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AN ANALYSIS OF RISK FACTORS OF *CLOSTRIDIUM DIFFICILE* INFECTION IN PATIENTS HOSPITALIZED IN THE TEACHING HOSPITAL IN 2008

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ABSTRACT

The last decade saw an increase in the incidence of *Clostridium difficile* infections. Approximately 80% of these infections occur in hospitalized patients. There are ongoing studies on the increase in the incidence of infections with this microorganism.

Objective: The objective of this study was to assess the risk factors of *C. difficile* infections in patients hospitalized in the teaching hospital in Warsaw in 2008.

Material and methods: A case-control study was conducted to achieve this objective. The study group comprised 266 patients. Patients in the study group were assessed for the following factors: sex; age; place of residence; hospitalizations; the use of proton pump inhibitors (IPP), various antibiotics, probiotics; hospital stay conditions, and the patient's condition.

Results: The statistical analysis showed that out of the assessed risk factors, hospitalization within the 3 months preceding hospital admission (OR 5.02; P<0.003) and antibiotic therapy (OR 4.85; P<0.003) were associated with the highest risk of *C. difficile* infection. Hospital stay conditions, including a stay in a multi-bed room (OR 1.64; P=0.05) or in a room without an *en suite* toilet (OR 1.59; P=0.01), were also shown to play a role. The risk of a *C. difficile* infection was also increased in the case of "bed-ridden" patients requiring the assistance of hospital staff for their daily hygiene (OR 1.69; P=0.01).

Conclusions: This study demonstrated that hospitalization itself, including the frequency and conditions of hospital stay, as well as receiving antibiotic therapy were significant risk factors of *C. difficile* infections in patients hospitalized at the SP CSK. Therefore, our analysis showed that *C. difficile* infections are mostly nosocomial.

Key words*: Clostridium difficile, risk factors, nosocomial infections.*

INTRODUCTION

Clostridium difficile is an obligate anaerobic gram-positive spore-forming bacillus.(1) The spores are highly resistant to environmental factors, including high temperature, drying, low pH, alcohol, antibiotics, and disinfectants. *C. difficile* is a cosmopolitan microorganism found all over the world. It causes conditions with diarrhea in people and animals. In humans, *C. difficile* infection may take various clinical presentations including asymptomatic, diarrhea of various severity, or even fulminant colitis.(2) Hence, this syndrome is known as Clostridium Difficile Associated Disease or Clostridium Difficile Associated Diarrhea (CDAD) or Clostridium Difficile Infection (CDI).(2) Although the natural *C. difficile* reservoir has not been definitively identified, the sources as well as routes of infection for this microorganism have been determined. The source of infection for humans may include another human, either affected or a carrier, animal, contaminated food, or spore-contaminated objects and surfaces.(3,4) A *C. difficile* infection typically develops via the oral route i.e. either as a result of ingesting contaminated food or via the fecal-oral route. It has also been suggested that *C. difficile* spores may be transmitted via air and this way enter the oral cavity and subsequently the gastrointestinal tract, however, these observations require further evidence.(5) The first stage of infection is colonization of the human gastrointestinal tract, including mainly the large intestine (colon).(2,6) The sprouting

spores begin to compete for food with normal intestinal flora colonizing the colon, including: *Bacteroides* spp., *Clostridium* spp., *Escherichia coli*, *Klebsiella* spp., *Proteus* spp., *Enterococcus* spp. and other.

Not all strains of *C. difficile* that colonize the human gastrointestinal tract are pathogenic. Colonization requires a capacity for adhesion and chemotaxis but it is the A, B, and binary toxins that are responsible for pathogenic properties of the bacterium.(2,7) In recent years, the amount of toxin produced and an increased capacity for spore formation have been considered to influence the pathogenic properties and/or infectiousness.(2,4)

Features of the host, including susceptibility to infection and the immune response, as well as features of the microorganism, such as virulence, both play a role in contracting a *C. difficile* infection and in its course. Exposure duration, the size of infective dose, and the presence of risk factors are also important.

