for diarrheal diagnostics.
- Epidemiologists employ pulsed-field gel electrophoresis (PFGE) and multilocus sequence typing (MLST) for highly discriminative strain typing during outbreaks.

Therapy
- Treatment varies according to pathogen and the severity of the clinical syndrome.
- Close attention must be given to antimicrobial resistance patterns owing to increasing prevalence and multiple mechanisms.

Prevention
- Hand and food hygiene decrease the risk of person-to-person and foodborne outbreaks.
- Strict infection control measures target nosocomial spread of antibiotic-resistant strains.
- There are no currently licensed vaccines.

The family Enterobacteriaceae belongs to the domain Bacteria, phylum Proteobacteria, class Gammaproteobacteria, and order Enterobacteriales (http://www.bacterio.net). The Enterobacteriaceae are gram-negative, non–spore-forming, facultative anaerobes that are typically motile by means of peritrichous flagella, ferment glucose, reduce nitrate to nitrite, and produce catalase but not oxidase. The family includes multiple genera implicated in human infection (Table 218.1).

This chapter begins by discussing prototypical shared structural and pathogenic features of the family Enterobacteriaceae—mostly extrapolated from the study of *Escherichia coli*— followed by more detailed descriptions of particular pathogens. *Salmonella, Shigella* (now considered an *E. coli* pathotype), and *Yersinia* are discussed elsewhere because of their distinct pathogenesis and clinical syndromes (see Chapters 223, 224, 229A, and 229B).[1]

GENERAL PROPERTIES

Epidemiology

The lower gastrointestinal tract is the predominant reservoir for Enterobacteriaceae, although these bacteria are also widely distributed in the environment. Furthermore, enterobacterial species frequently colonize the human genitourinary tract and oropharynx, especially in frequently hospitalized or immunosuppressed individuals.[2–5] Consequently, the Enterobacteriaceae are the most common gram-negative pathogens isolated in microbiology laboratories, capable of causing both community-acquired and nosocomial infection in virtually every organ system.

Antimicrobial resistance among the members of the Enterobacteriaceae family is a major global health threat.[6] Extended-spectrum β-lactamase (ESBL)–producing, metallo-β-lactamase (MBL)–producing, and carbapenem-resistant Enterobacteriaceae (CRE) isolates are frequently encountered in varied clinical settings throughout both the developed and developing world.[7–9] Plasmid-encoded resistance genes on transposons or integrons have allowed rapid and promiscuous dissemination of antibiotic resistance even between disparate enterobacterial genera.[10] Pulsed-field gel electrophoresis (PFGE), multilocus sequence typing (MLST), and whole-genome sequencing can facilitate rapid identification then containment of clonal outbreaks, including highly drug-resistant Enterobacteriaceae.[11–13]

General mechanisms of antibiotic resistance are discussed in Chapter 18.

Structural and Surface Antigenic Features

The Enterobacteriaceae are rod-shaped organisms, generally 1 to 3 μm in length and 0.5 μm in diameter, with a genome typically consisting of a single circular chromosome, although multiple plasmids may be present in the cytoplasm. As with any gram-negative organism, members of the Enterobacteriaceae family feature a peptidoglycan cell wall in the periplasmic space between an inner and outer membrane (Fig. 218.1).

Inner Membrane

The inner—or cystoplasmic—membrane is a phospholipid bilayer that maintains the proton motive force for energy storage and regulates the passage of molecules to and from the cytoplasm. The inner membrane may feature hundreds of proteins, including integral membrane proteins with transmembrane domains traversing the bilayer, lipoproteins inserted in the outer leaflet, and peripheral membrane proteins in either the inner or outer leaflet.[14] The inner membrane includes the proteins involved in electron transfer and oxidative phosphorylation, and the F_1F_0 adenosine triphosphatase that couples proton transport to adenosine triphosphate (ATP) synthesis. Furthermore, the inner membrane may feature efflux pumps, solute transporters, protein translocation systems, polysaccharide export systems, and histidine kinase signaling proteins.[15]

Periplasmic Space

The periplasmic space between the inner and outer membrane is an aqueous, oxidizing environment containing the peptidoglycan cell wall and a high concentration of proteins.[16] These proteins include oxidoreductases, isomerases, chaperones, proteases involved in protein

TABLE 218.1 Clinically Important Enterobacteriaceae

GENUS	SPECIES
Chronobacter	*sakazakii*
Citrobacter	*freundii* *koseri* *braakii*
Edwardsiella	*tarda*
Enterobacter	*cloacae* *aerogenes*
Escherichia	*coli* *albertii*
Hafnia	*alvei*
Klebsiella	*pneumoniae* *oxytoca* *granulomatis*
Morganella	*morganii*
Pantoea	*agglomerans*
Plesiomonas	*shigelloides*
Proteus	*mirabilis* *vulgaris*
Providencia	*stuartii* *rettgeri*
Salmonella	*enterica*
Serratia	*marcescens* *liquefaciens*
Shigella (now considered an E. coli pathotype)	*dysenteriae* *flexneri* *sonnei* *boydii*
Yersinia	*pestis* *enterocolitica* *pseudotuberculosis*

folding and degradation, lipoprotein-sorting proteins, detoxifying enzymes, and solute-binding proteins that ferry molecules across the periplasmic space. The periplasmic space also features enzymes involved in the biogenesis of peptidoglycan, lipopolysaccharide (LPS) and the capsule.

Peptidoglycan Cell Wall

The gram-negative cell wall is a thin layer of peptidoglycan—also called murein—composed of alternating *N*-acetylglucosamine and *N*-acetylmuramic acid amino sugars joined by β-1,4 linkages with a short peptide composed of L-alanine, D-glutamic acid, L-*meso*-diaminopalmelic acid, and D-alanine attached to the carboxyl group of the muramic acid.[17] Linear strands of peptidoglycan are linked by amide bonds to adjacent strands, with murein lipoprotein anchoring the layer to the inner leaflet of the outer membrane. The peptidoglycan layer of each bacterium is believed to comprise a single contiguous molecule enveloping the organism, responsible for shape and osmotic stability yet undergoing constant remodeling as the organism elongates and divides.

Outer Membrane

The outer membrane is an asymmetrical lipid bilayer featuring a mostly phospholipid inner leaflet and an outer leaflet composed primarily of LPS. The outer leaflet also features porins—membrane proteins with a conserved β-barrel fold enclosing a central aqueous channel—that regulate the passage of hydrophilic molecules.[18] The polarity of LPS in the outer leaflet prevents penetration of lipophilic molecules, including detergents, dyes, and hydrophobic antimicrobials.[19]

Because LPS is an essential component of the outer membrane of all gram-negative bacteria, some argue it is not a virulence factor per se. Regardless, LPS will be discussed in more detail later; the repeating oligosaccharide attached to the LPS core is referred to as the O antigen, which is the basis for serogroup classification. Each species within the Enterobacteriaceae has multiple O-antigen types.

Other Surface Polysaccharides and Capsules

Other potential surface polysaccharides of the Enterobacteriaceae family include enterobacterial common antigen, colonic acid, and a wide variety of polysaccharide envelopes known as capsules. Enterobacterial common antigen is remarkably conserved throughout the family, suggesting an important although poorly understood function.[20] Many of the Enterobacteriaceae—including *E. coli* strains—feature a colonic acid layer, which resembles a subset of capsule types. True capsules are linked to either LPS or α-glycerol phosphate and are the basis of the K-antigen serotyping scheme.[21] Some enterobacterial capsules can be quite luxuriant, imparting the highly mucoid colonial morphology to *Klebsiella* and *Enterobacter* species, for example.[22]

Flagella

Flagella are flexible, rotating surface appendages that propel bacteria through liquid environments. Most members of the Enterobacteriaceae are motile by way of flagella, often emanating from all sides of the organism (Fig. 218.2A). Even nonmotile Enterobacteriaceae sometime retain genes specific to flagellar expression and are then capable of motility under certain conditions.[23] Like LPS, flagellin is recognized by host innate immune system pattern recognition receptors, which can lead to neutrophil recruitment and initiation of an inflammatory response.[24–26]

Flagellar biogenesis proceeds from base to tip and is orchestrated by a complex regulatory network, responding to a variety of extracellular signals. The flagellar filament is composed of a hollow helical array of the protein flagellin whose amino and carboxyl termini is highly conserved within and across a species. In contrast, the surface-exposed middle of the flagellin molecule is highly variable as a result of both diversifying mutations and recombination after horizontal gene transfer.[27] This diversity is represented in the H-antigen typing scheme, the third component of O:K:H serotyping.

Pili

Most of the Enterobacteriaceae produce thin nanofilaments extending from the bacterial surface called pili—also known as fimbriae—that mediate autoaggregation and adhesion to host cells.[28] Furthermore, pili facilitate bacterial conjugation and are often encoded by plasmids—capable of harboring virulence and antimicrobial resistance genes—to mediate intercellular contact and genetic exchange.[29] The family Enterobacteriaceae features a variety of pilus types, which differ in morphology and function. An individual strain may produce multiple different pili even of the same type.[30–32]

Chaperone-usher type pili are common among the Enterobacteriaceae and include the ubiquitous type 1 pili (see Fig. 218.2B). Assembly begins with secretion machinery exporting subunits of the major structural protein pilin to the periplasmic space to bind with chaperones to prevent premature subunit interactions. The pilin-chaperone complexes are then delivered across the outer membrane—in a specific order starting with the tip—by a membrane channel protein known as the usher.[33,34] Most chaperone-usher type pili feature a rigid rod composed of a helical array of pilin joined end to end with a thinner, more flexible tip.[35,36] The tip often features adhesin proteins that serve as critical virulence factors.

