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Clostridium difficile-associated Intestinal Disease and Probiotics

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Abstract

Probiotics are traditionally defined as viable microorganisms that have a beneficial effect in the prevention and treatment of pathologic conditions when they are ingested. Although there is a relatively large volume of literature that supports the use of probiotics to prevent or treat intestinal disorders, the scientific basis behind probiotic use has only recently been established, and clinical studies on this topic are just beginning to get published. Currently, the best studied probiotics are lactic acid bacteria, particularly *Lactobacillus* and *Bifidobacterium* species. Other organisms used as probiotics in humans include *Escherichia coli*, *Streptococcus* sp., *Enterococcus* sp., *Bacteroides* sp., *Bacillus* sp., *Propionibacterium* sp., and various fungi, and some probiotic preparations contain more than one bacterial strain. Probiotic use for the prevention and treatment of antibiotic-associated diarrhea caused by *Clostridium difficile* induced intestinal disease as well as for other gastrointestinal disorders has been discussed in this review.

Keywords: Probiotics, *Clostridium difficile*, *Lactobacillus*, antibiotic associated diarrhea

Clostridium difficile

Clostridium difficile is a spore-forming, obligate anaerobic, gram-positive, rod-shaped organism that is acquired from the environment or the fecal-oral route. The organism was first discovered by Hall and O'Toole in 1935 in the stool of newborns and was referred to as *Bacillus difficilis* owing to the difficulty in its isolation and study (Hall and O'Toole, 1935).

Although *C. difficile* was identified as the main causative agent of antibiotic-associated diarrhea (AAD) and pseudo-membranous colitis during the late 1970s (Bartlett *et al*., 1978a; Bartlett *et al*., 1978b), it has only received substantial attention since the late 1980s because of its increased incidence in hospitals worldwide (Cartmill et al., 1994; Johnson et al. 1999), after which it was renamed *C. difficile*.

C. difficile is indeed the most common cause of antimicrobial-associated diarrhea currently; 85% of *C. difficile*-infected patients receive antibiotics within 28 days of the onset of symptoms (Morris *et al*., 2002). The use of almost all antibiotics has been associated with *C. difficile* infection, including cephalosporin, penicillin, and clindamycin (McFarland *et al*., 1990; Chang and Nelson, 2000; Thomas *et al*., 2003). Clinical symptoms of infection vary widely, ranging from asymptomatic colonization to pseudomembranous colitis with bloody diarrhea, fever, severe abdominal pain, peritonitis, and toxic megacolon that can result in death (Clements *et al*., 2010).

The prevalence of *C. difficile* infections has increased dramatically since 2000 (Kuijper *et al*., 2006). The number of fatal cases of *C. difficile* infection in England increased from approximately 500 in 1999 to nearly 3400 in 2006 (Kelly and LaMont, 2008). Zilberberg *et al*. (2010) reported a recent increase in the number of severe cases of *C. difficile* infection in children, from 3565 cases in 1997 to 7779 cases in 2006. This increase was associated with the presence of the newly discovered hypervirulent strain B1/NAP1/027 (Winter and Jayasekera, 2013). Related epidemic strains of *C. difficile* have been identified as the causes of hospital outbreaks within North America and Europe, and *C. difficile* infections in