Risk factors of *C. difficile* infections as well as the development of CDAD/CDI have been extensively described in literature.(4,8,9) Moreover, these factors can be divided into modifiable, i.e. those that can be changed and non-modifiable factors, i.e. those that cannot be changed. The former include antibiotic therapy, hospitalization, proton pump inhibitor (PPI) use, H2-receptor-antagonist treatment, chemotherapy, immunotherapy, surgical procedures, endogastric tubes, parenteral nutrition. The factors of the latter group include age, sex, the underlying condition.(9) The presented classification is dictated by the appropriate preventive measures.

The objective of this study was to assess risk factors of *C. difficile* infections among patients hospitalized at the Independent Public Central Clinical Hospital (SP CSK) in 2008.

The following risk factors were evaluated: sex; age; place of residence; hospital stays; the use of PPIs, antibiotics, probiotics; conditions of hospitalization, and the patient's condition.

MATERIALS AND METHODS

We conducted a case-control study to assess the risk factors for *C. difficile* infection. The study group comprised 266 patients hospitalized at the SP CSK in 2008, who had developed diarrhea and were diagnosed with a *C. difficile* infection, i.e. a positive culture for *C. difficile* was obtained and/or A/B toxins were detected in the feces. The study group was composed of 146 women and 120 men, with the mean age for women and men 65.8.

The control group comprised 221 patients who were hospitalized at the SP CSK in 2008 and underwent diagnostic tests for *C. difficile* infection due to their diarrhea but *C. difficile* infections were excluded. The control group comprised 115 women and 106 men, with the mean age for men and women 65. The subjects were assigned to the control group based on the site where there had been hospitalized and randomly selected from the group of patients meeting the inclusion criteria.

The data was obtained from the Microbiology Laboratory, the hospital IT network, and the patients' records.

Statistical analysis

The association of the disease and the exposure factor was evaluated with the use of 2x2 tables calculating the odds ratio of an undesirable event and its 95% confidence interval and critical level. The factors with $P<0.05$ were considered statistically significant. The statistical analysis was conducted with the Wald test, for each analyzed risk factor separately. Moreover, the study was expanded to include multivariate methods. For this reason, a logistic regression method was used based on generalized linear models. In the first stage, a logistic regression model for 12 risk factors was used, and it helped identify significant factors from among all evaluated factors. In the second stage, the logistic regression model was adjusted for the 5 risk factors identified in the first stage that had demonstrated statistical significance at the level of $P<0.05$. Calculations were conducted with the use of R 2.12.1 (R Development Core Team (2008); R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org).

RESULTS

In 2008, 266 hospitalized patients were diagnosed with a *C. difficile* infection at the SP CSK. A total of 221 out of the 266 *C. difficile* infections were found to be nosocomial, which constituted 83% of all new cases; and the incidence rates in the Intensive Care Clinic, Internal Disease Clinics, and Surgical Clinics were 1.68, 0.8, and 0.5 per 1,000 person-days, respectively.

Table I presents the causes of hospitalization in patients diagnosed with a *C. difficile* infection.

This study evaluated the likelihood of developing a *C. difficile* infection depending on the exposure to risk factors. Table II presents the results of the analysis. The statistical analysis demonstrated that the greatest likelihood of contracting a *C. difficile* infection is associated with hospitalization during the 3 months preceding the current hospital admission (OR 5.02; $P<0.003$) and receiving antibiotic therapy (OR 4.85; $P<0.003$). The analysis also showed an increased risk of *C. difficile* infection depending on the conditions of hospitalization, including a stay in a multi-bed room (OR 1.64; $P=0.05$) and in a room without an *en suite* toilet (OR 1.59; $P=0.01$). The

Table I. Causes of hospitalization in patients diagnosed with a *C. difficile* infection

Underlying disease (ICD)	**%**
Diseases of the circulatory system (I00–I99)	22.8%
Malignant neoplasms of lymphoid, hematopoietic and related tissue (C81–C96)	15.6%
Diseases of the digestive system (K00–K93.8)	15%
Diseases of the genitourinary system (N00–N99)	12.5%
Diseases of the respiratory system (J00–J99) + tuberculosis	9.8%
Malignant neoplasms (C00–C80)	7.8%
Endocrine, nutritional and metabolic disorders (E00–E99 –without E36–46. E87.0)	7.3%
Diseases of the blood and blood-forming organs (D46.9–89.10)	2.6%
Sepsis (A40.1–A41.5) / septic shock	2%
Injuries (S00–S99)	1.4%
Diseases of the nervous system (G00–G99)	1.4%
Malnutrition (E40–46, E87.0)	0.9%
Benign neoplasms (D33.7, D46.2)	0.6%
Diseases of the musculoskeletal system and connective tissue (M06.9, M05.1)	0.3%

risk of *C. difficile* infection increased also in the case of "bed-ridden" patients requiring the help of hospital staff for their daily hygiene (OR 1.69; P=0.01).