Type IV pili are also widespread among the Enterobacteriaceae, often forming ropelike bundles expressed at the poles of the organism[37–41] (see Fig. 218.2C). The type IV pilin protein is processed to its mature form by a dedicated prepilin peptidase that also *N*-methylates the amino-terminal residue.[42] Type IV pili are retractable, which facilitates aggregation and disaggregation and a type of locomotion called "twitching motility."[43]

Virulence Factors

The Enterobacteriaceae relevant to the human host include a wide variety of organisms, ranging from commensal bacteria to highly virulent

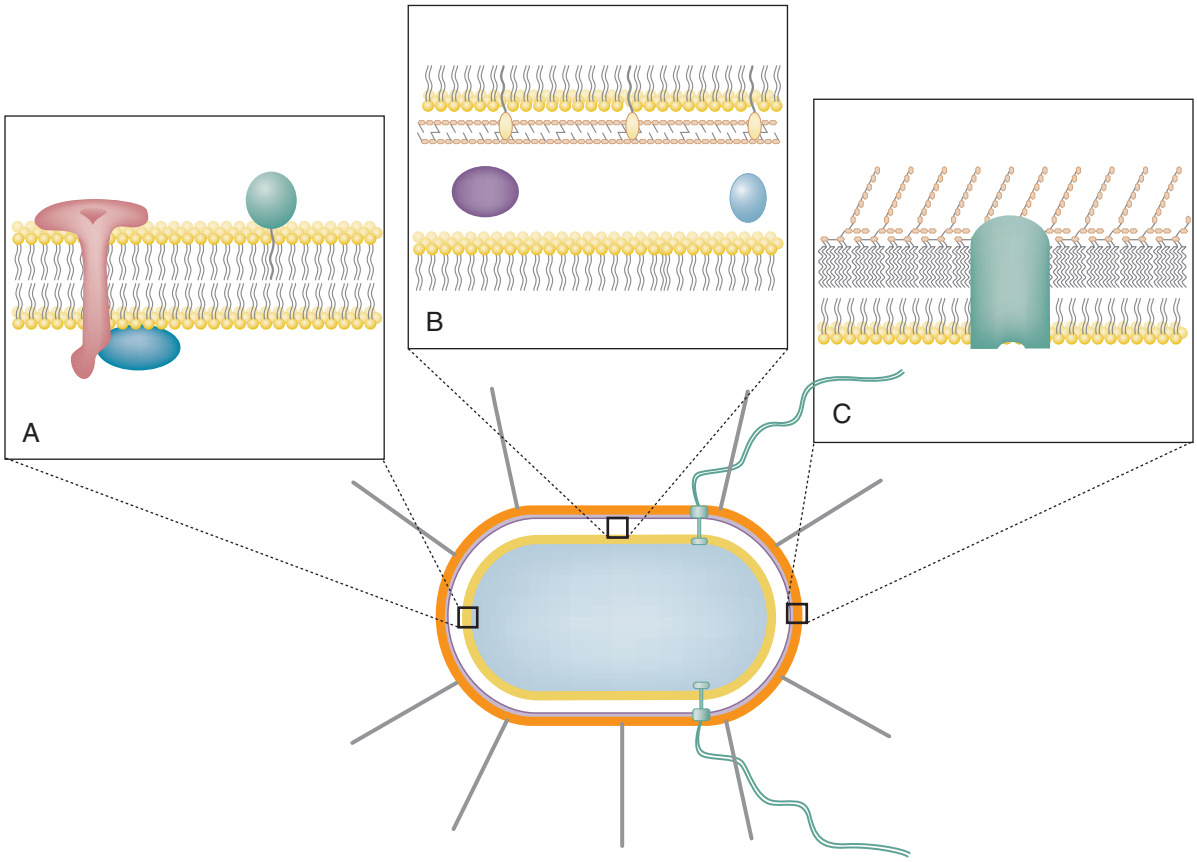

FIG. 218.1 Architecture of an enterobacterial cell. The cytoplasm is depicted in blue, the inner—or cytoplasmic—membrane in yellow, the periplasmic space in white, the peptidoglycan layer in purple, and the outer membrane in orange. Pili—also called fimbriae—are depicted in gray as linear organelles extending from the outer membrane, and flagella are shown in green as hollow, flexible organelles emanating from an assembly and secretion apparatus that spans both membranes and the periplasmic space. Insets show closer views of the envelope. (A) The cytoplasmic membrane has a phospholipid bilayer, transmembrane *(red)* proteins, peripheral membrane *(blue)* proteins, and lipoproteins *(green).* (B) The periplasmic space is surrounded by the inner and outer membranes and includes soluble proteins (*purple* and *blue*). The peptidoglycan layer, composed of disaccharide and peptide cross-linkages, is associated with the inner leaflet of the outer membrane through covalent attachment to lipoprotein *(yellow).* (C) The outer membrane layer has an inner phospholipid leaflet and an outer lipopolysaccharide leaflet. Traversing the membrane are porin proteins that regulate the passage of molecules *(green).*

pathogens. This spectrum of pathogenicity reflects the variable expression of virulence factors in a given strain. Although debate persists about what constitutes a true virulence factor, the principle of "molecular Koch's postulate" provides a helpful conceptual framework for discussing the general features of the Enterobacteriaceae discussed here. This principle defines a virulence factor as causing disease whose severity is attenuated by mutation of the gene encoding this factor, which is then restored by reintroduction of the wild-type allele.[44] As with all microbes, the common themes of pathogenicity include entry, establishment and multiplication, host defense avoidance, tissue damage, and exit.[45]

Adhesins

Many adhesins of the Enterobacteriaceae—including pili, outer membrane proteins, and even surface carbohydrates—have proven indispensable to infection.[46] An organism may feature a number of adhesins at once or in sequence in response to environmental cues or as a result of random phase variation.

Type 1 pili are the prototypical enterobacterial adhesin, composed of a rigid rod of repeating subunits of the protein FimA arranged in a helical array. The pilus tip contains a short fiber composed of FimG and FimH subunits—the so-called fimbrial adhesins—joined end-to-end with the FimA rod.[35] FimH binds to mannose residues found in glycoproteins and glycolipids on the surface of host cells.[47,48] Type 1 pili expression is subject to phase variation because of the invertible DNA element flanking the promoter of the *fim* operon, resulting in variable pili production depending on the organism's environment.[49] Furthermore, type 1 pili adhesion is enhanced by shear force such as the flow conditions of the genitourinary tract.[50] In addition to being well-established as virulence factors, type 1 pili are also important in colonization and transmission.[51,52] Furthermore, a given enterobacterial strain may produce multiple other chaperone-usher type pili and type IV pili, discussed in more detail later, depending on the particular species.

The invasin-intimin family of proteins are the best characterized outer membrane adhesins, described from studies of *Yersinia pseudotuberculosis* and enteropathogenic *E. coli* (EPEC).[53,54] These proteins—inserted into the bacterial outer membrane—share common structural features with similar amino-acid sequences, especially in their amino-terminal and central domains. The membrane portion is connected by a flexible hinge region to a rigid rod of repeating subunits similar in structure to portions of an immunoglobulin molecule. The carboxyl-terminal adhesin domain shares similarities to calcium-binding lectin molecules.[55–57]

Toxins and Secretion Systems

Many members of the Enterobacteriaceae family produce toxins released into the environment or directed to host cells. Interesting to note, despite having proven activity in animal models or tissue culture cells, many enterobacterial toxins do not fully reproduce disease in the absence of the bacteria itself, which suggests that some toxins play only an ancillary or even an unknown role in pathogenesis.

Because of the formidable barrier of the inner and outer membrane, toxin export often requires specific secretion machinery.[58] The autotransporter proteins produced by many gram-negative bacteria are a prime example.[59] Typical signal sequences at the amino termini of these proteins are cleaved as they are exported across the inner membrane

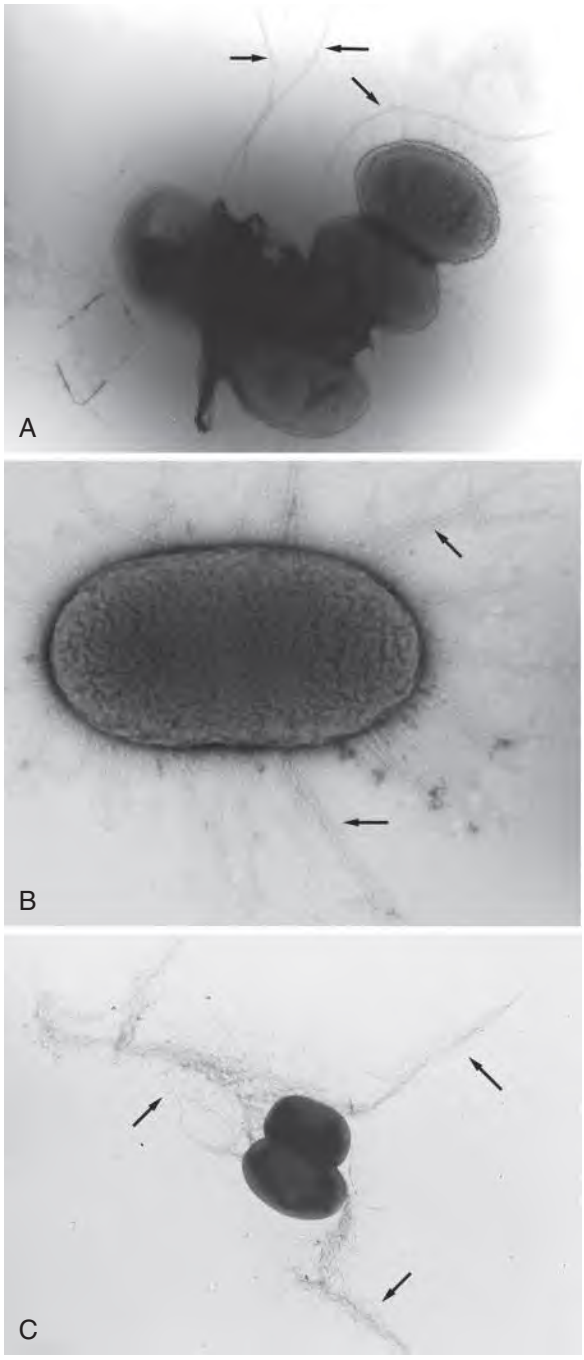

FIG. 218.2 Transmission electron micrographs of *Escherichia coli* cells. (A–B) An extraintestinal pathogenic *E. coli* strain grown under conditions favoring the expression of type 1 pili demonstrates both flagellae (*arrows* in A) and pili (*arrows* in B). (C) An enteropathogenic *E. coli* strain grown under conditions favoring expression of bundle-forming type IV pili *(arrows). (Courtesy Eric Buckles and Paula J. Fernandes.)*

then inserted into the outer membrane by the β-barrel structure of their mature amino termini. The passenger domain at the carboxyl terminus is then cleaved off and released. A subset of autotransporter proteins—termed serine protease autotransporters of the Enterobacteriaceae (SPATE)—are well characterized in *E. coli* and *Shigella* pathotypes.[60,61]

Hemolysins—named for inducing zones of clearing on blood agar plates—are toxins capable of lysing host cells. *E. coli* hemolysin is the best described and also introduces the concept of the secretion system. *E. coli* hemolysin is secreted in a single step across both the inner and outer membrane by an apparatus referred to as a type I secretion system (T1SS), composed of integral inner membrane proteins—HlyB and HlyD—that bind hemolysin. The HlyB-HlyD-hemolysin complex then engages a tunnel-channel protein, TolC, that spans the periplasmic space and outer membrane, followed by hemolysin release.[62,63] TolC serves a similar function for other toxin export, including *E. coli* heat-stable enterotoxin.[64] Hemolysin inserts into the host cell membrane with subsequent pore formation, and induces low-frequency intracellular calcium oscillations that stimulate signaling through nuclear factor kappa B (NF-κB).[65,66]

Most of the Enterobacteriaceae also feature a type II secretion system (T2SS), a multiprotein complex spanning the periplasmic compartment. T2SSs can serve a variety of functions including extracellular secretion of toxins in addition to hydrolytic enzymes.[67] For example, enterohemorrhagic *E. coli* (EHEC) O157:H7 features a plasmid-encoded T2SS that secretes a metalloprotease capable of degrading the C1 esterase inhibitor involved in complement activation.[68]

Several important enterobacterial pathogens have a type III secretion system (T3SS) that not only exports proteins through the bacterial membrane but also injects them into the host cell membrane.[69,70] The complex T3SS machinery is similar to the apparatus required for flagella assembly, featuring a "needle" extending from the bacterial outer membrane. Proteins secreted through this needle form a pore in the host cell membrane through which a variety of "effector" proteins pass, which subvert host cell pathways through structural mimicry.[71]