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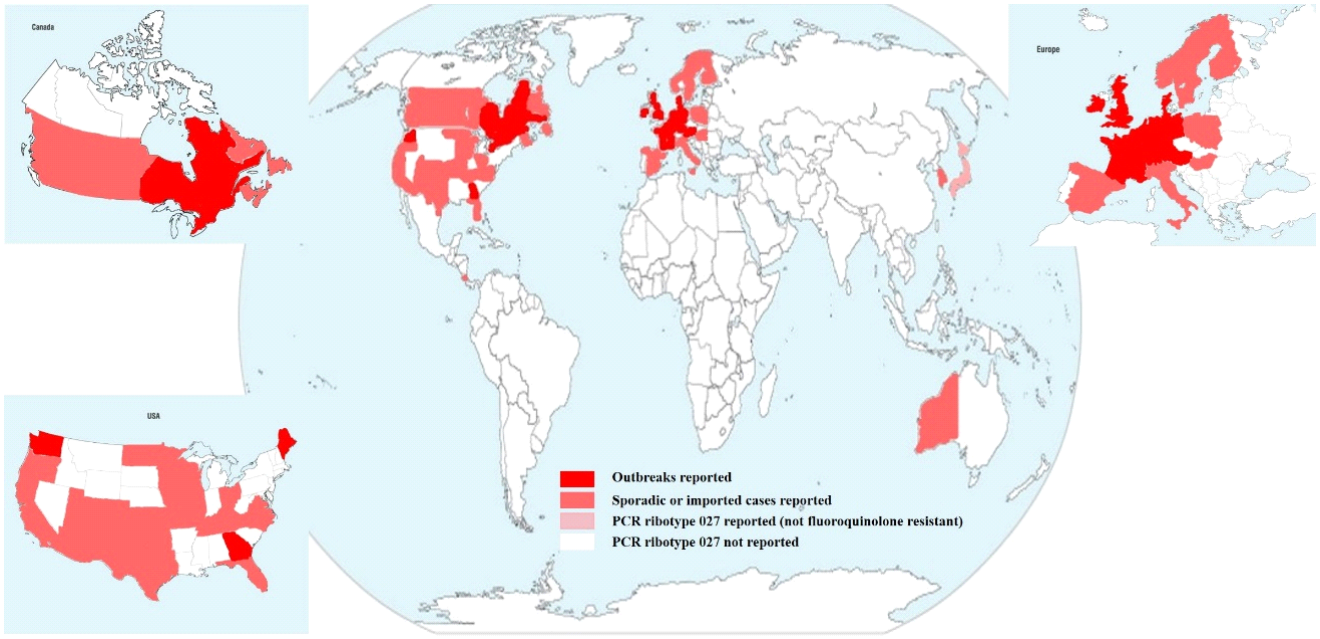


(Adapted from Clements et al., 2010)

Fig. 1. Worldwide incidence of *C. difficile* infection attributed to PCR ribotype 027. The rate of *C. difficile* infection acquisition has increased dramatically since 2000. This increase was associated with the newly discovered hypervirulent strain B1/NAP1/027 that produces a binary toxin associated with severe diarrhea.

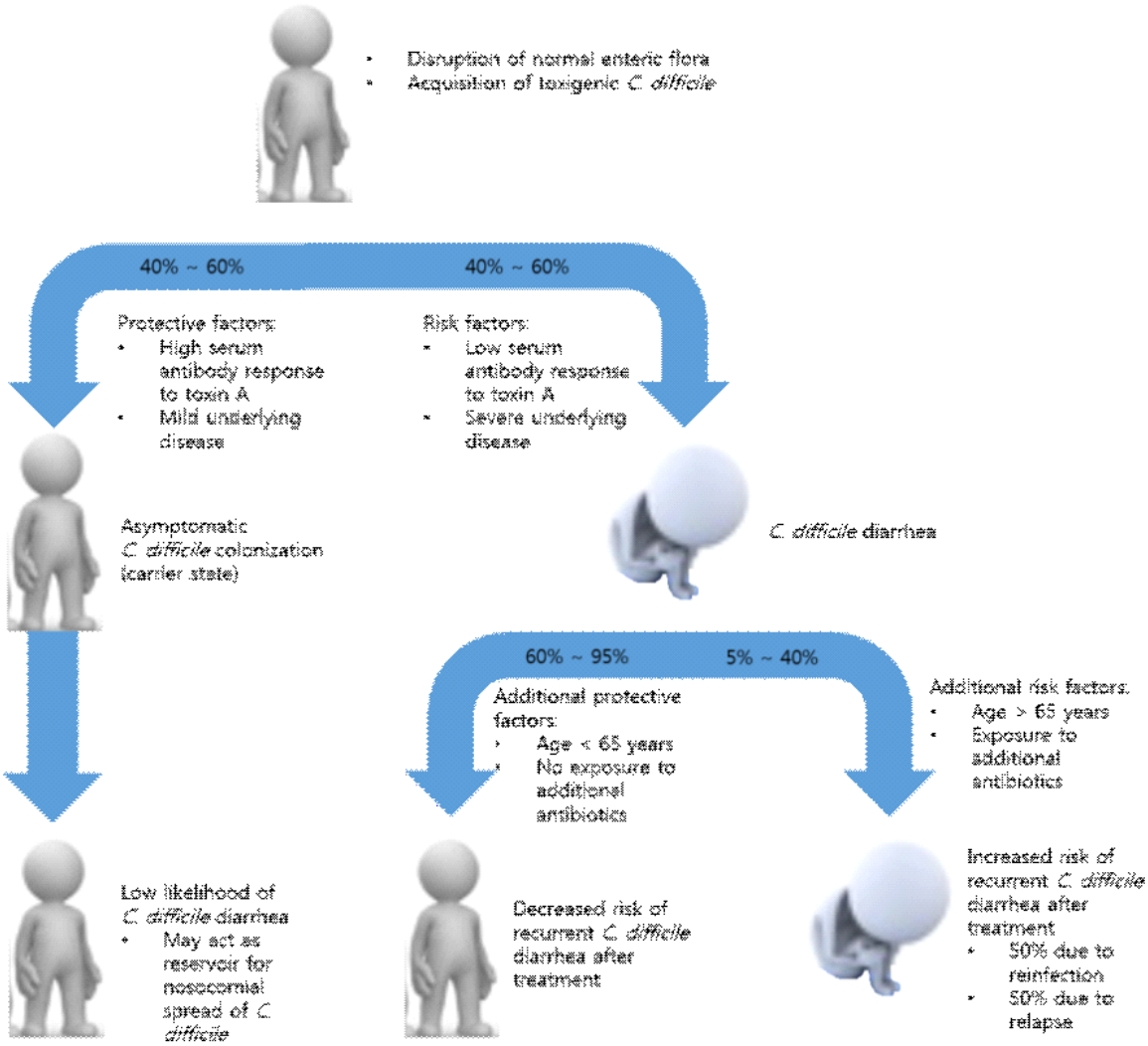
Asia during 2008–2010 have been attributed to PCR ribotype 027 (Fig. 1) (Clements *et al*., 2010). Furthermore, the number of fatal *C. difficile* infections in England recently rose from approximately 500 in 1999 to nearly 3400 in 2006 (Kelly and LaMont, 2008).

