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REVIEW ARTICLE

Clinical impact of *Clostridium difficile* colonization

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Clostridium difficile can cause antibiotic-associated diarrhea in hospitalized patients. Asymptomatic colonization by *C. difficile* is common during the neonatal period and early infancy, ranging from 21% to 48%, and in childhood. The colonization rate of *C. difficile* in adult hospitalized patients shows geographic variation, ranging from 4.4% to 23.2%. Asymptomatic carriage in neonates caused no further disease in many studies, whereas adult patients colonized with toxigenic *C. difficile* were prone to the subsequent development of *C. difficile*-associated diarrhea (CDAD). However, the carriage of nontoxigenic *C. difficile* strains appears to prevent CDAD in hamsters and humans. Risk factors for *C. difficile* colonization include recent hospitalization, exposure to antimicrobial agents or gastric acid-suppressing drugs (such as proton-pump inhibitors and H2 blockers), a history of CDAD or cytomegalovirus infection, the presence of an underlying illness, receipt of immunosuppressants, the presence of antibodies against toxin B, and Toll-like receptor 4 polymorphisms. Asymptomatic *C. difficile* carriers are associated with significant skin and environmental contamination, similar to those with CDAD, and contact isolation and hand-washing practices should therefore be employed as infection control policies for the prevention of *C. difficile* spread. Treating patients

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with asymptomatic *C. difficile* colonization with metronidazole or vancomycin is not suggested by the currently available evidence. In conclusion, asymptomatic *C. difficile* colonization may lead to skin and environmental contamination by *C. difficile*, but more attention should be paid to the clinical impact of those with *C. difficile* colonization.

Introduction

Clostridium difficile is the leading cause of antibiotic-associated diarrhea in hospitalized patients through the production of toxins A and B and, most likely, a binary toxin. The clinical manifestations range from mild diarrhea to pseudomembranous colitis, toxic megacolon, and even death. The incidence of *C. difficile*-associated diarrhea (CDAD) is increasing worldwide and in Taiwan.[1] In the USA, the reported case number of CDAD in 2005 (84/100,000) was nearly three times that in 1996 (31/100,000). The identified risk factors for CDAD include advanced age,[2,3] previous hospitalization,[2,4] the use of feeding tubes,[5] antimicrobial exposure,[2,4,6] and the use of proton pump inhibitors (PPIs).[4] Avoidance of unnecessary antimicrobial agents or gastric acid-suppressing agents, such as PPIs, in addition to contact isolation and hand washing, have been regarded as important infection control policies to prevent the spread of *C. difficile* in hospitals.[7] In addition to patients with CDAD, those with *C. difficile* colonization (CdC) have been regarded as potential reservoirs of *C. difficile* and it is a general belief that the number of patients colonized with *C. difficile* outnumbers that of patients with CDAD.[8] The incidence of CdC could be as high as 23.2% among hospitalized patients,[9] particularly among vulnerable populations, such as patients with cystic fibrosis (32.4%)[10] (Table 1).[8–10,16,18–22,25,26,30,35,37,40–42,45–48]

C. difficile isolates that are capable of producing toxins A and B are regarded as toxigenic; otherwise, they are considered nontoxigenic. Toxigenic *C. difficile* colonization (tCdC) has been described as an independent risk factor for the subsequent development of CDAD,[8,9] and nontoxigenic *C. difficile* has been used to treat relapsing CDAD.[11] However, the clinical significance of tCdC or nontoxigenic CdC (ntCdC) remains controversial, warranting more attention from clinicians, infection control staff, epidemiologists, and microbiologists. In this review, we aim to elucidate the epidemiology, clinical impact, risk factors, and infection control concerns of individuals with tCdC or ntCdC.

Epidemiology of *C. difficile* colonization

During the neonatal period and early infancy, asymptomatic colonization by *C. difficile* is common, ranging from 21% to 48% in some reports, particularly among those with prolonged hospitalization, low birth weight (<2500 g), or younger gestational age (<37 weeks) as well as those nursing in an incubator or delivered by cesarean birth (Table 2).[12,14,15,49,50] There was geographic variation in *C. difficile* colonization rates among infants, with 35% observed among Swedish infants compared to 4% in Estonian infants.[12] Infant susceptibility to *C. difficile* colonization may be due to the inability of their intestinal microbiota to resist CdC. Rousseau et al[13] have demonstrated that the presence of *C. difficile* in the gut of infants is associated with changes in the microbiota composition. CdC has also been noted during childhood, particularly among those with malignancy (19%) or inflammatory bowel disease (17%).[14,15]

Likewise, the colonization rates of *C. difficile* in adult hospitalized patients vary geographically. In Canada, as few as 4.4% of hospitalized patients had CdC at admission.[16] The prevalence rate of CdC has been reported as 4.4–3.3% in France, 7.9% in the UK, 2.1–18.4% in the USA, 14% in Israel, and 20–23.2% in Taiwan (Table 1). However, the rates cannot be compared between these studies, as their study designs (prospective or retrospective), detection methods (culture, cytotoxin assay, or polymerase chain reaction), target populations, and/or the inclusion of ntCdC varied. In the study by Lee et al,[17] changing incidence and clinical manifestations of CDAD were noted when introduction of the combination of glutamate dehydrogenase and toxin assay in Northern Taiwan.

