Clostridium difficile associated diseases (CDAD), ranging from antibiotic associated diarrhea to pseudomembranous colitis, are serious nosocomial problems, with annual costs in Europe of up to 3 billion euros due to factors such as more intensive treatment and prolonged hospitalization (1, 2). In recent years, outbreaks of a new, highly virulent strain of C. difficile have been reported, especially in North America, and have shown a 5- to 20-fold increase in incidence and a 3- to 5-fold increase in morbidity and mortality. Since 2003 the new strain has also been isolated in England, Belgium, the Netherlands, France, and Austria. To date there is no evidence that the pathogen has been introduced to Germany; an examination of approximately 900 isolates from the period 2000-06 did not show any evidence of the new strain (5). However, it is likely only a matter of time before the new strain is identified in this country as well.

In the present article, we discuss the requirements and necessary steps to prevent, diagnose, and treat the infection.

Bacteriology

The history of antibiotic associated colitis began in the 1970s, when the condition was observed, in particular, in surgical patients receiving clindamycin (4). In 1977, C. difficile was identified as one of the most important causes of antibiotic associated colitis (5).

C. difficile is an obligately anaerobic, gram-positive, spore-forming, rod-shaped bacterium. The pathogenicity locus of C. difficile contains 5 genes that code for the major virulence factors of the bacillus. An analysis of the pathogen's genome sequence showed that approximately 11% of the information contained therein consists of mobile genetic elements, facilitating changes related to pathogenicity and host adaptation (6). The expression of an enterotoxin (toxin A) and a cytotoxin (toxin B) is modulated by a positive (TcdR) and negative (TcdC) regulator. The isolates that caused the severe C. difficile infections seen in

SUMMARY

Introduction: C. difficile associated diseases (CDAD) are an emerging problem, especially in the management of severely ill and elderly patients. Methods: Medline search up to January 2007, and abstracts of conferences on this topic attended by the authors. Results: A new highly virulent C. difficile strain detected in North America and Europe is leading to increased infectivity, morbidity, and mortality. This nosocomial infection is contributing to a substantial rise in health-care costs. The dissemination of recent information on the diagnosis, prevention, and treatment of CDAD is therefore a priority. The molecular methods required for detecting the highly virulent C. difficile strain are not generally available in Germany. Discussion: Infection control policies must be applied more stringently than has previously been the case, if these bacteria are to be prevented from spreading through hospital units. More restrictive use of antibiotics will lead to a decreased incidence of CDAD. The standard treatment of CDAD is metronidazole, with vancomycin as a fallback option. New agents such as toxin binders are currently under investigation in multicenter clinical studies.

Key words: Clostridium difficile associated diarrhea, hypervirulent strain, treatment, prevention, epidemiology
Canada, the United States, Great Britain, France, Belgium, and the Netherlands exhibited partial deletions in the gene encoding for TcdC. Classified as ribotype 027, toxinotype III, and PFGE NAP1, the pathogen also carries genes for an additional, so-called binary toxin (7, 8, 9). The increased virulence of the strain is thought to be caused by increased toxin production demonstrated in vitro and resulting from the 18-bp deletion in tcdC.

**Epidemiology**

At the time of hospital admission, between 3% and 7% of patients are carriers of C. difficile. Depending on risk factors such as length of hospital stay, patient age, severity of underlying disease, and use of antibiotics, this number rises to between 16% and 35% (7, 8, 9, 10). Although the majority of patients who are carriers at the time of hospital admission remain asymptomatic, between 15% and 71% of those who acquire the infection during their hospital stay develop symptomatic C. difficile associated diarrhea (10). The pathogen can be isolated from at least 20% to 30% of patients with antibiotic associated diarrhea, from 50% to 70% of patients with antibiotic associated colitis, and from over 90% of patients with pseudomembranous colitis (7, 8).