The prevalence of *C. difficile* colonization has been estimated to be 7–11%, 5–7%, and 2% in hospital inpatients, long-term care facilities, and ambulatory adults, respectively (Aronsson *et al*., 1985; Samore *et al*., 1994). The primary reservoirs of *C. difficile* include colonized patients and contaminated hospital environments (Clabots *et al*., 1992; Cohen *et al*., 2000; Titov *et al*., 2000; Fawley and Wilcox, 2001). The intestinal microbiota in a healthy adult generally does not permit *C. difficile* colonization, but resistance to colonization is lost if this microbial changes. Any factor involved in altering the intestinal microbiota therefore increases the risk of *C. difficile* colonization after exposure (Fig. 2) (Poutanen and Simor, 2004).

Probiotics against Gastrointestinal Infection

Probiotics are defined as live microorganisms, which when consumed in adequate amounts, confer a health benefit for the host (FAO/WHO, 2002). They have been best studied with respect to their effects on gastrointestinal health. Probiotics have an emerging role in the treatment of gastrointestinal infections and are also effective against inflammatory bowel diseases (Lenoir-Wijnkoop *et al*., 2007). Probiotics have been used for fermented foods, yogurt, and cheese since thousands of years. The most commonly used probiotics are lactobacilli and bifidobacteria, but other organisms are used as well. The reported health benefits of probiotics in human intervention trials include the prevention and reduction of acute diarrhea and allergy (Szajewska *et al*., 2001; Sazawal *et al*., 2006; Ouwehand, 2007), relief from inflammatory bowel disease (Ewaschuk and Dieleman, 2006; Limdi *et al*., 2006) and antibiotic-associated gastrointestinal symptoms (Lenoir-Wijnkoop *et al*., 2007; Guglielmetti *et al*., 2011), anti-inflammatory effect (Tedelind *et al*., 2007; Maslowski *et al*., 2009), reduction of potentially pathogenic bacteria (Savard *et al*., 2011), and immunomodulatory effects (Bahrami *et al*., 2011). Specifically, *L. rhamnosus* LGG shortens the duration of acute childhood diarrhea caused by rotavirus and other pathogens (Isolauri *et al*., 1991; Majamaa *et al*., 1995), while *Saccharomyces boulardii* has been shown to be beneficial for the treatment of acute diarrhea in children and adults.