Because *C. difficile* is often acquired from a nosocomial environment, additional cases of CdC have been discovered during hospitalization, with the figures of CdC higher than those at admission or at initial screening. For example, the prevalence rate of CdC was 2.1% (11/517) at initial screening and 50% (64/128) during follow-up for more than 1 month in a study conducted by Clabots et al.[18] In our prospective study, we found a CdC prevalence rate of 20.0% among hospitalized patients at an initial screening, with an additional 25.4% of the patients developing CdC during follow-up.[8]

Many reports have explored the prevalence rates of CdC among the elderly. In general, the elderly in nursing care units or long-term care facilities (LTCFs) had higher prevalence rates of CdC. Fecal CdC was detected in 4% of the elderly outside LTCFs in the UK[19] and in 0.6% of elderly individuals in Belgium.[20] Arvand et al[21] reported a typical finding that the prevalence of CdC was 4.6% (11/240) in nursing home residents versus 0.8% (2/249) in the elderly living outside LTCFs. However, such a figure may range from 10% in continuing care institutions[22] to 51% among LTCF residents.[23] With the increase in the elderly population worldwide, the potential clinical sources of *C. difficile* among the elderly, particularly those in LTCFs, require more clinical attention.

Other susceptible populations have been investigated in addition to the elderly, including individuals with cystic fibrosis, inflammatory bowel disease, or human immunodeficiency virus (HIV) infection. CdC has been noted in

Table 1 The prevalence rates of *Clostridium difficile* colonization (CdC) in adults. Only studies with >200 cases or in specific populations are listed

Population	Country	Study year	*C. difficile* detection	Toxin detection	Prevalence rate, %		Refs
					CdC	Toxigenic CdC	
	USA	1985–1986	Culture	Cytotoxin assay	6.8% (29/428) IS; 13.0% (52/399) FU	2.0% (8/399) FU	45
	UK	1986[c]	Culture	Cytotoxin assay	32.4% (12/37) cystic fibrosis; 7% (3/40) controls	24.3% (9/37) cystic fibrosis; 5% (2/40) controls	10
Nursing home residents	USA	1987–1988	Culture	Cytotoxin assay	2.1% (11/517) IS; 50% (64/128) FU	ND	18
Nursing home residents	Israel	1998	Culture	Cytotoxin assay	14% (37/271)	79% (15/19)	30
Long-term care facilities	USA	1991	Culture	Cytotoxin assay	7.1% (16/225)[a]	4% (9/225)[a]	37
Long-term care facilities	USA	1991	Culture	EIA	11% (43/406) within 72 h of admission	ND	40
Long-term care facilities	Canada	2006–2007	Culture	ELISA	4.4% (184/4143) IS; 3% (123/4143) FU	33.5% (59/176) IS; 36.9% (45/122) FU	16
Acute geriatric ward	Israel	1992	Culture	ND	2% (2/100) IS; 12.2% (12/98) FU	ND	46
Admission to an infectious disease ward	France	1993	Culture or cytotoxin B assay	Cytotoxin B assay	13.3% (32/240)[b]	7.1% (17/240)[b]	25
Inpatients	France	1993–1994	Culture	Cytotoxin assay	4.4% (3/68) non-HIV; 3.8% (2/52) HIV	0	26
Acute rehabilitation facility	USA	2006[c]	Culture	EIA	16.7% (9/54)	16.7% (9/54)	47
Cases of spinal cord injury	USA	2008	Culture	Cytotoxin assay	ND	12% (18/149)	42
Elderly outside long-term care facilities	UK	2009	Culture	ELISA	4% (6/149)	2% (3/149)[d]	19
Elderly outside long-term care facilities	USA	2009	PCR	PCR	ND	9.7% (31/320)	41
Elderly in continuing care institution	Ireland	2009[c]	Culture	PCR	10% (10/100)	7% (7/100)	22
Elderly	Belgium	2009	Culture	Cytotoxin assay	0.6% (2/336) IS, 0.3% (1/336) FU	0.6% (2/336) IS; 0.3% (1/336) FU	20
Elderly	German	2010–2011	Culture	ELISA and PCR	4.6% (11/240) NHR; 0.8% (2/249) outside LTCFs	4.2% (10/240) NHR; 0.4% (1/249) outside LTCF	21
Elderly	Ireland	2011[c]	Culture	EIA and PCR	1.6% (2/123) community, 9.5% (4/43) outpatient settings, 21% (32/151) hospitalized patients		48
Elderly	Taiwan	2011	Culture	Real-time PCR	23.2% (39/168)	9.5% (16/168) IS; 7.1% (12/168) FU	9
Elderly	Taiwan	2011–2012	Culture	PCR	20.0% (84/441) IS; 25.4% (112/441) FU	13.2% (58/441) IS; 17.7% (78/441) FU	8
Elderly	UK	2012	Culture	Whole genome sequencing	7.9% (18/227)	5.7% (13/227)	35