Mortality attributable to CDAD ranges from 1% to 2%, but increases in the case of pseudomembranous colitis to between 6% and 30% (7, 11). It has been estimated that CDAD patients stay in the hospital up to three weeks longer than patients without CDAD (2). Managing CDAD cases in the clinical setting includes expensive hygienic measures such as isolating patients, decontaminating hospital rooms, and even closing entire wards. Currently, the health care costs associated with C. difficile total approximately 3 billion euros per year in Europe and approximately 1.1 billion dollars in the US (1, 2). Due to the growing proportion of older people in the population, the use of more intensive treatment methods, and the appearance of more aggressive pathogens, an increase in these costs appears very likely.

In recent years, various countries have reported a sharp rise in the number of CDAD cases. In the US, the number of patients with this diagnosis on discharge nearly doubled between 1996 and 2003, increasing from 31 to 61 per 100,000 population (12). This is attributable, in part, to the increasing spread of the new, highly virulent strain of C. difficile (12, 13). The US Centers for Disease Control and Prevention have also reported a rise in the incidence of CDAD in ambulatory populations (14).

Data on the incidence or prevalence of C. difficile infections in Germany are scarce. A recent study revealed that the pathogen could be detected in 12% of stool samples from
hospitalized patients with antibiotic associated diarrhea (15). In a recently completed pilot study in high-risk hematological patients, we found C. difficile in 11.2% of stool samples. To date, the highly virulent strain has not been identified in Germany (box 1).

Clinical manifestation
The typical clinical features of CDAD include acute watery diarrhea with crampy lower abdominal pain, fever, leukocytosis, and the presence of fecal leukocytes. Abdominal symptoms usually appear between 5 and 10 days after antibiotic treatment has been initiated, but in rare cases may develop on the second day, or several months after cessation, of treatment. The association between nosocomial CDAD and antibiotic therapy has been well established (7, 8). In contrast, a large percentage of patients who acquire CDAD in the ambulatory setting have no history of antibiotic exposure (7). Although rare, chemotherapy induced CDAD has been reported and may be attributable to the antibiotic effect of the cytostatic agents used or to their cytotoxic effect accompanied by a decrease in local immune response. The clinical spectrum of CDAD ranges from mild diarrhea without inflammatory mucosal damage, to different grades of colitis including in 10% to 20% of cases the formation of characteristic pseudomembranes (figure 1) with typical histological changes (figure 2), to fulminant colitis. Segmental inflammatory wall thickening can be demonstrated on CT in 50% of cases. Most frequently, the rectum and sigmoid colon are affected (figure 3). CT findings do not correlate with clinical parameters and cannot predict the need for surgical treatment (16).

The stools are of loose to watery consistency and are rarely bloody. Depending on severity, CDAD can lead to dehydration, electrolyte imbalance, or hypoproteinaemia. Severe complications with high mortality rates include toxic megacolon and intestinal perforation, the latter of which frequently leads to sepsis. A special clinical presentation is right-sided colitis with mild or even no diarrhea, accompanied by lower abdominal pain and local rebound tenderness. Although C. difficile has been identified as the most frequent cause of pathogen induced antibiotic associated colitis, it should be noted that diarrhea in most
patients who receive antibiotics is due to a disruption of bacterial flora and is not of infectious origin (box 2).

**Diagnosis**

If CDAD is suspected, the presence of the characteristic pseudomembranes on endoscopy (figure 1) can lead to rapid confirmation of the diagnosis. In most cases, however, additional microbiological testing is necessary. These tests can detect toxins and, in certain cases, the pathogen itself (or its antigens) in a fresh stool specimen. Because infections can also be caused by toxin A-negative/toxin B-positive isolates, an ELISA that detects both toxins (A and B) is recommended. The sensitivity (38% to 94%) and specificity (92% to 98%) of the test are influenced by the commercial assay used, the patient population, transport times for the stool specimens (i.e. due to possible toxin degradation during transport), and the particular C. difficile isolates. At the same time, a fecal culture should be performed. Negative findings do not rule out CDAD, and up to two repeat tests should be performed.
using new stool specimens collected on different days; if findings are positive, further tests are not indicated. Another approach is the stool cytotoxin assay, which can detect toxin B with very high sensitivity (94% to 100%) and specificity (99%). However, the test takes 1 to 3 days for results and requires cell culture facilities.