Furthermore, several probiotics have been evaluated for



(Adapted from Poutanen and Simor, 2004)

Fig. 2. Major factors contributing to the development of *C. difficile* colonization and diarrhea. *C. difficile* diarrhea occurs after 3 events: 1) change to the normal fecal microbiota; 2) colonic colonization; 3) growth of the organism with production of toxins.

prevention of AAD. The mortality rate of hospitalized patients with AAD is very high, at approximately 25%. *L. rhamnosus* LGG, *S. boulardii*, and other probiotic mixtures were found to be affective against this diarrhea, with the former 2 organisms being the most effective (Hickson *et al.*, 2007; Doron *et al.*, 2008; Surawicz, 2008). The strains of probiotics beneficial in the case of diarrhea are listed in Table 1.

C. difficile-associated Intestinal Disease

C. difficile infection is a classic example of the opportunistic proliferation of an intestinal pathogen after the breakdown of colonization resistance owing to antibiotic administration. After antibiotic intake by animals and humans, subsequent changes to the intestinal microbiota allow for the colonization of *C. difficile* within the intestine. *C. difficile* then releases two protein exotoxins, toxin A and toxin B, that mediate the diarrhea and colitis symptoms (Fig. 3. Poutanen and Simor, 2004). Toxigenic *C. difficile* is the underlying cause in approximately 20–40% of AAD cases (Clabots *et al.*, 1992, Fekety and Shah 1993). In fact, this microorganism is the major cause of nosocomial diarrhea in the US, infecting 15–25% of adult hospitalized patients. *C. difficile* infection can have serious clinical consequences, particularly in the elderly and debilitated; these effects include pseudomembranous colitis, toxic megacolon, intestinal perforation, and death.

Table 1. Probiotics use for gastrointestinal disease

Clinical condition	Strains
Diarrhea	
Infectious, childhood treatment	*Saccharomyces boulardii*, *Lactobacillus rhamnosus* LGG, and *Lactobacillus reuteri* SD2112
Prevention of infection	*Saccharomyces boulardii* and *Lactobacillus rhamnosus* LGG
Prevention of AAD	*Saccharomyces boulardii*; *Lactobacillus rhamnosus* LGG; and a combination of *Lactobacillus casei* DN114 G01, *Lactobacillus bulgaricus*, and *Saccharomyces thermophiles*
Prevention of recurrent CDAD	*Saccharomyces boulardii*, *Lactobacillus rhamnosus* LGG, and bacteriotherapy
Prevention of CDAD	*Saccharomyces boulardii* and *Lactobacillus rhamnosus* LGG
Inflammatory Bowel Disease (IBD)	
Pouchitis	
Preventing and maintaining remission	VSL#3
Induce remission	VSL#3
Ulcerative colitis	
Inducing remission	*Escherichia coli* Nissle, VSL#3
Maintenance	*Escherichia coli* Nissle, VSL#3
Crohn's	*Escherichia coli* Nissle, *Saccharomyces boulardii*, and *Lactobacillus rhamnosus* LGG
Irritable Bowel Syndrome (IBS)	*Bifidobacterium infantis* B5624, VSL#3, *Bifidobacterium animalis*, and *Lactobacillus plantarum* 299V

Adapted from Floch *et al*., 2011.