Table II. Risk of *C. difficile* infection in individuals exposed to a risk factor

Risk factors	Wald test		Logistic regression for 12 variables P	Logistic regression for 5 variables P
	OR	P		
Sex /women	1.12	0.58	0.32	
Age ≥ 65	1.09	0.64	0.49	
Place of residence – Warsaw	1.16	0.45	0.34	
Hospitalization within 3 months prior to current admission	5.02	<0.003	<0.0001	<0.0001
PPI use*	1.26	0.22	0.3	
Antibiotic therapy*	4.85	<0.003	<0.0001	<0.0001
No use of probiotics*	0.82	0.47	0.6	
Manual bed pan washing and disinfection	1.09	0.68	0.003	0.002
"Bed-ridden" care-dependent patient	1.69	0.01	0.08	0.08
"Bed-ridden" care-dependent patient with manual bed pan washing and disinfection	1.85	0.12		
Hospitalization in a room with ≥3 beds	1.64	0.05	0.93	
Hospitalization in a room without a shower	1.42	0.16	0.73	
Hospitalization in a room without a toilet	1.59	0.01	0.05	0.007

* during hospital stay

DISCUSSION

The association between the patient's condition that caused the hospitalization and the risk of contracting a *C. difficile* infection has been evaluated by various authors. Immune disorders and gastrointestinal conditions, including inflammatory bowel disease, have been listed in most papers as risk factors for *C. difficile* infections.(10,11) However, the present paper demonstrated that the conditions most commonly diagnosed in patients hospitalized at the SP CSK who developed the infection were: cardiovascular diseases (22.8%), malignant neoplasms of the lymphatic and hematopoietic tissue (15.6%), gastrointestinal conditions (15%), genitourinary conditions (12.5%) and respiratory system conditions (9.8%). The results are mostly consistent with the results by other authors, nonetheless, this also seems to be associated with the hospital's profile and the range of offered services. The SP CSK specializes in the treatment of cardiovascular diseases and the treatment of these conditions is conducted in 3 clinics, including two internal medicine clinics and one surgical clinic. The hospital also has a Hematology Clinic, where patients with lymphatic and blood neoplasms receive treatment. In 2008, these 4 clinics provided treatment to 30% of all patients with *C. difficile* infection. Hence, the results achieved show that the risk of infection was associated with the hospitalization itself and, to a lesser extent, with the patient's underlying disease.

Sex. Some studies have emphasized a greater proportion of women in the group infected with *C. difficile,* although in none of them the difference was significant. In the analysis conducted in Finland between 1996 and 2004, women constituted 62%, and in studies conducted in 34 European countries women constituted 56% in the CDAD group.(12,13) The present study evaluated the relationship between sex and the risk of developing a *C. difficile* infection and revealed the proportion of women in the study group was shown to be 55%, and in the control group – 52%, (OR 1.12; P=0.58). Similar results were achieved with the use of a logistic regression model (P=0.32). Thus, this study showed no increase in the risk of *C. difficile* infection in women.

Age. Many studies demonstrated that the age ≥65 years was a risk factor of *C. difficile* infection.(13,14) Bauer et al. demonstrated a 3 times greater incidence and Lyytikainen et al. — a 6 greater incidence among people ≥65 years old in comparison to the under 65 age group.(21,22) This study did not demonstrate an increased risk of *C. difficile* infections in individuals ≥65 years old (OR 1.09; P=0.64). Similar results were achieved with the use of a logistic regression model (P=0.49). This may be explained by a relatively young age of patients hospitalized at the SP CSK, as the mean

age of all those hospitalized in 2008 was 58.6. The incidence in younger age groups may also be a result of the in-hospital presence of *C. difficile* strain 027/NAP1/BI that is more pathogenous than other *C. difficile* strains and, as a result, it may cause infections in patients with no risk factors, including those in younger age groups.