Lipopolysaccharide and Capsules

LPS is an essential component of the outer membrane of all gram-negative bacteria that can also function as a virulence factor. LPS has different chemical compositions depending on the organism, with varying biologic activity and potency.[72] The three major domains of LPS are the lipid A—or endotoxin—backbone, the core phosphorylated oligosaccharide, and the repeating oligosaccharide side chains.[73]

Lipid A is composed of a β-1,6-disaccharide of glucosamine that is phosphorylated and substituted with saturated hydroxylated acyl chains. The acyl groups of lipid A are inserted into the outer leaflet of the outer membrane. The core oligosaccharide is composed of a pair of 8-carbon sugars known as KDO linked to lipid A, which are in turn linked to additional sugars, forming a branched chain. The repeating oligosaccharide—or O antigen—attached to the LPS core are highly variable and, again, are the basis of Kauffman O:K:H serotyping. Genes responsible for O antigen synthesis are subject to interspecies lateral transfer, with *E. coli* alone featuring more than 170 different O-antigen serotypes.[21,74]

LPS, often bound to the host acute-phase protein LPS-binding protein (LBP), binds to CD14, where lipid A is recognized by the host pattern recognition receptor Toll-like receptor 4 (TLR4). The TLR4-MD2 complex activates NF-κB by the MyD88 pathway to initiate transcription of proinflammatory cytokines including tumor necrosis factor (TNF), chemokines, and major histocompatibility complex (MHC) receptors. This inflammatory immune response is responsible for septic shock but is also critical to host recovery. For example, mice deficient in TLR4 fail to control gram-negative infection, and TLR4 polymorphisms in humans are associated with more severe gram-negative infection.[75,76]

Enterobacterial capsules can help the organism survive in human serum by avoiding phagocytosis.[77–79] Capsules from certain strains contribute particular pathogenic features—for example, the K1 capsule, implicated in neonatal meningitis–associated *E. coli* (NMEC), or the capsule responsible for the mucoid morphology of *Klebsiella pneumoniae.*[80,81]

Iron Acquisition

Virtually all organisms require iron as a metal cofactor for critical enzymatic processes. Many enterobacterial pathogens have developed highly efficient iron scavenger systems to compete with host iron-binding proteins, controlled by the ubiquitous transcription factor ferric uptake regular (Fur).[82,83] Enterobacterial siderophores—such as enterobactin and aerobactin—are low-molecular-weight compounds with high affinity for iron that are synthesized, secreted, and recaptured by the organism.[84–86] The cytoplasmic membrane protein TonB is common to all of these iron uptake systems, spanning the periplasmic space to siderophore receptors and other gated porins in the outer membrane.[87,88]

SPECIFIC ORGANISMS

Escherichia

The genus *Escherichia* is named for Theodore Escherich who described *Escherichia coli* in 1885 during pioneering studies of the neonatal fecal microbiota.[89,90] *E. coli*, the type genus of the family Enterobacteriaceae, is the most common facultative anaerobe in the human gastrointestinal tract.[91] Although most strains are commensal organisms in the human intestine, pathogenic *E. coli* causes a variety of diseases. The genus *Escherichia* contains other species that rarely cause human infection, such as *E. blattae, E. fergusonii, E. hermanii, E. marmotae, E. vulneris,* and *E. albertii* (an *eae*-positive species often misidentified as EHEC or EPEC, with some strains previously misclassified as *Hafnia alvei* or *Shigella boydii*).[92–97]

Most *E. coli* are motile and ferment lactose and other sugars to produce indole. The average genome of *E. coli* includes approximately 5000 genes, although only about 1500 are shared among all *E. coli*, reflecting the diversity of individual factors for a given pathogenic strain.[32,91,98–100] *E. coli* acquired many of its virulence factors from horizontal gene transfer, and these are now encoded by bacteriophages, on plasmids, or within stretches of chromosome known as pathogenicity islands, which were first described in *E. coli*.[101–104]

E. coli has historically been classified by pathotypes causing diarrheal syndromes associated with characteristic virulence factors and adhesion patterns[105–107] (Table 218.2). The term *diarrheagenic E. coli* (DEC) incorporates five pathotypes discussed here—enterotoxigenic *E. coli* (ETEC); EPEC; Shiga toxin–producing *E. coli* (STEC), which includes EHEC; enteroaggregative *E. coli* (EAEC); and enteroinvasive *E. coli* (EIEC). Two additional putative enteric *E. coli* pathotypes—diffusely adherent *E. coli* (DAEC) and adherent-invasive *E. coli* (AIEC)—are also discussed.[100,108]

In outbreak settings, laboratories still use the classic Kauffman serotyping scheme to identify *E. coli* based on antigenic variation of the O (somatic) polysaccharides, H (flagellar), and K (capsular) surface antigens (e.g., *E. coli* 0157:H7). However, *E. coli* lineages elaborated through whole-genome sequencing and the discovery of hybrid strains—such as the Shiga toxin–producing EAEC O104:H4 serotype—increasingly confound classification based on pathotype, serotype, or even phylogenetic grouping.[102,109] This ambiguity reflects the evolutionary complexity of *E. coli* in the context of ongoing lateral transfer of virulence factors between strains.[110,111]

Beyond diarrheal illness, *E. coli* has the capability to cause extraintestinal disease, including infection of the peritoneum, liver, and biliary system. Furthermore, *E.* coli is the single most common cause of urinary tract infection (UTI) and a major pathogen causing meningitis, septicemia, pneumonia, cellulitis and even bone and joint infection. The term *extraintestinal pathogenic E. coli* (ExPEC)—including

TABLE 218.2 Epidemiology, Clinical Syndrome, Characteristic Virulence Factors, and Treatment Considerations for Diarrheagenic *Escherichia coli* Pathotypes

PATHOTYPE	EPIDEMIOLOGY	CLINICAL SYNDROME	CHARACTERISTIC VIRULENCE FACTORS	TREATMENT CONSIDERATIONS[a]
ETEC	Pediatric diarrhea, particularly in the developing world Traveler's diarrhea Foodborne outbreaks	Acute nonbloody diarrhea	Heat-labile (LT) and/or heat-stable toxin (ST) Heterogeneous adhesins; also called colonization factors (CFs)	Traveler's diarrhea[b]
EPEC	Pediatric diarrhea, particularly in the developing world Atypical EPEC (aEPEC) increasingly recognized to cause diarrhea regardless of age Traveler's diarrhea Nosocomial and child care center outbreaks	Acute nonbloody diarrhea Associated with vomiting and sometimes protracted disease	Attaching and effacing lesions (A/E) (see Fig. 218.3) Typical EPEC (tEPEC) defined by type IV bundle-forming pili (BFP)	Traveler's diarrhea[b] Antimicrobial therapy for severe endemic disease, including nosocomial outbreaks
STEC (including EHEC)	Foodborne outbreaks; person-to-person and animal contact transmission	Acute, often bloody diarrhea Can progress to hemolytic-uremic syndrome (HUS)	Shiga toxin EHEC produces attaching and effacing lesions (A/E) like EPEC (see Fig. 218.3)	Antimicrobial and antimotility agents associated with increased risk of HUS
EAEC	Increasingly recognized to cause diarrhea in all ages, including developed countries Associated with growth impairment in children from developing countries Traveler's diarrhea Foodborne outbreaks	Acute nonbloody diarrhea Chronic diarrhea in some children and HIV patients	Aggregative adherence fimbriae (AAF) Enteroaggregative heat-stable toxin 1 (EAST-1) and several SPATE proteins	Traveler's diarrhea[b] Case reports support antimicrobial therapy for chronic disease in HIV patients
EIEC	Pediatric diarrhea, particularly in the developing world Foodborne outbreaks; person-to-person transmission	Acute, sometimes bloody diarrhea Clinically indistinguishable from shigellosis, although typically less severe	Cellular invasion then cell-to-cell spread orchestrated by *pInv* plasmid	Antimicrobial therapy recommended for severe cases, but must first exclude STEC
DAEC	Pediatric diarrhea in somewhat older children Not an established cause of diarrhea in adults	Acute nonbloody diarrhea	Afa/Dr family adhesins	
AIEC	Associated with Crohn disease pathogenesis, although not with diarrhea per se		Adherence to M cells of Peyer patches by type I and long polar fimbriae Cell invasion facilitated by membrane-bound protein OmpA	

[a]Volume resuscitation is the most important treatment for diarrhea regardless of etiology. Most diarrheagenic *E. coli* infections are self-limiting with supportive care.
[b]Data support the use of antimicrobial therapy for shortening the duration of traveler's diarrhea with loperamide in combination with single-dose azithromycin, levofloxacin, or rifaximin.
AIEC, Adherent-invasive *E. coli*; *DAEC,* diffuse-adhering *E. coli*; *EAEC,* enteroaggregative *E. coli*; *EHEC,* enterohemorrhagic *E. coli*; *EIEC,* enteroinvasive *E. coli*; *EPEC,* enteropathogenic *E. coli*; *ETEC,* enterotoxigenic *E. coli*; *HIV,* human immunodeficiency virus; *SPATE,* serine protease autotransporters of the Enterobacteriaceae; *STEC,* Shiga toxin–producing *E. coli.*

uropathogenic *E. coli* (UPEC), NMEC, and sepsis-associated *E. coli* (SEPEC)—has been used to describe nondiarrheagenic strains.[112]

Epidemiologists have often used PFGE for highly discriminative strain typing during outbreaks. MLST has emerged as another common means of identifying pathogenic strains by their allelic profile, allowing global comparison.[113] Future typing schemes will likely incorporate the clinical syndrome, virulence factors historically associated with a given pathotype, serotyping, and whole-genome sequence data to characterize pathogenic *E. coli* most fully.

General Clinical Principles Related to Diarrheagenic *Escherichia coli*

DEC has been associated with seasonal increases in temperature.[114,115] Recent meta-analysis has suggested as much as an 8% increased incidence of diarrhea due to *E. coli* for every 1°C increase in mean monthly temperature, regardless of season.[116] Anticipated increases in the global burden of pathogenic *E. coli* underscore the need for optimal strain characterization to prevent transmission and treat infection effectively, especially in regions most affected by climate change.