C. difficile can be cultured on selective media and generally needs to be incubated in an anaerobic environment for approximately 48 hours. Because the presence of C. difficile in culture does not, by itself, allow us to distinguish between toxigenic and non-toxigenic strains, cultures that grow C. difficile should be tested for toxins. In addition to markedly improved sensitivity, the advantage of culture lies in the ability to perform antibiotic sensitivity testing and to characterize the pathogen with serotyping and/or molecular methods of analysis (PCR, ribotyping). These are the only methods that can identify the new, highly virulent strain of C. difficile.

The pathogen can be identified in feces within 60 minutes using an ELISA to detect the enzyme glutamate dehydrogenase (GDH). Thanks to its negative predictive value of approximately 99%, the GDH test is well suited to rule out the presence of C. difficile in stool specimens (box 3).

**Treatment**

**General measures**

If the clinical situation permits, the first and most important step after a diagnosis of CDAD is to discontinue the precipitating antibiotic. The goal of supportive therapy should be to restore fluid and electrolyte balance. Antimotility agents such as loperamide are contraindicated. Asymptomatic carriers or patients with mild symptoms do not require specific therapy. Treatment with anti-C. difficile antibiotics is indicated for patients with symptoms of colitis (fever, leukocytosis, and, if applicable, endoscopic findings), severe diarrhea, persistent diarrhea after discontinuation of the offending antibiotic, or the need to continue antibiotic therapy.

**Antibiotic therapy**

Despite the frequency of the disease and its considerable socioeconomic impact, relatively few treatment studies have been conducted to date. In a recent meta-analysis, no differences in clinical response were observed between metronidazole, bacitracin, fusidic acid, or vancomycin (evidence level I–II) (17). One small study showed that teicoplanin was slightly superior in terms of eliminating C. difficile in stool (7).

To prevent the spread of vancomycin resistant enterococci and to control costs (daily costs of metronidazole are 2.50 euros versus 75 euros for vancomycin), metronidazole has been established as standard therapy for CDAD since the mid-1990s (18) (table 1). Most patients who respond to therapy experience rapid defervescence and resolution of diarrhea in 4 to 5 days.
Follow-up testing of stool for the presence of C. difficile or its toxins does not generally influence the length of therapy. When oral therapy is not possible, metronidazole can be administered intravenously, but vancomycin cannot. For the treatment of severely ill patients with (sub)ileus, experts currently recommend combination therapy with vancomycin (up to 4 x 500 mg) administered via gastric tube plus intravenous metronidazole (evidence level V) (7). Recent observational studies in North America point to an increase in the number of complicated cases among patients on

**TABLE 1**

**CDAD treatment**

<table>
<thead>
<tr>
<th>CDAD</th>
<th>1st recurrence</th>
<th>2nd and later recurrences</th>
<th>Very severe cases (intestinal perforation, peritonitis, toxic megacolon)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral (i.v.) metronidazole (4 x 250 mg or 3 x 500 mg) for 10 days*</td>
<td>Oral (i.v.) metronidazole (4 x 250 mg or 3 x 500 mg) for 10 days*</td>
<td>Oral metronidazole or oral vancomycin 14–21 days + Saccharomyces boulardii</td>
<td>Metronidazole (possibly i.v.) + vancomycin via gastric tube; consider (total) colectomy</td>
</tr>
</tbody>
</table>

* In case of intolerance, pregnancy, breast-feeding, or in children: oral vancomycin (4 x 125 mg) for 10 days; intravenous vancomycin is ineffective in CDAD.