[a] Percentage of stool cultures.
[b] Percentage of admissions.
[c] Publication year because there was no study period mentioned.
[d] One sample contained both toxigenic and nontoxigenic strains.
EIA = enzyme immunoassay; ELISA = enzyme-linked immunosorbent assay; FU = follow-up; HIV = human immunodeficiency virus; IBD = inflammatory bowel disease; IS = initial screening; LTFC = long-term care facility; ND = no data; NHR = nursing home residents; PCR = polymerase chain reaction.

Table 2 The prevalence rates of *Clostridium difficile* colonization (CdC) in children. Only studies with >200 cases or in specific populations are listed

Population	Country	Study year	C. *difficile* detection	Toxin detection	Prevalence rate, %		Refs
					CdC	Toxigenic CdC	
Neonates	UK	1984[a]	Culture	Cytotoxin assay	47% (31/66) 1 wk-old infants; 30.7% (46/150) 1-mo-old infants	14.7% (22/150) 1-mo-old infants	49
Newborns	UK	1982–1983	Culture	Cytotoxin assay	21% (31/490)	0	50
Infants	Sweden	1997	Culture	ND	35% (10/29) Swedish infants, 4% (1/27) Estonian infants	ND	12
Pediatric cancer patients	Australia		Culture			19%	14
Children with IBD	USA	2007–2009	Culture	Cytotoxin assay and PCR	17% (12/72) IBD, 3% (2/62) controls	9.6% (25/261)	15

[a] Publication year because there was no study period mentioned.
CIE = counter immunoelectrophoresis; IBD = inflammatory bowel disease; ND = no data; PCR = polymerase chain reaction.

32.4% (12/37) of cystic fibrosis patients, compared to 7.5% (3/40) of control patients.[10] The greater extent of CdC may be related to younger age, more severe pulmonary disease, and more exposure to antimicrobial agents in cystic fibrosis patients,[24] in whom, for unknown reasons, *C. difficile* strains are often nontoxigenic (77% *vs.* 17%). Asymptomatic *C. difficile* carriage is significantly more prevalent among patients with inflammatory bowel disease than controls (17% vs. 3%, $p = 0.012$).[15]

It is surprising that there are few CdC data among HIV-infected patients. In a ward in which 74% of patients were HIV infected, Hutin et al[25] reported a CdC prevalence rate of 13.3% (32/240). In another study, the prevalence rate of CdC was 3.8% (2/52) among HIV-infected patients, which was lower than that in non–HIV-infected patients (4.4%, 3/68).[26] However, the prevalence rate of CdC in HIV-infected patients may have been underestimated, as both studies were conducted prior to 1998. By contrast, in a study in an HIV-infected cohort, the incidence of CDAD was 8.3 cases/1000 patient-years, which was twice that previously reported and increased independently of a CD4 cell count ≤50 cells/μL.[27] The prevalence rate and clinical impact of CdC in HIV-infected patients require further large-scale investigations.

tCdC and ntCdC: good or bad?

As described earlier, fecal carriage of *C. difficile*, either toxigenic or non-toxigenic, is common among neonates. Such a carrier state is well tolerated by infants, and the immunoglobulin G antitoxin response that develops during the carrier state appears to provide durable protection against subsequent *C. difficile* disease.[28] However, the clinical impact of CdC in adults remains unsettled. An early study suggested that CdC was associated with a low risk of CDAD[29] (Table 3).[8,9,23,29,30,40,42] However, other studies have reported that patients with CdC are more likely to develop CDAD.[8,9,30] In a recent study, diabetes mellitus and prior receipt of piperacillin–tazobactam or PPIs were found to be independent risk factors for the development of CDAD among hospitalized patients with tCDC.[31] These studies were diverse in study design, including the definition of diarrhea, toxigenic or nontoxigenic isolates, hospital settings,