Place of residence. Since the SP CSK is a super specialty teaching hospital, where patients from all over Poland are treated, the relationship between the infection and the place of residence was evaluated based on locations either in, or outside of, Warsaw. The analyzed data showed no increase in the risk of *C. difficile* infection in patients from Warsaw (OR 1.16; P=0.4) in comparison to those from outside of Warsaw. This was consistent with the results obtained with the use of a logistic regression model (P=0.34). These results cannot be compared with those by other authors because such studies have not been conducted.

Antibiotic therapy. *C. difficile* bacilli are an established etiological factor of antibiotic-associated diarrheas. This was demonstrated by many authors in different studies (13,15,16,17) that showed a relationship between *C. difficile* infection and antibiotic therapy. The antibiotics described in the literature as particularly predisposing to *C. difficile* infections belong to three different classes, i.e. fluoroquinolones, amoxicillin/clavulanate, and cephalosporins, especially third-generation cephalosporins. A clear relationship between antibiotic therapy and the risk of *C. difficile* infection was demonstrated in this, and other studies. The risk of contracting a *C. difficile* infection was 5 times higher in patients who received antibiotics than patients who did not receive antibiotics (OR 4.85; P<0.003). Similar results were shown with the use of a logistic regression model (significance level P<0.0001).

Proton pump inhibitor (PPI) use. There is a lack of consensus as to the relationship between PPI use and the risk of *C. difficile* infection; namely, Howell et al. demonstrated a relationship between the use of PPIs and the risk of *C. difficile* infection (OR 3.6) whereas Henrich et al. demonstrated no such relationship (OR 0.88; P=0.83).(10,18) Nonetheless, there are a number of publications showing the existence of a relationship between PPI use and *C. difficile* infections, and demonstrating that the risk of a *C. difficile* infection is between 1.7 and 3.5 times higher in the PPI group.(19,20,21,22) Therefore, the present study aimed to compare the rate of PPI use in the *C. difficile*-infected and non-infected study groups. The results showed no increase in the risk of *C. difficile* infection in the PPI group (OR 1.26; P=0.22). Similar results were obtained with the use of a logistic regression model (P=0.3).

Probiotic use. There have been many publications on the therapeutic and preventive roles of various probiotic compositions in association with *C. difficile* infections. (23,24) Out of the probiotic strains, it is only *Saccharomyces boulardii* that has a standardized dose of 1 g/24 h. According to a meta-analysis, *Saccharomyces boulardii* at 1 g per day prevents antibiotic-associated diarrhea (incidence lowered from 17.2% to 6.7%).(40) The present study assessed the risk of *C. difficile* infection in a group who did not receive probiotics during hospitalization. No increase in the risk of contracting a *C. difficile* infection was demonstrated in the group receiving no probiotics during hospitalization versus the group receiving probiotics (OR 0.82; P=0.42). Similar results were obtained with the use of a logistic regression model (P=0.6).

Hospitalization. There have been a number of studies showing hospitalization to be one of the main risk factors of contracting a *C. difficile* infection due to the ease with which the infection spreads in a hospital setting. The risk of colonization was shown to increase with each week of hospital stay, with approximately 20% of patients colonized with *C. difficile* after 2 weeks, and up to 50% after 4 weeks.(5,9) Henrich et al. showed that not only there is a relationship between frequent hospitalizations and the infection but also that the rate of severe CDAD in patients hospitalized for the second time is 2 times higher.(10)

The present study demonstrated that in 83% out of 266 patients infected with *C. difficile* the infection was nosocomial and it met the pre-defined criteria for nosocomial infections.