E. coli strains are implicated in many diarrheal syndromes, including traveler's diarrhea, which are discussed in detail elsewhere (see Section J, "Gastrointestinal Infections and Food Poisoning," and Chapter 319). Recent guidelines emphasize the diagnostic and therapeutic strategies for suspected infectious diarrhea for both endemic disease and the returning traveler.[117–119]

The advent of multiplex gastrointestinal molecular assays—termed culture-independent diagnostic tests (CIDTs)—has helped in optimizing antibiotic therapy and identifying possible DEC outbreaks.[120,121] Guidelines suggest limiting testing to patients with diarrhea who are experiencing fever, bloody or mucoid diarrhea, severe abdominal cramping, or signs of sepsis or immunocompromised hosts.[117] Additional criteria include diarrhea lasting >7 days or epidemiologic investigation during known or suspected diarrheal outbreaks.[119]

DEC is a predominant pathogen in traveler's diarrhea.[118,122–124] Data support the use of antimicrobial therapy for shortening the duration of traveler's diarrhea with loperamide in combination with single-dose azithromycin, levofloxacin, or rifaximin.[118,125–128] However, antibiotic therapy introduces adverse side effects and increases risk for colonization with ESBL-producing Enterobacteriaceae.[128,129] Debate continues as to the severity of traveler's diarrhea warranting antibiotic treatment.[130,131]

Clinicians continue to recommend strict avoidance of contaminated food and water to prevent traveler's diarrhea, although the data are mixed as to whether this counseling reduces infection, given practical limitations of adherence.[119,132–135] Prophylactic bismuth can reduce the risk of traveler's diarrhea, but further research is needed to delineate who would clearly benefit from prophylactic antimicrobial therapy.[119,131,136,137]

Antimicrobial therapy for DEC should be directed by the identification of a clinically plausible organism.[117] Treatment specific to particular *E. coli* pathotypes will be discussed later. As a general principle, regardless of pathotype, volume resuscitation is the cornerstone of therapy for all diarrheal illness. Empirical antimicrobial therapy is discouraged in immunocompetent children and adults pending the results of appropriate diagnostic tests. Empirical therapy is recommended for infants <3 months of age with suspected bacterial diarrhea, immunocompromised patients with severe and bloody diarrhea, and ill-appearing immunocompetent patients or returning travelers with fever and/or signs of sepsis with suspected enteric fever.

Enterotoxigenic *Escherichia coli*

ETEC infection results from ingestion of contaminated water or food and is a common cause of diarrhea in the developing world, especially in children, and in returning travelers.[105,132,138] For example, ETEC producing heat-stable toxin (with or without heat-labile toxin) is one of the leading causes of pediatric diarrheal disease in sub-Saharan Africa and south Asia.[185] ETEC foodborne outbreaks occur in developed countries often as a consequence of improper sanitation.[139,140]

The ETEC incubation period ranges from hours to 2 days. The severity of diarrhea ranges from mild to cholera-like, although typically it occurs without fever, vomiting, or bloody stool. Symptoms usually last less than 5 days.[141]

ETEC pathogenesis starts with adherence, then colonization of the small intestinal mucosa, which is mediated by adhesins—or colonization factors (CFs)—on the bacterial surface.[124,142] More than 20 distinct CFs have been described, including chaperone-usher type and type IV pili, thinner fibrillae, and even afimbrial factors.[143–145] This heterogeneity of CFs may partly account for ETEC recurrence during childhood in the developing world followed by eventual immunity.[146,147] The degree to which exposure to a particular CF, toxin, or other antigen confers protection remains an important debate for vaccine development.[148]

Nonpilus adhesins, such as Tia and TibA invasins and the secreted protein EtpA, also promote bacterial adherence to intestinal cells.[149] The SPATE protein EatA has been shown to degrade EtpA, modulating adhesion and actually accelerating toxin delivery.[150]

ETEC produces heat-labile enterotoxin (LT) or heat-stable enterotoxin (ST); these toxins are responsible for secretory diarrhea. About a third of ETEC strains produce both toxins.[151] LT has an almost identical quaternary structure and mechanism of action as cholera toxin.[152] LT features a single catalytic A subunit and a pantameric B receptor-binding subunit, secreted by a T2SS.[153] The B subunit binds to the apical membrane of enterocytes and, after endocytosis, the A subunit finds its target on the basolateral membrane—the regulatory subunit of a heterotrimeric G protein ($G\alpha_s$)—and locks it in its active form. $G\alpha_s$ then constitutively activates its target, adenylyl cyclase, increasing intracellular cyclic adenosine monophosphate (cAMP) and ultimately protein kinase A, which activates the cystic fibrosis transmembrane conductance regulator (CFTR). This cascade results in active secretion of chloride then resultant sodium and water loss into the small intestinal lumen.[154,155] LT has impressive genetic diversity with varying expression in clinical syndromes, which suggests that a particular LT type confers variable virulence.[156–159]

ST is produced by 75% to 80% of ETEC isolates and is associated with more severe disease than ETEC strains that produce LT only.[124,151] ST is a small peptide resembling the mammalian peptide hormone guanylin and binds to the guanylin receptor on the enterocyte apical membrane.[160] Traditional dogma has attributed ST-induced secretory diarrhea to receptor binding resulting in elevated cyclic guanosine monophosphate (cGMP) and protein kinase G activation then CFTR activation with resultant chloride, sodium, and water secretion.[161] However, in vivo experiments with ST in rat models have questioned this doctrine.[162,163] As with LT, new ST variants continue to be reported with varying impact on severity.[164]

The diagnosis of ETEC diarrhea can be confirmed with immunoassays, polymerase chain reaction (PCR) assays, or DNA probe assays designed to detect genes encoding ETEC toxins. Furthermore, multiplex PCR assays have been developed for diarrhea suspected to be due to *E. coli,* which includes ETEC toxin or virulence factor detection.[165–168] Less expensive approaches in development include single-chain variable fragments (scFv) against LT and ST.[169]

Antimicrobial therapy for ETEC diarrhea has been studied mostly with respect to traveler's diarrhea, in which ETEC is implicated in 20% to 40% of cases.[118,122–124] Evidence for antibiotic efficacy in traveler's diarrhea was discussed earlier (see also Chapter 319), although clinicians should consider regional differences in ETEC resistance patterns.[170]

ETEC vaccine efforts have been thwarted by antigen heterogeneity and the poor immunogenicity of ST. Killed oral vaccine that produced an immune response to ETEC CFs reduced the rate of severe diarrhea in returning travelers but proved ineffective otherwise.[171,172] A cutaneous LT vaccine successfully induced LT-specific immunoglobulin, but clinical results proved disappointing.[173,174] Research is ongoing for an oral multivalent vaccine composed of four ETEC strains overexpressing CFs and an LT toxoid[175] in addition to various vaccine formulations exploiting EtpA immunogenicity to prevent host colonization.[150,176–179]

Enteropathogenic *Escherichia coli*

EPEC was the first pathotype of DEC described, reported after devastating neonatal outbreaks in the 1940s.[114,180,181] EPEC is characterized as either typical (tEPEC) or atypical (aEPEC); aEPEC lacks the type IV bundle-forming pili (BFP) encoded by the EPEC adherence factor (EAF) plasmid.[182,183]

The EPEC incubation period is rapid, with diarrhea starting as early as 3 hours after ingestion in human volunteers and with protracted disease lasting as long as 2 weeks.[105,184] tEPEC causes substantial childhood diarrhea in the developing world and is the pathogen associated with the highest risk of death in infants aged 0 to 11 months.[185] aEPEC is a highly heterogeneous group found commonly in asymptomatic children, although increasingly it is recognized as an emerging cause of diarrhea in the developed world, affecting children, adults, and the immunocompromised.[100,183,186–195] Because transmission occurs principally through person-to-person contact, EPEC is a significant contributor to nosocomial and child care center outbreaks.[196–198]

EPEC infection produces watery nonbloody diarrhea, mostly affecting children younger than 6 months.[154,190] EPEC diarrhea is often associated with vomiting, resulting in severe dehydration and with protracted disease capable of yielding malnutrition and considerable morbidity.[199–203]

The ropelike BFP (see Fig. 218.2C) of tEPEC forms three-dimensional microcolonies on tissue culture cells in a pattern termed localized adherence (LA) (see Fig. 218.4A later).[106,107,204] BFP are involved in biofilm formation but are then able to retract after host cell binding, enhancing infectivity through more efficient colonization and effector translocation.[205–207] EPEC classification has been complicated somewhat by the discovery of strains—including both tEPEC and aEPEC—displaying diffuse or aggregative adherence patterns.[190,208] tEPEC strains with a hybrid localized and aggregative-like adherence phenotype may cause more severe and persistent diarrhea owing to augmented biofilm formation.[209]

The hallmark feature of EPEC is the formation of attaching and effacing (A/E) lesions on the apical surface of intestinal epithelial cells (Fig. 218.3).[210] A/E is orchestrated by genes on a pathogenicity island known as the locus of enterocyte effacement (LEE), which encodes a T3SS, the adhesin intimin—encoded by the *eae* gene—and the translocated intimin receptor (Tir) and other EPEC-secreted protein (Esp) effectors.[101,211,212] Other important effector proteins are encoded outside the LEE.[213,214]

The protein EspA is a filamentous extension of the EPEC T3SS that connects and forms a canal to the host cell.[215] Tir transits the EspA canal, enters a pore in the host cell membrane formed by the proteins EspB and EspD, and then inserts into the host membrane to function as a receptor for intimin.[54] Each intimin receptor dimer binds two intimin molecules protruding from the bacterial surface, activating the N-WASP and Arp2/3 actin-nucleating and filament machinery,[216] resulting in host cell microvilli loss and the formation of a cuplike actin and cytoskeletal pedestal (see Fig. 218.3).[56,216–221] Other effector proteins injected by the EPEC T3SS interrupt the NK-κB pathway to inhibit the host's innate immune response.[222–224] Both intimin and EspB are proven EPEC virulence factors,[184,225] and triple-mutant tEPEC strains that do not express BFP, EspA, or intimin fail to adhere to intestinal epithelial cells.[226]

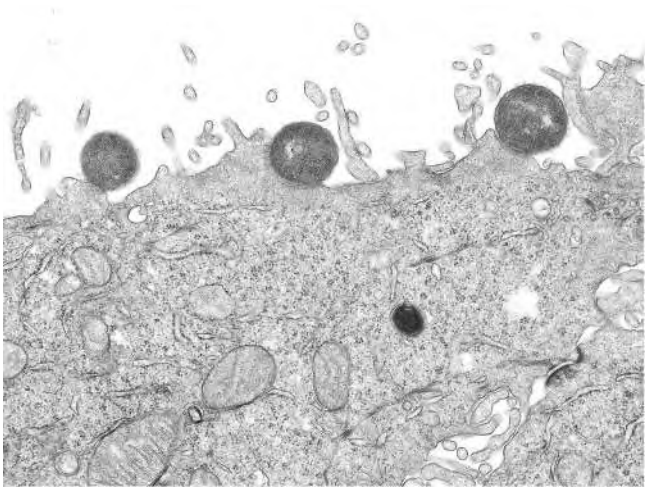

FIG. 218.3 Transmission electron micrograph of the attaching and effacing effect of enteropathogenic *Escherichia coli* on a tissue culture cell. Note the loss of microvilli, the intimate attachment of bacteria to the cell, and the cuplike pedestal composed of cytoskeletal proteins to which the bacteria are attached. Attaching and effacing effects are also seen with enterohemorrhagic *E. coli* strains and *Escherichia albertii* strains.