Table 3 Clinical impact of *Clostridium difficile* colonization in adults

Country	Study year	Follow-up duration	Clinical impact	Refs
			Development of *C. difficile* infection	
USA	1983–1993	9 wk–20 mo	1.0% (2/192) CdC vs. 3.6% (22/618) no CdC	29
USA	1991	5 mo	47% (9/19) tCdC	40
Israel	1998	5 mo	51.3% (19/37) CdC at admission vs. 12.0% (28/234) no CdC ($p < 0.0001$)	30
Taiwan	2011	6 mo	17.9% (5/28) tCdC vs. 1.4% (2/140) no tCdC ($p = 0.002$)	9
Taiwan	2011–2012	18 mo	14.1% (11/79) tCdC vs. 0.9% (3/328) no CdC, 0% (0/34) ntCdC ($p < 0.001$)	8
			Contamination	
USA	2008	90 d	17% (3/18) CdC: skin and/or environmental contamination	42
USA	2006	3 mo	Carriers *vs.* noncarriers: skin (61% vs. 19%; $p = 0.001$) or environmental contamination (59% vs. 24%; $p = 0.004$)	23

CdC = *C. difficile* colonization; ntCdC = nontoxigenic CdC; tCdC = toxigenic CdC.

calendar years, follow-up periods, and antibiotic pressure, which may underlie the differences in their conclusions.

The ability of hosts to mount an immune response might be a factor determining whether a patient with CdC will develop CDAD. Mulligan et al[32] found that immunoglobulin (Ig) A and IgM concentrations were significantly higher in asymptomatic carriers than in symptomatic patients. Another study found that asymptomatic carriers had greater increases in serum levels of IgG against toxin A and in serum levels of IgM against non-toxin antigens than did patients with CDAD.[30] The variable capability to mount immune responses can, at least partially, explain the different clinical evolutions among individuals acquiring *C. difficile*. The hypothesis that serum immunoglobulins against composite antigens of *C. difficile* can be used as surrogate markers of CDAD development among *C. difficile* carriers warrants further clinical study.

As early as in 1987, a promising result of oral therapy with nontoxigenic *C. difficile* was reported for difficult-to-treat CDAD. Two patients with relapsing CDAD following metronidazole and vancomycin therapy were treated by a nontoxigenic *C. difficile* strain given orally, and both responded well without side effects.[11] However, the role of nontoxigenic *C. difficile* as a therapy is more controversial than its preventive use. Nontoxigenic *C. difficile* has been examined for the prevention of the establishment of toxigenic *C. difficile* in the intestinal tract in animal models, including hamsters. Pretreatment with nontoxigenic *C. difficile* protected hamsters from subsequent lethal challenge with toxigenic *C. difficile*.[33] Notably, an epidemiological survey revealed that the incidence of CDAD was low in individuals with ntCdC (0/34, 0%) and in those without colonization (3/328, 0.9%).[8] It is plausible that precolonization of the intestinal tract with nontoxigenic *C. difficile* can exclude toxigenic CdC by outcompeting the toxigenic *C. difficile* for a limiting nutrient. The ability of nontoxigenic *C. difficile* administration to prevent CDAD in animal models has prompted human clinical trials, which are in the early stages. Among these, an oral suspension of VP20621, spores of a nontoxigenic *C. difficile* strain (M3), are evaluated for its ability to protect against the colonization of human guts by toxigenic strains, and have been shown to be in good tolerability in healthy adults.[34]

Risk factors of *C. difficile* colonization

Because *C. difficile* is a nosocomial pathogen, it is not surprising that previous hospitalization is a risk factor for CdC, as revealed by several studies (Table 4).[8,9,15,16,18,23,25,35–37,40,41,45,50] For example, Loo et al[16] reported that patients with recent hospitalization, i.e., in the previous 2 months, were more likely to have CdC. Similarly, Eyre et al[35] found that CdC at admission was correlated with a hospital stay within the previous 6 months. Because there was an increased risk of CdC in hospitals, prolonged hospitalization is a likely risk factor for CdC.[36]

Previous exposure to cephalosporins,[8,37] penicillins,[25] clindamycin,[25] co-trimoxazole,[37] or fluoroquinolones[23] has been linked to CdC. However, the effects of cephalosporins on the development of CdC are diverse and dependent on the cephalosporin generation. We previously reported that prior prescription of cefepime, a fourth-generation cephalosporin, was associated with tCdC, whereas cefuroxime, a second-generation cephalosporin, was associated with ntCdC.[8]

The use of gastric acid-suppressing drugs, including antacids,[19] PPIs,[15,16] and H2 blockers,[16] is a risk factor CDAD in addition to nosocomial CdC because vegetative *C. difficile* can survive exposure to gastric contents with reduced acidity.[38] In addition, PPIs decrease extracellular and intracellular reactive oxygen production and the bactericidal activity of neutrophils,[39] which mediate the host defense against *C. difficile*.

Additional host variables related to CdC have been identified, including a history of CDAD[40] or cytomegalovirus infection,[40] hemodialysis,[41] the receipt of immunosuppressants[35] or corticosteroids,[35] renal insufficiency,[40] admission to vascular surgery services,[40] and liver transplantation.[40] Of interest, the presence of antibodies against toxin B has been associated with CdC.[16] However, many variables have not been consistently related to CdC in the literature. In our previous work, patients with the TLR4 rs1927914 polymorphism (GG genotype) had a higher risk of CdC.[8] These risk factors suggest that CdC is associated with the host's immune status.