The incidence rates in the Intensive Care Clinic, Internal Disease Clinics, and Surgical Clinics were 1.68, 0.8, and 0.5 per 1,000 person-days, respectively. These results are consistent with the results obtained by other investigators, indicating that approximately 80% of diagnosed *C. difficile* infections were associated with a stay in a medical facility, and the places with the highest rate of infections were Intensive Care Units.(12,13)

Moreover, the present study showed an increased risk of contracting a *C. difficile* infection in patients hospitalized multiple times. Thus, the risk of developing a *C. difficile* infection in patients hospitalized within 3 months prior to the index hospitalization (when they were diagnosed with the infection) was 5 times higher than in patients with no recent hospitalizations (OR 5.02; P<0.003). Similar results were obtained with the use of a logistic regression model (significance level P<0.0001). This should be construed as a result of colonization acquired during the subsequent hospitalizations. These results are consistent with those by other authors who conducted such an evaluation.(10,13,25)

Hospital stay conditions. The *C. difficile* bacillus has been a generally established agent in nosocomial infections, and the present study demonstrated that it is also an agent in nosocomial infections at the SP CSK. Therefore, we evaluated the effect of hospital stay conditions

at the SP CSK on the risk of developing an infection. Many publications have described the risk factors of an infection in a hospitalized patient based on the hospital stay conditions. These include mainly the presence of sanitary ware in the room, a stay in a multiple-bed room, and the possibility of transmitting the infection via the hands of hospital staff.(10,14,15,26,27) The study demonstrated a higher risk of *C. difficile* infections in the case of the lack of an *en suite* toilet in the patient's room. The risk of contracting a *C. difficile* infection by patients using a common toilet was 1.5 times hither (OR 1.59; P=0.01) in comparison to patients who used an *en suite* toilet. Similar results were obtained with the use of a logistic regression model (P=0.07). The present study also demonstrated a higher risk of contracting a *C. difficile* infection by a "bed-ridden" care-dependent patient in comparison with patients who did not require the help of hospital staff in maintaining their daily hygiene. The risk of contracting a *C. difficile* infection by a "bed-ridden" care-dependent patient was nearly 1.5 times higher than that in a patient who did not require such care (OR 1.69; P=0.01). The increased risk was probably due to transmitting the infection via the hands of hospital staff. Similar results were obtained with the use of a logistic regression model (P=0.08).

An analysis with the use of the Wald test demonstrated a higher risk of contracting a *C. difficile* infection in patients staying in a multi-bed room (of more than 2 patients per room) in comparison with the patients staying in two-bed and single-bed rooms (OR 1.64; P=0.05). However, this was not confirmed by a logistic regression test (P=0.9).

An assessment of the risk of contracting a *C. difficile* infection in the case of a lack of automated bedpan washers at the Clinic yielded contradictory results; namely, an analysis with the Wald test did not reveal an increased risk of contracting a *C. difficile* infection in patients hospitalized in a Clinic with no automated bedpan washers (OR 1.09; P=0.68), whereas, a logistic regression test showed an increase in such risk (P=0.002).

Also, the results showed no increased risk of a *C. difficile* infection in patients using common shower stalls (OR 1.42; P=0.16). Similar results were obtained with the use of a logistic regression test (P=0.73).

RESULTS

Clostridium difficile is an opportunistic bacterium, thus an infection and subsequent development of CDAD/CDI requires a combination of risk factors. Those include the so-called modifiable risk factors, such as hospitalization, antibiotic therapy and non-modifiable risk factors, such as age, underlying disease. Determining the risk factors, especially identifying the modifiable factors, is the basis for the introduction of targeted preventive measures against *C. difficile* infections. The present study demonstrated that a significant risk factor of *C. difficile* infections in patients hospitalized at the SP CSK was the hospitalization itself, including the frequency and conditions of hospital stay, as well as antibiotic therapy. However, this study did not show an increased risk of contracting a *C. difficile* infection in association with such risk factors as age, sex, type of underlying disease, the use of PPIs or a lack of probiotic use. Thus, our evaluation showed *C. difficile* infections to be mostly nosocomial. An active source of infection in a hospital setting are patients infected with *C. difficile* with symptoms of diarrhea. Hospitalization of infected patients is associated with contamination of hospital environment and further transmission of spores on the hands of staff, patients, and visitors. Spores of *C. difficile* are resistant to commonly used disinfectants and antibiotics, which makes these pathogens especially dangerous in a hospital setting. Therefore, our analysis demonstrated that in order to prevent *C. difficile* infections, emphasis should be placed on primary prophylaxis i.e. limiting the transmission of spores in a hospital setting. The basis for prevention of *C. difficile* infections in a hospital setting are: an early established diagnosis, reliable diagnostic assessments, isolation of infected patients, hand hygiene, disinfection of the hospital environment with agents active against spores and the use of rational antibiotic therapy.

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