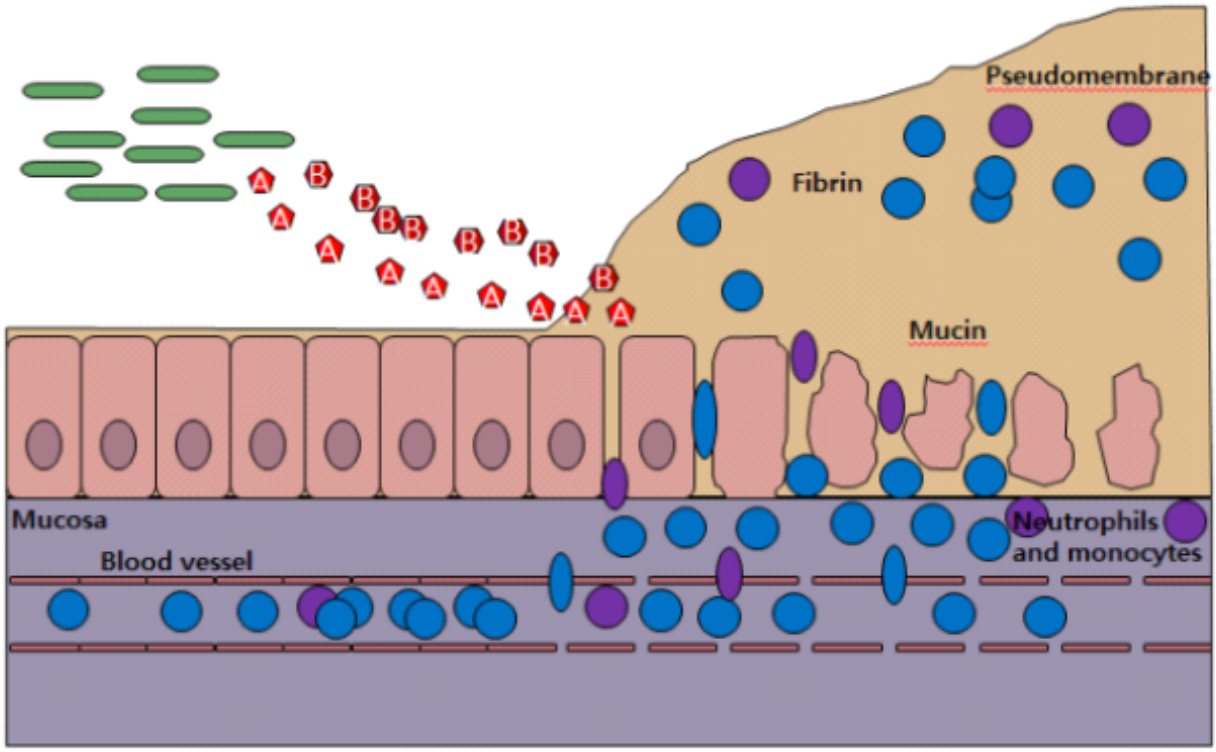


Fig. 3. Pathogenesis of *C. difficile*-associated intestinal disease. After antibiotic intake by animals and humans, subsequent changes to the intestinal microbiota allow for the colonization of *C. difficile* within the intestine. *C. difficile* then releases two protein exotoxins, toxin A and toxin B, that mediate the diarrhea and colitis symptoms.

The standard treatment for *C. difficile*-associated intestinal disease, involving either vancomycin or metronidazole, can be expensive and difficult. In addition, approximately 25% of patients experience relapse once treatment is discontinued (Bartlett *et al*., 1980; Fekety *et al*., 1989). Multiple relapses can occur and relapses can be more severe than the initial disease. The precise reason for relapse is currently unknown, but it is likely attributable to the survival of *C. difficile* spores within the intestinal tract during antibiotic treatment (Walters *et al*., 1983). Subsequently, when the therapy is completed, the spores germinate and produce toxin, especially because the treatment prevents the normal flora from reestablishing itself. To date, no effective therapy has been found for preventing *C. difficile* recurrences in intractable patients.

An attractive option for restoring intestinal homeostasis after antibiotic therapy is to use probiotics. Patients at risk for *C. difficile* intestinal disease can be identified by the fact that if they had a previous relapse of *C. difficile* infection they are more likely to have another relapse. Some preventative treatments against reoccurring *C. difficile*-associated intestinal disease have been recently evaluated. Rectal administration of feces from healthy adults has been examined in a very limited number of uncontrolled studies (Bowden *et al*., 1981; Schwan *et al*., 1984), and although it appears to be somewhat successful, there is obvious concern about the use of a complex, mixed, and undefined flora that could contain potential pathogens. Additional uncontrolled studies have investigated the rectal infusion of 10 different aerobic and anaerobic bacteria, as well as the use of a non-toxigenic strain of *C. difficile* (Borriello 1988; Tvede and RaskMadsen, 1989). Presumably, these bacteria occupy niches that the toxigenic strain would otherwise find available.

S. boulardii has demonstrated the most promise for use against *C. difficile*-associated intestinal disease. In a placebo controlled study, McFarland *et al*. (1994) examined clinical symptoms following standard antibiotic therapy (metronidazole or vancomycin) with concurrent *S. boulardii* or placebo administration in 124 adult patients, 64 of whom had an initial episode of *C. difficile* disease and 60 of whom had a history of *C. difficile* disease. The researchers found that in patients presenting with *C. difficile* disease for the first time, there was no significant difference in the likelihood of disease recurrence between the placebo and *S. boulardii* groups. However, in patients with prior *C. difficile* disease, *S. boulardii* administration significantly inhibited additional disease recurrences. The researchers concluded that the use of *S. boulardii* ingestion in combination with standard antibiotics is an effective and safe therapy for patients with recurrent *C. difficile* infections.

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