Management of *C. difficile* colonization

In addition to CDAD, patients with CdC have been regarded as a reservoir of *C. difficile* contamination. In a study by Riggs et al,[23] 51% of asymptomatic patients carried toxigenic *C. difficile*, of which 37% belonged to the epidemic strain. In the same study, compared with noncarriers, asymptomatic carriers had higher percentages of skin (61% vs. 19%; $p = 0.001$) and environmental contamination (59% vs. 24%; $p = 0.004$). Spores on the carriers' skin were easily transferred to their hands. Importantly, 87% of the isolates from the skin and 58% of isolates from the environment were matched to concurrent isolates in the stool.[23] By comparison, in CDAD patients in the Cleveland VA Medical Center (Cleveland, Ohio, USA), the prevalence of skin and/or environmental contamination was lower among asymptomatic carriers (3/18, 17% vs. 5/6, 83%; $p = 0.007$), but the carriers outnumbered CDAD patients by a factor of 3:1. This finding underlines an important message: asymptomatic carriers have great potential to contribute to *C. difficile* transmission in hospitals.[42] Additionally, nosocomial acquisition of a *C. difficile* strain has been reported to be preceded by a documented introduction into the ward by another asymptomatic ward admission in 16 (84%) of 19 instances, suggesting that *C. difficile*-colonized new admissions are a major source of nosocomial *C. difficile* infections.[18] Thus, CdC may be a source for the nosocomial spread of *C. difficile*, but the detailed pathways by which it spreads should be carefully explored when designing infection control policies for CdC. In fact, there are voices in favor of *C. difficile* screening coupled with isolation precautions in terms of economic dominance and health benefits in hospitals.

Infection control for *C. difficile* colonization

Because environmental contamination is important for *C. difficile* spread from carriers, contact isolation and hand

Table 4 Risk factors for *Clostridium difficile* colonization

Population	Country	Identified risk factors (by univariate analysis or multivariate analysis*)				Refs
		Recent hospitalization	Antimicrobial exposure	Gastric acid-suppressing agents	Others	
Adults	USA	Recent hospitalization (OR: 3.1)			Prior CDAD (OR: 9.5), renal insufficiency (OR: 6.7); admission to vascular surgery service (RR: 2.3); liver transplantation (RR: 4.2)	40
	USA	Hospitalized within 30 d (16% vs. 7%, $p < 0.001$)				18
	USA	Recent hospitalization (OR: 2.45)*			Chronic dialysis (OR: 8.12)*, corticosteroid use (OR: 3.09)*	41
	Canada	Hospitalization in previous 2 mo (OR: 2.18)		Proton-pump inhibitors (OR: 1.71); H2 blockers (OR: 2.14)	Use of chemotherapy (OR: 2.37), presence of antibodies against toxin B (OR: 1.75)	16
	UK	Hospital stay within 6 mo (OR: 5.5)			Steroids or other immunosuppressant within 6 months (OR: 7.2)	35
Admission to an ID ward	France		Clindamycin (OR: 9.4)*; penicillin (OR: 3.9)*		History of cytomegalovirus infection (OR: 4.2)*	25
Long-term care facilities	USA		Cephalosporin (RR: 4.66)*; co-trimoxazole (RR: 8.45)*	Histamine-2 antagonist use (RR: 3.27)*		37
	USA		Antibiotic within 3 mo (OR: 3.4)*		CDAD history (OR: 20.7)*	23
	Taiwan		>1 class of antibiotic (OR: 6.67)			9
	Taiwan		Cefepime (OR: 5.3)* associated with tCdC; cefuroxime (OR: 11.7)* and glycopeptide (OR: 10.9)* associated with ntCdC		TLR4 rs1927914 polymorphism (GG genotype; OR: 4.4)	8
	USA			Antacids (RR: 1.80)*	Stool softeners (RR: 2.04)*	45
Newborns	USA			PPI use higher in CdC (54% vs. 25%, $p < 0.05$)		15
	UK				Prolonged hospitalization, a lower birth weight (<2500 g), a younger gestational age (<37 wk), nursing in an incubator	50
	USA				Increased length of stay in the nursery ($p < 0.001$); delivery by cesarean birth ($p < 0.001$)	36

CDAD = *C. difficile*-associated diarrhea; CdC = *C. difficile* colonization; ID = infectious diseases; ntCdC = nontoxigenic CdC; OR = odds ratio; PPI = proton pump inhibitor; RR = relative risk; tCdC = toxigenic CdC.