The precise mechanism of EPEC diarrhea is not fully understood, although it does not appear to be mediated by toxin production.[100] It has been shown that EspF—another protein secreted by the EPEC T3SS, although not involved directly in A/E lesion formation—participates in protein-protein interactions including sorting nextin 9 (SNX9), N-WASP, and the tight-junction protein occludin, resulting in decreased intestinal barrier function.[227–230] Other TSS3 proteins, including EspG and EspG2, have been implicated in inhibiting luminal chloride transport.[231] Furthermore, research shows that malnutrition resulting from protracted disease may be worsened by localized EPEC inhibition of thiamine uptake.[208]

EPEC diagnostics typically rely on probes targeting genes encoding hallmark EPEC genetic elements, including *eae* +/− *bfpA* but lacking *stx*.[232] A rapid agglutination test specific to EspB was developed for use in low income countries.[233]

Like ETEC, EPEC is a significant pathogen contributing to traveler's diarrhea (discussed earlier and in Chapter 319).[118,122,123,128] Data suggest that antibiotics are warranted in severe endemic EPEC disease, including nosocomial outbreaks,[234,235] although EPEC resistance to multiple antibiotic classes is a formidable concern.[236–240]

Human breast milk contains factors—including antibodies against BFP, intimin, EspA, EspB, and O antigens—that inhibit EPEC adherence.[198,241–249] Vaccines targeting BFP, EspB, and intimin have been explored, and EPEC strains transformed into "bacterial ghosts" by lysis plasmids elicit protective immunity in mouse models.[250] Unfortunately, immunogenicity data from human studies have proven underwhelming, presumably owing to antigen heterogeneity.[191,225,251–254]

Shiga Toxin–Producing *Escherichia coli* Including Enterohemorrhagic *Escherichia coli*

STEC incorporates any of the more than 400 *E. coli* serotypes that produce Shiga toxin, sometimes referred to as verotoxin.[255] STEC infection results in watery, often bloody diarrhea potentially complicated by hemolytic-uremic syndrome (HUS). STEC hemorrhagic colitis is often clinically mistaken as noninfectious hematochezia because of lack of fever.

HUS—a thrombotic microangiopathy resulting in anemia, thrombocytopenia, and renal failure—is the leading cause of renal failure in children. Thrombosis from HUS can lead to organ failure elsewhere including ischemic bowel, peripheral necrosis, and neurologic complications with cerebrovascular accident.[256–258] Approximately 30% of patients develop long-term sequelae including cardiac, renal, gastrointestinal, neurologic, and even endocrine complications.[259,260] HUS is more likely to complicate pediatric or elderly infections, with mortality ranging from 0.5% to 11% depending on the strain and patient age.[260–264]

STEC includes the pathotype EHEC, which has historically been responsible for large HUS outbreaks, including those due to the infamous O157:H7 serotype. This being said, the largest HUS outbreak to date was caused by an enteroaggregative O104:H4 STEC strain in northern Germany.[265,266] The prevalence and virulence of other STEC serotypes has varied considerably by geography.[265,267,268]

Large herbivorous mammals, such as cattle, are the primary natural reservoir for STEC.[269,270] STEC biofilm formation—observed on bovine hides, carcasses, and even abiotic surfaces—affords certain strains the ability to persist in the environment.[271,272]

STEC infection results from ingesting a small inoculum—as few as 100 organisms—from undercooked beef or any contaminated produce, water, or other beverage. A multistate outbreak of STEC serotypes O121 and O26 was attributed to raw flour consumption from a common production facility.[273] Fecal-oral transmission has also been documented from farm animal exposure, petting zoos, and person-to-person contact.[274–284]

The median incubation period for STEC infection is approximately 3 days, with bloody diarrhea typically developing after 2 days.[285] Most STEC cases resolve spontaneously after 1 week, although 4% to 22% of patients may progress to develop HUS, depending on infecting strain

and patient age.[262,264,266,285–287] In one large person-to-person child care outbreak, STEC shedding continued for a median duration of 30 days and up to 52 days from symptomatic individuals.[288]

Shiga toxins are a group of cytotoxic virulence factors—including Stx1 and Stx2 variants— with structural similarities to *Shigella dysenteriae* toxin that readily spread by transfection as *stx* genes are encoded on bacteriophages.[289] Shiga toxin is composed of a pentameric B subunit that binds to globotriaosylceramide and glycosphingolipids on the surface of host cells. After endocytosis, a single A subunit catalyzes the depurination of an adenine residue from the 28S rRNA of the host ribosome, rendering it nonfunctional.[290,291] Shiga toxin also affects cell signal transduction, prompting inflammation and apoptotic responses.[292,293] Furthermore, Shiga toxin can bind to leukocytes, facilitating dissemination through the bloodstream where endothelial cells elsewhere are then intoxicated, increasing procoagulant expression and subsequent microvascular thrombosis.[294–296]

EHEC is considered distinct from other STEC strains for producing the A/E lesions on intestinal cells also observed in EPEC (see Fig. 218.3).[297] As detailed earlier regarding the pathogenesis of EPEC, EHEC A/E lesion formation is orchestrated by the pathogenicity island LEE. However, in contrast to EPEC stains, genomic research shows that EHEC has a much larger effector repertoire, with the EHEC T3SS linked to a complex phage "metagenome," reflecting the complexity of LEE-positive STEC virulence, especially for O157:H7 serotypes.[298–300] Increasingly, LEE-negative STEC strains are recognized agents of diarrhea and even HUS, with virulence factors suspected to be similar to those of EAEC.[110]

Suspicion of STEC should be triggered by careful clinical history, physical examination, and laboratory parameters corresponding with HUS, such as schistocytes on peripheral smear. Sorbitol-MacConkey agar plates can be used to detect O157:H7 colonies but will fail to reveal other STEC serotypes; the microbiology laboratory should be notified of any concern for STEC because PCR assays and other immunoassays exist to readily detect *stx* genes.[117,301–304] The delayed occurrence of HUS—sometimes more than a week after onset of diarrhea—can confound proper diagnosis because STEC may no longer be detectable in stool.[154] STEC or HUS cases should prompt immediate alert of public health officials to the threat of an outbreak.[261]

Swift STEC diagnosis is critically important because proper identification will alter therapeutic management. Cellular stress—observed after exposure to fluoroquinolones and trimethoprim—induces high levels of Shiga toxin, which then lyses STEC strains.[305] Despite in vitro studies suggesting that certain antibiotics—such as azithromycin—may reduce Shiga toxin levels, antibiotic therapy is not recommended for STEC based on clinical data associating treatment with higher risk of HUS, observed across multiple antibiotic classes.[149,262,306,307]

The cornerstone of STEC treatment remains volume resuscitation and avoidance of antimotility agents.[308–310] Intensive care including renal replacement therapy may be required depending on the severity of disease or progression to HUS, although there is no evidence to support plasmapheresis for HUS due to STEC.[266,311,312] Case series have suggested improved outcomes with eculizumab—a monoclonal antibody against complement factor C5—in cases of HUS due to STEC, but this benefit was not observed during the German O104:H4 outbreak.[266,313–315]

Preventing diarrhea from STEC relies on safe food handling and consumption, including cooking ground beef to at least 155°F (68.3°C) (until "the juices run clear") and avoiding cross-contamination from undercooked meat.[278,316] Unpasteurized juices and milk also carry risk for STEC infection.[317] Proper hygiene after exposure to water in public swimming areas and animals at farms or petting zoos also reduces disease risk.

Research into STEC treatment other than antimicrobial therapy continues, including work with monoclonal antibodies against Stx1 and Stx2.[318–325] Oral toxin receptor analogues showed promise in protecting mice from sequelae of STEC infection but failed to reduce the severity of disease in children in a clinical trial.[326] STEC vaccine development is ongoing, targeting O157 antigens, Shiga toxin, intimin, and other T3SS secreted proteins.[327–332] Construction of STEC "bacterial ghosts" has shown promise as a vaccine candidate, as has Shiga toxin neutralization by a receptor mimic.[333,334]

Enteroaggregative *Escherichia coli*

The term *enteroadherent-aggregative E. coli* was initially coined to characterize strains isolated from the diarrhea of Chilean children with non-EPEC patterns of adherence observed in HEp-2 cells.[335] This group was ultimately separated into two pathotypes: EAEC and DAEC.[154,335,336]

EAEC strains are defined by their characteristic "stacked brick" aggregative adherence (AA) pattern on tissue culture cells, which has since been observed on intestinal mucosa and even abiotic surfaces (Fig. 218.4B).[337,338] EAEC was first described in 1987 and is increasingly recognized as a major cause of diarrhea in both developed and developing countries.[335,339,340] In large prospective studies in the United States, EAEC was the most common bacterial pathogen isolated from diarrheal samples.[187,341]

EAEC transmission is via the fecal-oral route, with different strains accounting for a wide variety of pathogenicity ranging from asymptomatic carriage to the severe European HUS outbreak attributed to fenugreek sprouts contaminated with the Shiga toxin–producing O104:H4 strain.[286,342–346] EAEC has been implicated in large foodborne outbreaks and traveler's diarrhea—with a frequency rivaling ETEC—and in postinfectious irritable bowel syndrome.[118,122,347–351] EAEC infection contributes significant morbidity to endemic cases of chronic diarrhea in children and HIV patients.[352–358]

Even in the absence of diarrhea, children in resource-poor environments with EAEC isolated in their stool have significant growth impairment, associated with increased interleukin (IL)-8 production[359,360] regulated through mitogen-activated protein kinases (MAPKs) that induce transcription factors NFκB and AP-1.[361] Several single nucleotide polymorphisms (SNPs)—including an SNP in the IL-8 gene promoter—have been linked to susceptibility to EAEC diarrhea.[362–365]

EAEC causes mucosal damage after adherence, resulting in microvilli loss and cell death, although the heterogeneity of a given EAEC strain—observed even in a single outbreak—defies a unifying account of pathogenesis.[338,349] EAEC aggregative adherence has been linked to the expression of chaperone-usher type adhesins encoded on plasmids, known as aggregative adherence fimbriae (AAF), which are related to the Dr family of adhesins found in DAEC and some UPEC strains.[366–369] AggR—a positive regulator of the AAF operon—has been proposed as the defining characteristic of typical EAEC, responsible for expression of multiple virulence factors associated with disease, including AAF, the surface protein dispersin, and the Aai T6SS.[91,370–372] Nevertheless, so-called atypical EAEC strains without AggR are still widely associated with diarrheal illness, reflecting the diversity of EAEC virulence yet to be described.[373]

EAEC strains can form a thick biofilm on the intestinal mucosa and on abiotic surfaces, which can harbor organisms in thick bacterial aggregates and hamper host nutrient absorption.[374–376] Pic—a mucinolytic SPATE protein—helps EAEC penetrate this biofilm but also induces mucus hypersecretion by host intestinal cells, suggesting a role in the creation of mucoid diarrhea.[377]

Several EAEC toxins have been described, including the enteroaggregative heat-stable enterotoxin 1 (EAST-1), also found in many non-EAEC strains.[378] EAST-1 shares structural similarities with the heat stable toxins of ETEC, and its pathogenesis also begins with increase of cGMP.[379,380] Some EAEC strains produce other SPATE family toxins such as plasmid-encoded enterotoxin (Pet), which causes intestinal crypt dilation and cell damage.[381,382]

The diagnosis of EAEC can be made by specific clump-formation tests, although most laboratories rely on tissue culture adhesion assays.[335,383,384] PCR and DNA probes for AggR have proven useful but are not commercially available.[385] Regardless, confirming EAEC as the causative pathogen is complex, given the spectrum of clinical disease and asymptomatic colonization.