**TABLE 2**

**Overview of hygienic measures in cases of C. difficile infection**

(based on recommendations by the Robert Koch Institute for managing patients with C. difficile associated diarrhea)

<table>
<thead>
<tr>
<th>Isolation and preventive measures</th>
<th>Isolation preventive measures</th>
</tr>
</thead>
<tbody>
<tr>
<td>Room arrangements</td>
<td>Single rooms with en-suite facilities are highly advisable for patients with severe symptoms or incontinence</td>
</tr>
<tr>
<td></td>
<td>Cohort isolation is possible if patients have same pathogen</td>
</tr>
<tr>
<td>Contact precautions</td>
<td>Ensure proper staff training</td>
</tr>
<tr>
<td></td>
<td>Use aprons and disposable gloves before close contact with patients or when handling infected bodily fluids</td>
</tr>
<tr>
<td></td>
<td>Meticulous hand hygiene (esp. hand washing) after direct patient contact is essential</td>
</tr>
<tr>
<td>Disinfection and cleaning of surfaces</td>
<td>Daily wipe cleaning and disinfection of environmental surfaces with which patients have come in (skin) contact</td>
</tr>
<tr>
<td></td>
<td>If necessary, larger areas should be cleaned and disinfected</td>
</tr>
<tr>
<td>Sterilization of medical equipment</td>
<td>Medical items such as stethoscopes and thermometers should be limited to individual patients</td>
</tr>
<tr>
<td></td>
<td>Transport used medical products in closed containers</td>
</tr>
<tr>
<td></td>
<td>When preparing medical devices, thermal sterilization methods are preferred</td>
</tr>
<tr>
<td></td>
<td>Dishes and eating utensils should be transported to the dishwasher in closed containers</td>
</tr>
<tr>
<td></td>
<td>Laundry should be washed using disinfection procedures</td>
</tr>
<tr>
<td></td>
<td>Beds and mattresses should be protected with covers that can be wiped with an appropriate surface disinfectant</td>
</tr>
<tr>
<td>Final disinfection</td>
<td>Disinfection of all surfaces</td>
</tr>
<tr>
<td>Waste disposal</td>
<td>Follow normal procedures for hospital waste</td>
</tr>
</tbody>
</table>
metronidazole therapy (19, 20). However, because these data are retrospective, metronidazole or vancomycin were not administered randomly; moreover, the CDAD epidemic characterized by the emergence of the new, more virulent strain occurred during the observation period. Thus, the findings of these studies are not definitive enough to change the recommendations described above.

Treatment of recurrent infection

A major problem in the treatment of CDAD is the significant risk of clinical relapse. This is seen in approximately 20% of patients treated with metronidazole or vancomycin, usually occurs within 3 to 21 days, and is caused in almost 50% of cases by a different strain (7). Risk factors for recurrent C. difficile infection include exposure to additional antibiotics after treatment, age > 65 years, severe underlying illness, hypoalbuminemia, prolonged hospitalization, stay in an intensive care unit, and bacterial factors that have yet to be characterized in detail. Because treatment of the first recurrence has the same probability of success as the initial episode, retreatment with metronidazole is advised. In the event of a second recurrence, 7 weeks of tapered and pulsed antibiotic therapy with vancomycin have been recommended (evidence level V) (21). In a randomized study, the probiotic agent Saccharomyces boulardii administered as an adjunct to antibiotic therapy was able to reduce the risk of relapse in patients with recurrent CDAD, but not in patients with initial CDAD (evidence level I) (22) (table 1).