washing are essential infection control measures.[7] *C. difficile* spores survive routine environmental cleaning with detergents and hand hygiene practices using alcohol-based gels.[43] Enhanced environmental cleaning with 10% sodium hypochlorite and hand washing with chlorhexidine or soap and water can reduce *C. difficile* burden, and the use of barrier precautions can attenuate *C. difficile* transmission.[43] More aggressively, metronidazole and vancomycin have been used individually to eradicate CdC, although with discrepant results. In a study involving 30 asymptomatic *C. difficile* carriers, fecal *C. difficile* was absent during and immediately after oral vancomycin therapy in nine of 10 patients, compared to three of 10 patients treated by oral metronidazole ($p = 0.02$) and two of 10 patients treated with placebo ($p = 0.005$). Nevertheless, eight of nine patients with transient clearance of fecal CdC following vancomycin treatment excreted *C. difficile* again within an average of 20 days after completing treatment.[44] Therefore, the investigators did not favor antimicrobial eradication of CdC. There is no adequate evidence to support metronidazole or vancomycin therapy in the routine management of fecal colonization by *C. difficile*.

Conclusion

In conclusion, asymptomatic fecal colonization by *C. difficile* can cause skin and environmental contamination, similar to that observed among patients with CDAD, and more attention should be given to the clinical impact of *C. difficile* carriers and infection control policies.

Conflicts of interest

All contributing authors declare no conflicts of interest.

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References

1. Chung CH, Wu CJ, Lee HC, et al. *Clostridium difficile* infection at a medical center in southern Taiwan: incidence, clinical features and prognosis. *J Microbiol Immunol Infect* 2010;**43**:119—25.
2. Vesteinsdottir I, Gudlaugsdottir S, Einarsdottir R, Kalaitzakis E, Sigurdardottir O, Bjornsson ES. Risk factors for *Clostridium difficile* toxin-positive diarrhea: a population-based prospective case-control study. *Eur J Clin Microbiol Infect Dis* 2012;**31**:2601—10.
3. Lai CC, Lin SH, Tan CK, Liao CH, Huang YT, Hsueh PR. Clinical manifestations of *Clostridium difficile* infection in a medical center in Taiwan. *J Microbiol Immunol Infect* 2013 Aug 23. pii: S1684-1182(13)00111-4.
4. Dial S, Delaney JA, Barkun AN, Suissa S. Use of gastric acid-suppressive agents and the risk of community-acquired *Clostridium difficile*-associated disease. *JAMA* 2005;**294**:2989—95.
5. O'Keefe SJ. Tube feeding, the microbiota, and *Clostridium difficile* infection. *World J Gastroenterol* 2010;**16**:139—42.
6. Cho SM, Lee JJ, Yoon HJ. Clinical risk factors for *Clostridium difficile*-associated diseases. *Braz J Infect Dis* 2012;**16**:256—61.
7. Cohen SH, Tang YJ, Muenzer J, Gumerlock PH, Silva Jr J. Isolation of various genotypes of *Clostridium difficile* from patients and the environment in an oncology ward. *Clin Infect Dis* 1997;**24**:889—93.
8. Hung YP, Lin HJ, Wu TC, Liu HC, Lee JC, Lee CI, et al. Risk factors of fecal toxigenic or non-toxigenic *Clostridium difficile* colonization: impact of Toll-like receptor polymorphisms and prior antibiotic exposure. *PLoS One* 2013;**8**:e69577.
9. Hung YP, Tsai PJ, Hung KH, Liu HC, Lee CI, Lin HJ, et al. Impact of toxigenic *Clostridium difficile* colonization and infection among hospitalized adults at a district hospital in southern Taiwan. *PloS one* 2012;**7**:e42415.
10. Peach SL, Borriello SP, Gaya H, Barclay FE, Welch AR. Asymptomatic carriage of *Clostridium difficile* in patients with cystic fibrosis. *J Clin Pathol* 1986;**39**:1013—8.
11. Seal D, Borriello SP, Barclay F, Welch A, Piper M, Bonnycastle M. Treatment of relapsing *Clostridium difficile* diarrhoea by administration of a nontoxigenic strain. *Eur J Clin Microbiol* 1987;**6**:51—3.
12. Naaber P, Klaus K, Sepp E, Björksten B, Mikelsaar M. Colonization of infants and hospitalized patients with *Clostridium difficile* and lactobacilli. *Clin Infect Dis* 1997;**25**(Suppl. 2):S189—90.
13. Rousseau C, Levenez F, Fouqueray C, Doré J, Collignon A, Lepage P. *Clostridium difficile* colonization in early infancy is accompanied by changes in intestinal microbiota composition. *J Clin Microbiol* 2011;**49**:858—65.
14. Burgner D, Siarakas S, Eagles G, McCarthy A, Bradbury R, Stevens M. A prospective study of *Clostridium difficile* infection and colonization in pediatric oncology patients. *Pediatr Infect Dis J* 1997;**16**:1131—4.
15. Hourigan SK, Chirumamilla SR, Ross T, Golub JE, Rabizadeh S, Saeed SA, et al. *Clostridium difficile* carriage and serum antitoxin responses in children with inflammatory bowel disease. *Inflamm Bowel Dis* 2013;**19**:2744—52.
16. Loo VG, Bourgault AM, Poirier L, Lamothe F, Michaud S, Turgeon N, et al. Host and pathogen factors for *Clostridium difficile* infection and colonization. *N Engl J Med* 2011;**365**:1693—703.
17. Lee YC, Wang JT, Chen AC, Sheng WH, Chang SC, Chen YC. Changing incidence and clinical manifestations of *Clostridium difficile*-associated diarrhea detected by combination of glutamate dehydrogenase and toxin assay in Northern Taiwan. *J Microbiol Immunol Infect* 2012;**45**:287—95.
18. Clabots CR, Johnson S, Olson MM, Peterson LR, Gerding DN. Acquisition of *Clostridium difficile* by hospitalized patients: evidence for colonized new admissions as a source of infection. *J Infect Dis* 1992;**166**:561—7.
19. Miyajima F, Roberts P, Swale A, Price V, Jones M, Horan M, et al. Characterisation and carriage ratio of *Clostridium difficile* strains isolated from a community-dwelling elderly population in the United Kingdom. *PLoS One* 2011;**6**:e22804.
20. Schoevaerdts D, Swine C, Verroken A, Huang TD, Glupczynski Y. Asymptomatic colonization by *Clostridium difficile* in older adults admitted to a geriatric unit: a prospective cohort study. *J Am Geriatr Soc* 2011;**59**:2179—81.
21. Arvand M, Moser V, Schwehn C, Bettge-Weller G, Hensgens MP, Kuijper EJ. High prevalence of *Clostridium difficile* colonization among nursing home residents in Hesse, Germany. *PLoS One* 2012;**7**:e30183.