The bulk of data regarding EAEC antimicrobial therapy is limited to the study of traveler's diarrhea, suggesting benefit in appropriate cases, although small case reports also support antibiotics to treat chronic diarrhea attributed to EAEC in HIV patients.[118,128,386,387]

In contrast to most EAEC strains, antimicrobial management was well studied during the 2011 Shiga toxin–producing EAEC outbreak in Germany. Whereas worse outcomes have been associated with antibiotic therapy for STEC diarrhea, one medical center provided

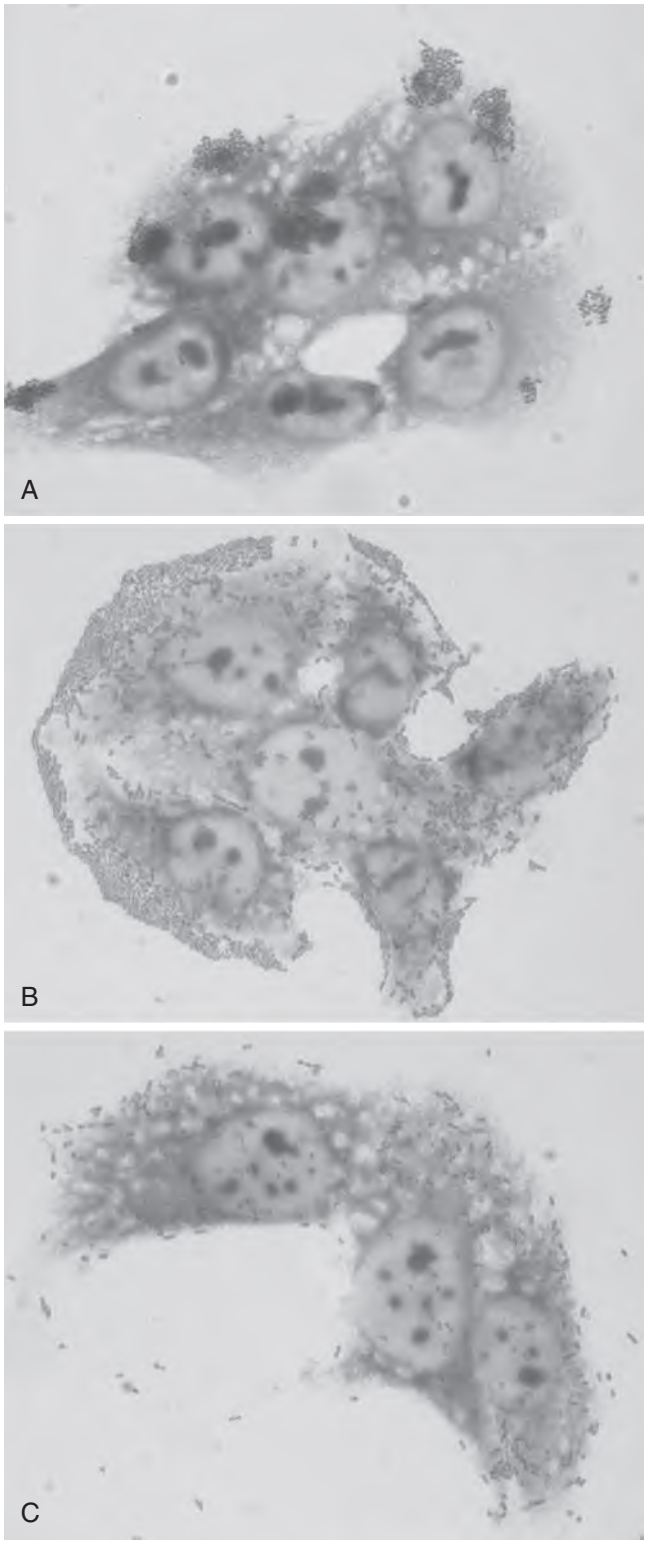

FIG. 218.4 Patterns of cellular adherence displayed by various diarrheagenic *Escherichia coli* pathotypes. (A) The localized adherence pattern exhibited by typical strains of enteropathogenic *E. coli* is characterized by discrete three-dimensional microcolonies of bacteria. (B) The aggregative adherence pattern exhibited by enteroaggregative *E. coli* strains is characterized by two-dimensional "stacked brick" aggregates. (C) Diffuse adherent *E. coli* strains adhere individually at random to tissue culture cells.

aggressive antimicrobial therapy to patients with HUS due to the German O104:4 strain, resulting in shorter duration of EAEC excretion, fewer seizures, and fewer deaths compared with historical patients untreated with antibiotics.[266] Patients studied elsewhere during the outbreak who received azithromycin for meningitis prophylaxis as part of a regimen including eculizumab were less likely to have long-term O104:H4 carriage, suggesting a role for antibiotics to prevent colonization.[388] Antibiotic therapy for this unique EAEC strain and the risk for HUS remain controversial.[389,390] Treatment data from the O104:H4 outbreak should not be widely extrapolated to less idiosyncratic EAEC strains.[110,391] Furthermore, any empirical antibiotic regimen for EAEC infection is potentially confounded by reports of widespread multidrug resistance, including ESBL and AmpC variants.[392]

Early vaccine studies against ETEC heat-labile toxin incidentally showed decreased burden of EAEC in the treatment group, although this ultimately failed to protect travelers from diarrhea due to any organism in a phase III clinical trial.[174,393]

Enteroinvasive *Escherichia coli*

The pathogenesis of EIEC infection is quite similar to that of *Shigella* and is discussed in more detail elsewhere (see Chapter 224). Although EIEC and *Shigella* represent distinct genetic lineages within the genus *Escherichia,* their similarities reflect convergent evolution, including plasmid exchange and the loss of antivirulence factors—termed pathoadaptation—through deletion, insertion, and point mutations.[1,394–396] *Shigella* retains its genus for historical reasons but is now considered to belong to *E. coli* based on phylogenetic data.[397] Like *Shigella*, EIEC are typically not motile, cannot ferment lactose, and are lysine decarboxylase negative.[398]

EIEC infection—first described in the 1940s—is clinically indistinguishable from shigellosis, although the inoculum required for disease appears considerably higher and typically results in less severe diarrhea.[399,400] EIEC diarrhea produces watery stools that can progress to dysentery, especially affecting children in the developing world.[400–405] Person-to-person and foodborne outbreaks—implicating contaminated cheese, milk products, and beef—occur in the developed world, with large outbreaks in Europe attributed to ingestion of lettuce contaminated with the motile serotype O96:H19, not previously reported in EIEC or *Shigella*.[406–408]

The hallmark feature of EIEC is cell invasion, orchestrated by a large plasmid—*pInv*—that encodes a T3SS, allowing the organism to escape macrophage phagocytosis, invade the intestinal epithelial cell, and usurp the host actin-filament machinery to spread from cell to cell.[409–411] EIEC is as capable of invasion as *Shigella,* but subsequent expression of virulence genes is lower.[412,413]

Historically, *Shigella* and EIEC were characterized by a positive Sereny test result wherein the isolate was observed to cause keratoconjunctivitis in guinea pigs.[414] Because of their close genetic relationship, distinguishing *Shigella* from EIEC is difficult. A combination PCR and biochemical assay strategy has been proposed, although often MLST analysis is required to confirm the diagnosis.[415]

EIEC diarrhea is usually self-limiting, but antibiotic therapy is warranted for severe cases. However, the clinician must be careful to first exclude the possibility of STEC infection.[416] As with other DEC strains, multidrug-resistant (MDR) EIEC strains have been reported, prompting consideration of local resistant patterns when empirical therapy is initiated.[417]

Preventing EIEC infection relies on proper hygiene and sanitation to avoid ingestion of contaminated food or water. *Shigella* vaccine development is addressed in Chapter 224.

Diffusely Adherent *Escherichia coli*

Diffuse adherence (DA) in tissue culture was observed in the initial adherence assays for non-EPEC strains, but this pattern was not clearly associated with diarrhea[335,336,418] (see Fig. 218.4C). Adult volunteers challenged with DAEC did not develop infection.[419] DAEC is now generally accepted as a DEC pathotype based on its association with watery diarrhea typically affecting somewhat older children worldwide.[100,154,420–424] Although virulence has been well established in UTI, the role of DAEC in diarrhea outside of childhood remains unclear.[425]

DAEC organisms are frequently isolated in asymptomatic children and adults, and molecular testing may have overestimated DAEC detection in diarrhea because of DNA probe cross-reactivity with EAEC strains.[425–427]

The characteristic diffuse adherence of DAEC has been attributed to both fimbrial and afimbrial adhesins from the Afa/Dr family, described initially in a diffusely adherent UPEC strain.[425,428,429] Operons of the *afa* family produce heterogeneous adhesins, with some recognizing the Dr blood group antigen as a receptor on human decay-accelerating factor (DAF), termed Afa/Dr+ adhesins.[430] Afa/Dr+ adhesins remain the defining feature of so-called typical DAEC, whereas diarrheagenic DAEC strains not expressing Afa/Dr have been grouped into the atypical EPEC pathotype.[100]

Afa/Dr+ adhesin binding to DAF on the brush border of epithelial cells increases permeability due to cytoskeletal changes, defective brush border proteinsm and microvilli loss.[431–437] DAEC flagellar stimulation of Toll-like receptor 5 (TLR5) induces IL-8 secretion then transepithelial migration of neutrophils, with subsequent inflammation further promoting DAF expression.[438–440] The presence of ileal DAEC and increased expression of DAF in patients with Crohn disease (CD) has led to speculation about the role for DAEC in CD pathogenesis.[428,441,442]

Diarrheal DAEC strains from a Brazilian pediatric case-control study showed genetic expression of a wide variety of previously described toxins—including EAST-1, ShET1 (Shigella enterotoxin 1), and hemolysin—which has made reliable laboratory detection and meaningful vaccine development challenging.[422] Antimicrobial and vaccine strategies for extraintestinal DAEC—namely, uropathogenic strains—are discussed later.