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DIAGRAM

Management of Patients with Suspected CDAD, algorithm for diagnosis

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Abbreviations: CDAD = Clostridium difficile associated disease; AAD = antibiotic associated diarrhea; DD = differential diagnoses; CIID = chronic inflammatory intestinal disease (e.g. colitis)

* Endoscopy should be performed in patients with severe disease or unclear diagnosis;
** Culture improves the sensitivity of the toxin test and allows for bacteria typing and antibiotic sensitivity test;
*** C. perfringens and S. aureus are also potential causes of AAD; **** if discontinuation of the offending antibiotic is insufficient
Non-antibiotic approaches
The increasing number of problems caused by antibiotic use underscore the great importance of developing novel substances for treating CDAD. A monoclonal antibody directed at C. difficile toxin A is currently in phase II testing (23). An ongoing international, multicenter study is investigating the efficacy of a toxin binder (tolevamer) compared to metronidazole and vancomycin. In a first clinical study in patients with mild to moderate CDAD, tolevamer demonstrated comparable efficacy to orally administered vancomycin (evidence level I) (24). Although vancomycin was associated with a trend towards more rapid response, a lower recurrence rate was observed in patients on tolevamer (24). The ongoing, large-scale studies will need to demonstrate whether this finding can be confirmed, and if the trend towards hypokalemia observed in patients on tolevamer is clinically relevant.

Surgical treatment
In rare cases, CDAD can lead to intestinal perforation, peritonitis, and toxic megacolon. These complications are associated with increased mortality and may require surgery. In a small retrospective study, total colectomy was superior to other resective procedures. Total colectomy was associated with a lower mortality rate of 11% (1/9) compared to hemicolectomy, which was associated with a mortality rate of 100% (4/4) (25).

Prevention/hygienic measures
The pathogen can be acquired by the fecal-oral route, from contact with contaminated environmental surfaces, and from health care workers who have cared for a colonized or infected patient (10). Prevention consists of three steps: 1) antibiotic restriction/control of inappropriate antibiotic use (26), 2) contact isolation of affected patients, and 3) environmental decontamination.

Hygienic measures, which consist primarily of placing CDAD patients in contact isolation, can be viewed on the website of the Robert Koch Institute (www.rki.de) (table 2). Among the measures to be observed when caring for symptomatic patients is the use of gloves and, if applicable, an apron when invasive procedures are planned or when expecting to handle body fluids. The decision as to which patients should be placed in single rooms depends on patient compliance, severity of symptoms, and risk to other patients. For oncology patients, isolation in a single room is obligatory.

After each patient contact, hands should be washed to aid in the mechanical removal of microorganisms; this should be followed by hygienic hand disinfection. This procedure should also be performed every time that gloves are removed.

When cleaning and disinfecting surfaces, the mechanical action of wiping is even more important than the germicidal action of the disinfectant used because the spores are resistant to common environmental surface disinfectants. The preventive effect of sporicidal surface disinfectants has not been confirmed. The adjunctive isolation and preventive measures should be continued for 48 hours after symptoms of enteritis have resolved. In the event of an outbreak, it is essential to continue the isolation and preventive measures described above, consult with the hospital’s infection control specialist, and, if necessary, use sporicidal surface disinfectants (peracetic acid, sodium hypochlorite). If an epidemiological link is suspected, any increase in the frequency of C. difficile associated disease in hospitals or nursing homes must be reported to the appropriate public health authorities.

Conclusion for clinical practice
CDAD is becoming an increasing clinical challenge. In particular, the newly emerging, highly virulent strain of C. difficile is associated with increased morbidity and mortality. The infection is primarily nosocomial, and severely ill and elderly patients are those at the highest risk. Recommended treatment is with oral metronidazole. The duration of treatment is a clinical decision. In severely ill patients with intestinal perforation, peritonitis, or toxic megacolon, total colectomy should be considered. Finally, ensuring early diagnosis (diagram), observing hygienic procedures, and isolating CDAD patients can help prevent the spread of the pathogen in the hospital setting.

Conflict of interest statement
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Dedication
The authors dedicate this paper to Prof. Dr. Ernst-Otto Riecken, former director of the Medizinische Klinik I (Gastroenterology and Infectious Diseases), on his 75th birthday.

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REFERENCES
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