22. Ryan J, Murphy C, Twomey C, Paul Ross R, Rea MC, MacSharry J, et al. Asymptomatic carriage of *Clostridium difficile* in an Irish continuing care institution for the elderly: prevalence and characteristics. *Ir J Med Sci* 2010;**179**:245—50.
23. Riggs MM, Sethi AK, Zabarsky TF, Eckstein EC, Jump RL, Donskey CJ. Asymptomatic carriers are a potential source for transmission of epidemic and nonepidemic *Clostridium difficile* strains among long-term care facility residents. *Clin Infect Dis* 2007;**45**:992—8.
24. Bauer MP, Farid A, Bakker M, Hoek RA, Kuijper EJ, van Dissel JT. Patients with cystic fibrosis have a high carriage rate of non-toxigenic *Clostridium difficile*. *Clin Microbiol Infect* 2013 Nov 1. http://dx.doi.org/10.1111/1469-0691.12439.
25. Hutin Y, Casin I, Lesprit P, Welker Y, Decazes JM, Lagrange P, et al. Prevalence of and risk factors for *Clostridium difficile* colonization at admission to an infectious diseases ward. *Clin Infect Dis* 1997;**24**:920—4.
26. Mainardi JL, Lacassin F, Guilloy Y, Goldstein FW, Leport C, Acar JF, et al. Low rate of *Clostridium difficile* colonization in ambulatory and hospitalized HIV-infected patients in a hospital unit: a prospective survey. *J Infect* 1998;**37**:108—11.
27. Haines CF, Moore RD, Bartlett JG, Sears CL, Cosgrove SE, Carroll K, et al. *Clostridium difficile* in a HIV-infected cohort: incidence, risk factors, and clinical outcomes. *AIDS* 2013;**27**:2799—807.
28. Jangi S, Lamont JT. Asymptomatic colonization by *Clostridium difficile* in infants: implications for disease in later life. *J Pediatr Gastroenterol Nutr* 2010;**51**:2—7.
29. Shim JK, Johnson S, Samore MH, Bliss DZ, Gerding DN. Primary symptomless colonisation by *Clostridium difficile* and decreased risk of subsequent diarrhoea. *Lancet* 1998;**351**:633—6.
30. Kyne L, Warny M, Qamar A, Kelly CP. Asymptomatic carriage of *Clostridium difficile* and serum levels of IgG antibody against toxin A. *N Engl J Med* 2000;**342**:390—7.
31. Lin HJ, Hung YP, Liu HC, et al. Risk factors for *Clostridium difficile*-associated diarrhea among hospitalized adults with fecal toxigenic *C. difficile colonization*. *J Microbiol Immunol Infect* 2013 Sep 21. pii: S1684-1182(13)00148-5.
32. Mulligan ME, Miller SD, McFarland LV, Fung HC, Kwok RY. Elevated levels of serum immunoglobulins in asymptomatic carriers of *Clostridium difficile*. *Clin Infect Dis* 1993;**16**(Suppl 4):S239—44.
33. Nagaro KJ, Phillips ST, Cheknis AK, Sambol SP, Zukowski WE, Johnson S, et al. Nontoxigenic *Clostridium difficile* protects hamsters against challenge with historic and epidemic strains of toxigenic BI/NAP1/027 *C. difficile*. *Antimicrob Agents Chemother* 2013;**57**:5266—70.
34. Villano SA, Seiberling M, Tatarowicz W, Monnot-Chase E, Gerding DN. Evaluation of an oral suspension of VP20621, spores of nontoxigenic *Clostridium difficile* strain M3, in healthy subjects. *Antimicrob Agents Chemother* 2012;**56**:5224—9.
35. Eyre DW, Griffiths D, Vaughan A, Golubchik T, Acharya M, O'Connor L, et al. Asymptomatic *Clostridium difficile* colonisation and onward transmission. *PloS one* 2013;**8**:e78445.