Adherent-invasive *Escherichia coli*

The term *adherent-invasive E. coli* was coined in 1999 to describe *E. coli* strains isolated from resected ileal tissue in patients with CD. AIEC does not express any traditional DEC virulence factors but can adhere and efficiently invade intestinal epithelial cells.[441,443]

Concordance studies have long implicated environmental factors in CD pathogenesis with many organisms identified as potential culprits, including viral and mycobacterial agents.[444,445] Whereas AIEC is found in less than 10% of healthy ileums, AIEC is isolated from approximately 30% of CD ileal lesions, suggesting its role as a possible pathobiont in the multifactorial etiology of CD.[446–448] AIEC is a diverse group, although particular strains such as those from the B2 phylogroup—associated with extraintestinal infection—have been isolated with higher prevalence from CD patients.[449–452]

AIEC adherence involves type I fimbriae and long polar fimbriae (LPF) adhering to M cells at the surface of the Peyer patches (PPs) and CAECAM6 on host intestinal epithelial cells.[453] Invasion is then facilitated by the expression of a membrane-bound protein, OmpA, that promotes fusion of outer membrane vesicles (OMVs) with Gp96—a host ER stress response chaperone—that is overexpressed in the ileum of CD patients.[454–456] AIEC then replicates in macrophages of the lamina propria, inducing TNF-α but not provoking macrophage death.[457,458]

Although CD treatment is beyond the scope of discussing AIEC alone, it is worth noting that CD symptoms improve when luminal bacteria burden is decreased, leading clinicians to use antibiotics targeting enteric bacteria during CD flares.[459]

Extraintestinal Pathogenic *Escherichia coli*

ExPEC strains have distinct epidemiologic, phylogenetic and virulence factor profiles relative to commensal *E. coli* and DEC.[460,461] ExPEC strains are capable of infection—community acquired and nosocomial—involving almost any organ or anatomic site.[112] Based on characteristic virulence factors and on MLST and serotyping data, particular ExPEC variants are often described as *uropathogenic E. coli, neonatal meningitis–associated E. coli,* or *sepsis-associated E. coli.*[462]

Despite such syndromic classification, ExPEC strains can share virulence factors irrespective of their anatomic niche.[462] For example, prototypical UPEC strains have been isolated from cases of pneumonia, and characteristic NMEC strains frequently cause UTI.[463,464] Certain serotypes—such as *E. coli* O18:K1:H7—have been recovered from urine, feces, and CSF, and these isolates are clearly clonal derivatives.[465] Furthermore, *E. coli* isolates thought to represent commensal strains can express ExPEC virulence factors and cause infection—including bacteremia—in the appropriate clinical scenario, including immune compromise, retained nidus of infection, or obstruction.[102,113,466–469]

ExPEC strains are typically derived from the B2 or D phylogroup with virulence genes usually encoded on plasmids or within pathogenicity islands (PAIs) distinct from DEC.[112] Some researchers have defined ExPEC as expressing at least two of the following genes: *papA* or *papC* (P fimbriae major subunit and assembly), *sfa/foc* (S and F1C fimbriae), *afa/draBC* (Dr-binding adhesins), *kpsM II* (group 2 capsule), and *iutA* (aerobactin receptor).[470] ExPEC virulence genes are detected in approximately 10% of healthy individuals, supporting the concept that the intestinal microbiota serves as a reservoir for ExPEC strains.[469]

The pandemicity of hypervirulent and MDR strains underscores the importance of further characterization of ExPEC.[471,472] Particular ExPEC sequence types—such as ST101, ST131, and ST405—are associated with extensive antimicrobial resistance, including plasmid-mediated AmpC β-lactamases, CTX-M ESBLs, NDM MBLs, and other carbapenemases.[473–479] General principles of antibiotic resistance are discussed elsewhere (see Chapter 18).

Uropathogenic *Escherichia coli*

E. coli causes the majority of UTIs and pyelonephritis.[480] The host vaginal or fecal microbiota is the most common source for these strains—termed *uropathogenic E. coli*—although UPECs are not necessarily the predominant clones from these reservoirs.[481,482] UPEC clonal outbreaks have prompted investigation into common virulence factors particularly suited for genitourinary colonization and infection.[483–489] Murine and primate models have improved understanding of urinary tract pathogenesis, although UPEC strain heterogeneity continues to inspire research into novel virulence factors and potential vaccine targets.[490–494]

Many adhesins have been implicated in UPEC pathogenesis, but the ubiquitous *E. coli* type 1 fimbria—encoded by the *fim* operon—has proven essential to infection.[32,46,493,495–497] The apical layer of the normally impermeable bladder transitional epithelium features "umbrella cells" composed of highly ordered plaques featuring lipid components and uroplakin particles.[498] UPEC type 1 fimbriae bind to uroplakin, where the innate defense system induces host epithelial cell exfoliation followed by bacterial penetration through lipid rafts.[499–501]

UPEC expression of type 1 fimbriae genes is controlled by an invertible promoter capable of phase variation, allowing the virulence of the organism to change depending on the stage of infection.[49,502–504] Flow chamber models show that expression and adhesion of type 1 fimbriae actually increase with shear stress after they settle on bladder epithelial cells or even catheter surfaces, reflecting more pathogenic sessile subpopulations.[505–507] Furthermore, UPEC type 1 fimbriae have been implicated in decreased ureteral contractility in rat models, suggesting a role in ascending infection.[508]

The chaperone-usher type P fimbriae—encoded by *pap* genes—was the first virulence factor to be discreetly associated with UPEC infection, especially pyelonephritis.[509,510] P fimbriae bind to both galactosyl-galactose moieties on erythrocytes of the P blood group and glycosphingolipids on the surface of renal epithelial cells. Mutations in *papG*—encoding the PAP adhesin on the P fimbrial tip—resulted in less virulent pyelonephritis in a cynomolgus monkey model.[511–515] *E. coli* has higher prevalence of vaginal colonization in women who do not secrete histo-blood group antigens, which increases risk for recurrent cystitis.[516]

Although described earlier in the discussion of DAEC pathogenesis, the family of Afa/Dr adhesins was first characterized in UPEC strains associated with recurrent urogenital infections, especially in pregnant women.[488,517–521] Whereas Afa/Dr+ adhesins are the defining characteristic of DAEC, both Afa/Dr+ and Afa/Dr– adhesins have been described in cystitis, pyelonephritis, and sepsis from a urinary source.[430] Afa/Dr+ adhesins bind to type IV collagen, resulting in tropism for the renal basement membrane, observed to cause chronic pyelonephritis in a mouse model.[522,523] UPEC also shares virulence factors with EAEC, which is thought to be a recent common ancestor.[524,525] For example, a large Danish community-acquired EAEC UTI outbreak confirmed the urovirulence of AAF.[526]

After adherence, UPEC strains are capable of invading and replicating in host epithelial cells, forming intracellular bacterial communities

(IBCs), which affords protection from host defenses and antimicrobial therapy and potentially serves as a reservoir for recurrent UTI.[527–531]

UPEC strains produce hemolysin—the predominant toxin found in ExPEC—which creates pore formation and induces low-frequency calcium oscillations in the renal epithelium, stimulating IL-6 and IL-8.[66,532] Hemolysin results in uroepithelial sloughing and rapid bladder hemorrhage in mouse models, with induction of caspase-dependent inflammatory death observed in human urothelial cells.[533,534] A third of UPEC strains also produce cytotoxic necrotizing factor 1 (CNF1), which inactivates Rho GTPases, resulting in reorganization of actin assembly and multinucleation in culture cells.[535,536] CNF1 results in submucosal edema in the mouse bladder with attenuated disease observed in CNF1-negative strains.[533,537]

UPEC pathogenicity is augmented by multiple iron acquisition systems. UPEC strains with mutated TonB outer membrane receptors failed to utilize the siderophores enterobactin and aerobactin and are less infective than wild-type strains.[87] Many other putative virulence factors have been suggested by experiments showing attenuated disease in mutated UPEC strains, including mutations in the O antigen, K capsule and various regulatory genes.[538–543]

Owing to the ubiquity and common recurrence of UPEC infection, prophylactic therapeutics other than antimicrobials continue to be an active area of research.[544] Data supporting prophylactic *Lactobacillus* vaginal suppositories—intended to alter the colonizing UPEC reservoir—have not proven convincing, although they may confer some protection when administered immediately after antibiotic treatment for cystitis.[545–547] Anecdotal reports of cranberry product ingestion as prophylaxis against UPEC were supported by in vitro evidence showing inhibition of both type 1 and P pilus-mediated adherence, but, unfortunately, results from placebo-controlled trials are largely disappointing.[548–552] Meta-analyses have suggested benefit from short-term prophylactic methenamine hippurate in preventing UTI in patients without renal tract abnormalities, and vaginal estrogen in preventing recurrent UTI in postmenopausal women.[4,553–555] Oral FimH antagonists have shown success in preventing UTIs in mouse models.[556,557] The general clinical approach to UTI otherwise is discussed elsewhere (see Chapter 72).

Vaccine development to prevent UPEC infection has been ongoing since the 1950s, although success has been limited by the overwhelming heterogeneity of strains.[544,558,559] Iron acquisition systems—such as the yersiniabactin receptor FyuA—appear to be a compelling vaccine target, with success demonstrated in protecting mice against pyelonephritis.[560,561] Vaginal delivery of a multivalent whole-cell vaccine—featuring strains from many uropathogenic bacteria—seemed to reduce recurrent UPEC cystitis, particularly in sexually active women younger than 52 years.[562] An intramuscular biconjugate vaccine composed of O antigens from four UPEC serotypes induced significant immunoglobulin G (IgG) response and appeared safe in human phase I trials.[563] Finally, an oral vaccine composed of bacterial extracts from multiple UPEC strains is available in Europe and may have some possible benefit as prophylaxis against recurrent UTI, although the quality of evidence is low.[564–566]

Neonatal Meningitis–Associated *Escherichia coli*

E. coli is the most common gram-negative pathogen recovered from neonatal meningitis, typically preceded by high-grade bacteremia.[567,568] *E. coli* sepsis in the newborn is presumed to result from immature host immune defenses; deaths from NMEC infection are associated with preterm birth.[569,570]

Approximately 80% of NMEC strains feature the K1 polysialic acid capsule, which is immunochemically identical to the capsular polysaccharides of group B *Neisseria meningitidis* that confer protection from phagocytosis.[569,571,572] Early rat models suggested that the K1 capsule was critical in the development of *E. coli* meningitis, with both K1+ and K1− strains recovered in the cerebrospinal fluid (CSF) but only K1+ strains remaining viable, which suggests that the capsule is not necessary for invasion but facilitates survival after bacterial cells cross the blood-brain barrier (BBB).[80,573]

The NMEC outer membrane protein A (OmpA) allows survival in the serum by binding C4b-binding protein (C4bp) to inhibit complement activation.[574] Both type 1 and type S fimbriae and a number of other NMEC proteins—including OmpA and invasion of brain endothelial cell (Ibe) proteins—bind to particular epitopes on human brain microvascular endothelial cells (HBMECs), then facilitate invasion and loss of BBB integrity.[575–582]

NMEC strains are capable of expressing hemolysin and CNF—both encoded by a common pathogenicity island described in UPEC pathogenesis—and cytolethal distending toxin, which is associated with diarrheal disease.[575,583–586] Multiple NMEC gene mutations—including *cnf* and *clb* mutants—produce strains less capable of invading HBMECs, implicating a variety of virulence factors in central nervous system (CNS) disease worthy of ongoing research.[587–589] A French study of 325 NMEC cases recently characterized the largest number of isolates described to date, with more than 75% of the 141 different strains expressing genes implicating iron-uptake systems as a virulence factor, including *fyuA* (yersiniabactin), *iucC* (aerobactin) and *iroN* (salmochelin).[569] This study also revealed an association between disease severity and *pap*GII expression, previously linked with pyelonephritis but not bacteremia.[462,590]

The clinical approach to meningitis is discussed elsewhere (see Section H, "Central Nervous System Infections"), but clinicians should be cautious in choosing empirical antimicrobial therapy because epidemiologic data show increasing rates of drug resistance in NMEC isolates compared with UPEC, even of the same sequence type.[591]