36. Bacon AE, Fekety R, Schaberg DR, Faix RG. Epidemiology of *Clostridium difficile* colonization in newborns: results using a bacteriophage and bacteriocin typing system. *J Infect Dis* 1988;**158**:349—54.
37. Walker KJ, Gilliland SS, Vance-Bryan K, Moody JA, Larsson AJ, Rotschafer JC, et al. *Clostridium difficile* colonization in residents of long-term care facilities: prevalence and risk factors. *J Am Geriatr Soc* 1993;**41**:940—6.
38. Jump RL, Pultz MJ, Donskey CJ. Vegetative *Clostridium difficile* survives in room air on moist surfaces and in gastric contents with reduced acidity: a potential mechanism to explain the association between proton pump inhibitors and *C. difficile*-associated diarrhea? *Antimicrob Agents Chemother* 2007;**51**:2883—7.
39. Zedtwitz-Liebenstein K, Wenisch C, Patruta S, Parschalk B, Daxbock F, Graninger W. Omeprazole treatment diminishes intra- and extracellular neutrophil reactive oxygen production and bactericidal activity. *Crit Care Med* 2002;**30**:1118—22.
40. Samore MH, DeGirolami PC, Tlucko A, Lichtenberg DA, Melvin ZA, Karchmer AW. *Clostridium difficile* colonization and diarrhea at a tertiary care hospital. *Clin Infect Dis* 1994;**18**:181—7.
41. Leekha S, Aronhalt KC, Sloan LM, Patel R, Orenstein R. Asymptomatic *Clostridium difficile* colonization in a tertiary care hospital: admission prevalence and risk factors. *Am J Infect Control* 2013;**41**:390—3.
42. Guerrero DM, Becker JC, Eckstein EC, Kundrapu S, Deshpande A, Sethi AK, et al. Asymptomatic carriage of toxigenic *Clostridium difficile* by hospitalized patients. *J Hosp Infect* 2013;**85**:155—8.
43. Gerding DN, Muto CA, Owens Jr RC. Measures to control and prevent *Clostridium difficile* infection. *Clin Infect Dis* 2008;**46**(Suppl 1):S43—9.
44. Johnson S, Homann SR, Bettin KM, Quick JN, Clabots CR, Peterson LR, et al. Treatment of asymptomatic *Clostridium difficile* carriers (fecal excretors) with vancomycin or metronidazole. A randomized, placebo-controlled trial. *Ann Intern Med* 1992;**117**:297—302.
45. McFarland LV, Surawicz CM, Stamm WE. Risk factors for *Clostridium difficile* carriage and *C. difficile*-associated diarrhea in a cohort of hospitalized patients. *J Infect Dis* 1990;**162**:678—84.
46. Rudensky B, Rosner S, Sonnenblick M, van Dijk Y, Shapira E, Isaacsohn M. The prevalence and nosocomial acquisition of *Clostridium difficile* in elderly hospitalized patients. *Postgrad Med J* 1993;**69**:45—7.
47. Marciniak C, Chen D, Stein AC, Semik PE. Prevalence of *Clostridium difficile* colonization at admission to rehabilitation. *Arch Phys Med Rehabil* 2006;**87**:1086—90.
48. Rea MC, O'Sullivan O, Shanahan F, O'Toole PW, Stanton C, Ross RP, et al. *Clostridium difficile* carriage in elderly subjects and associated changes in the intestinal microbiota. *J Clin Microbiol* 2012;**50**:867—75.
49. Bolton RP, Tait SK, Dear PR, Losowsky MS. Asymptomatic neonatal colonisation by *Clostridium difficile*. *Arch Dis Child* 1984;**59**:466—72.
50. Phua TJ, Rogers TR, Pallett AP. Prospective study of *Clostridium difficile* colonization and paracresol detection in the stools of babies on a special care unit. *J Hyg (Lond)* 1984;**93**:17—25.