Sepsis-Associated *Escherichia coli*

E. coli is the most common pathogen in gram-negative bacteremia, featuring a wide variety of strains sometimes described as *sepsis-associated E. coli*.[592] Compared with rectal strains from uninfected controls, *E. coli* virulence factor profiles—including expression of *pap* (P fimbriae), *hyl* (hemolysin), *fyuA* (yersiniabactin), *kpsM* II (capsule) and *ompT* (outer membrane protein)—serve as a better predictor of bloodstream infection than phylogenetic grouping.[593] However, in a study of more than 1000 adult patients with *E. coli* bacteremia, no single bacterial virulence factor predicted severity of disease. Age, cirrhosis, immune compromise and a cutaneous portal of entry were predictive of death, whereas *E. coli* bacteremia from UTI carried a more favorable prognosis.[594]

The usefulness of the encompassing term *sepsis-associated E. coli* to characterize these strains is confounded by the fact that the majority of *E. coli* bacteremia in adults results from UPEC. Likewise, NMEC is frequently isolated in the context of bacteremia. The pathogenesis attributed to UPEC and NMEC was described earlier in this chapter. Research is ongoing into *E. coli* virulence factors specific to bacteremia from gut translocation in the absence of other identifiable sources.[595,596]

Possible mechanisms contributing to *E. coli* gut translocation include intestinal overgrowth, host immune deficiency, and damage to the intestinal mucosa.[597] *E. coli* gut translocation is observed frequently in patients with hematologic malignancy. Up to 70% of leukemic patients with *E. coli* bacteremia do not have an identifiable source, although gut translocation is suspected based on DNA similarity between blood and bowel isolates.[598] A subsequent analysis implicated multiple virulence genes contributing to bowel-blood translocation.[599] A murine chemotherapy-induced translocation model showed that *E. coli* with *fimH* deletion were less capable of translocation, suggesting a role for FimH in gut translocation.[600]

E. coli translocation is also frequently observed, although poorly investigated, in infants. One study defined *E. coli* from infantile gut translocation as a bloodstream isolate with the same virulence genotype and serotype as that recovered from gastric fluid or stool without a source identified elsewhere, including a negative urine culture. Analysis of 100 such isolates appeared similar to UPEC strains determined with MLST, but gut translocation isolates more frequently expressed *ibeA*, which has been implicated in neonatal meningitis, although only one infant had meningitis in the study.[601]

Considering the increasing frequency of ExPEC infection—including UPEC, NMEC, and SEPEC—in the setting of increasing antimicrobial resistance, researchers are actively identifying antigen targets for vaccine development.[602] Subtractive reverse vaccinology identified more than 200 ExPEC antigens absent in nonpathogenic *E. coli*, with a subset of these antigens showing promise in a mouse challenge model.[603]

Klebsiella

The genus *Klebsiella* was named by Trevisan to honor the microbiologist Edwin Klebs. *K. pneumoniae, K. oxytoca,* and *K. granulomatis* are the primary species associated with illness in humans. *K. planticola, K. ornithinolytica*, *K. terrigena, and K. variicola* are rarely associated with human clinical disease and are now classified in the genus *Raoultella*[604,605]; these organisms have been associated with scrombroid fish poisoning. *Klebsiella ozaenae* and *Klebsiella rhinoscleromatis* are considered non-fermenting subspecies of *K. pneumoniae* based on DNA homology studies and exhibit characteristic clinical manifestations. With those exceptions, strains within this genus ferment lactose, most produce highly mucoid colonies on plates, and all are nonmotile. *Klebsiella* organisms are found commonly in the environment, but also can asymptomatically colonize the human nasopharynx, gastrointestinal tract, and, less frequently, other sites.[606,607] Gastrointestinal colonization may precede abdominal infections, increasing risk of abdominal infection up to sevenfold, with increasing colonization rates in hospitalized patients, those with prolonged lengths of stay, and those with prior antibiotic receipt.[606] Given higher rates of intraabdominal *Klebsiella* infections in East Asian countries, especially Taiwan, ethnicity was believed to confer increased risk, but intestinal colonization rates that differ by geography account for increased disease.[608,609] Seasonal variation has been noted in *Klebsiella* bacteremia, similar to *E. coli*, but not *Enterobacter* or *Serratia.*[116,610,611]

Most *K. pneumoniae* infections are nosocomially acquired,[612–615] particularly in ICU settings,[616] but they can also occur in otherwise healthy people. UTIs, pneumonia, and bacteremia are the most common infections encountered, followed by liver abscess and other intraabdominal infections, wound infections, infections of intravascular and other invasive devices, and meningitis and postneurosurgical infections. Patients with various underlying conditions including alcohol use, diabetes, malignancy, end-stage renal disease, and immune suppression are at greatest risk.[608,617,618] Nosocomial outbreaks involving *K. pneumoniae* and, less commonly, *K. oxytoca* have frequently been reported.[613,618,619] Nosocomial meningitis, especially with preceding neurosurgical procedures, varies based on procedure type[620,621] and prophylactic antibiotic regimen.[622] *Klebsiella* is an infrequent cause of community-acquired meningitis,[623,624] with the notable exception of metastatic disease related to *Klebsiella* spp. liver abscess (KLA). Only *E. coli* is more common than *Klebsiella* in spontaneous bacterial peritonitis in both the hospital and community.[625,626] Splenic abscesses are often related to KLA (see following text). Geographic variations exist for both infectious syndrome and phenotype of recovered *Klebsiella* isolate, with most variation seen in KLA and its sequelae.

K. pneumoniae is a significant cause of primary, invasive, community-acquired liver abscesses, often associated with metastatic spread.[627–631] Initially primarily reported in Taiwan, this syndrome remains widespread in East Asian countries but is emerging globally where it likely is underrecognized. KLA is often monomicrobial, commonly associated with preexisting hepatobiliary disease, and typically manifests with single, unilobular, and multiloculated fluid collections.[608,630,632] Prior antibiotic exposure increases KLA risk.[633] Isolates exhibiting hypermucoviscosity and a hypervirulent phenotype are often implicated in metastatic infection. Metastatic spread is more common with KLA than non-KLA infections, likely because of a combination of a variety of virulence factors observed both in KLA and severe *Klebsiella* spp. infections (see discussion of virulence, later). The most common metastatic manifestations are endophthalmitis, meningitis, and brain abscess, although other sites have been described.[634–637] Treatment of KLA requires parenteral antibiotic therapy (which is often prolonged) and often drainage. When endophthalmitis is present, severe visual deficits predominate, and when suspected, prompt evaluation, antibiotics, ophthalmologic consultation, and use of intravitreal antibiotics should be considered.[608]

K. pneumoniae can cause UTIs in individuals with normal or abnormal urinary tracts or with indwelling catheters. Pathogenesis of *Klebsiella* UTIs is similar to that related to other Enterobacteriaceae. Uncomplicated UTIs, ascending acute pyelonephritis, emphysematous genitourinary infections, perinephric abscess, and prostatic involvement caused by *K. pneumoniae* do not have distinguishing clinical features from those caused by other bacterial species. *K. pneumoniae* is second only to *E. coli* as a cause of both all-cause gram-negative bacteremia and bacteremia from a urinary source.[612,638,639] Community-onset bacteremia can occur and is often severe, necessitating ICU admission, partly because of overlap with the hypervirulent phenotype seen in community syndromes, which is associated with septic shock and poor outcomes.[640–643]

Pneumonia caused by *K. pneumoniae* classically invoked particular distinguishing features, warranting the eponym *Friedländer disease.* Among these classic features are its severity, frequency in alcoholics, propensity to affect the upper lobes, production of "currant jelly" sputum resulting from hemoptysis, the bulging fissure sign on radiography from edematous lobar consolidation, and tendency for abscess formation. Despite these compelling descriptions, pneumonia caused by *K. pneumoniae* cannot be reliably distinguished clinically from other organisms, and many of the aforementioned features may be the result of misdiagnosis, inferred from culture of expectorated sputum, of anaerobic pulmonary infections.[644] CAP is infrequently caused by *K. pneumoniae* in the United States,[645,646] although higher rates have been seen in Southeast Asia,[647] and carries with it increased mortality.[648] In geriatric populations, *Klebsiella* also has been implicated in aspiration pneumonia.[649]

Klebsiella has less frequently been described in various syndromes, including uncomplicated skin and soft tissue infections indistinguishable from those caused by typical pathogens, and necrotizing fasciitis, spinal infections, septic arthritis, cardiac infections, mycotic aneurysm, and infected grafts.

Several virulence factors have been extensively investigated for *K. pneumoniae* including its luxuriant polysaccharide capsule, mucoviscous phenotype, and siderophore regulation. At least 77 capsular antigenic varieties have been described, with important geographic variation. Capsular virulence is due to phagocytosis inhibition (see earlier discussion) and to protection from bactericidal serum factors.[608,627] Capsular types K1 and K2 appear to be more virulent and cause more severe human disease,[627,650] as supported by increased lethality in animal models.[651] This increased severity is partly related to a hypermucoviscous phenotype; conversely, mutant acapsular strains exhibit reduced virulence.[652] The microbiologic observation of the mucoid phenotype as indicated by a "positive string test" (Fig. 218.5) correlates with this hypermucoviscous phenotype and clinically invasive strains.

There is not a direct relationship between virulence factors and *Klebsiella* strain identification or capsular serotype, challenging initial concepts of virulence.[627] *Klebsiella* spp. exhibiting a hypermucoviscosity phenotype have long been thought to be more invasive and implicated in KLA,[653–656] but the variety of virulence factors and observance of nonmucoid KLA isolates make *hypervirulent phenotype* a more

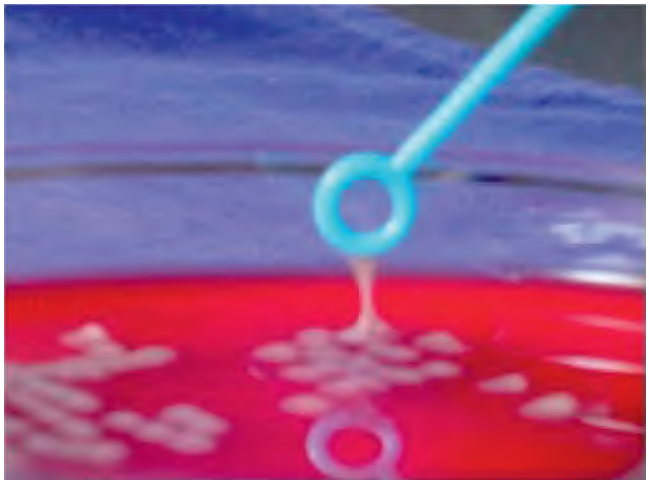

FIG. 218.5 Mucoid phenotype demonstrated by positive "string test." Viscous strings >5 mm in length are produced by stretching colony by loop on growth agar. *(From Siu LK, Yeh KM, Lin JC, et al. Klebsiella pneumoniae liver abscess: a new invasive syndrome.* Lancet Infect Dis. *2012;12:881–887.)*