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Analysis of *Clostridium difficile* infections in patients hospitalized at the nephrological ward in Poland

Analiza zakażeń wywołanych przez *Clostridium difficile* u chorych hospitalizowanych na oddziale nefrologicznym w Polsce

Authors' Contribution:
A Study Design
B Data Collection
C Statistical Analysis
D Data Interpretation
E Manuscript Preparation
F Literature Search
G Funds Collection

Agata Kujawa-Szewieczek [B C E F], **Marcin Adamczak** [A D E], **Katarzyna Kwiecień** [B E], **Sylwia Dudzicz** [B E F], **Zofia Prażak** [D], **Andrzej Więcek** [A D E]

Department of Nephrology, Transplantation and Internal Medicine, Medical University of Silesia in Katowice, Poland

Summary

Background: Few studies have evaluated the incidence and risk factors of *Clostridium difficile* infection (CDI) in the adult Polish population, in particular in solid organ recipients hospitalized at the nephrological ward.

Aim: The aim of this study was to analyze *Clostridium difficile* infections (CDI) among patients hospitalized in the Department of Nephrology, Transplantation and Internal Medicine, Medical University of Silesia in Katowice.

Material/Methods: Thirty-seven patients with *Clostridium difficile* infection diagnosed between October 2011 and November 2013 (26 months), identified among a total of 3728 patients hospitalized in this department during this period, were included in this retrospective, single-center study. The CDI definition was based on the current recommendations of the European Society of Clinical Microbiology and Infectious Diseases.

Results: The observation period was divided into two 13-month intervals. Increased incidence (of borderline significance) of CDI in the second period compared to the first period was observed (1.33% vs 0.65% respectively; p=0.057). Patients after kidney (n=11), kidney and pancreas (n=2) and liver (n=5) transplantation represented 48% of the analyzed CDI patients, and in half of these patients (50%) CDI symptoms occurred within the first 3 months after transplantation. *Clostridium difficile* infection leads to irreversible deterioration of graft function in 38% of kidney recipients. Most incidents of CDI (70%) were identified as nosocomial infection.

Conclusions: 1. *Clostridium difficile* infection is particularly common among patients in the early period after solid organ transplantation. 2. *Clostridium difficile* infection may lead to irreversible deterioration of transplanted kidney function.

Keywords: ***Clostridium difficile*** **• risk factors • kidney transplantation • kidney and pancreas transplantation • liver transplantation**

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Author's address: Prof. Andrzej Więcek, Department of Nephrology, Transplantation and Internal Medicine, Medical University of Silesia, Francuska 20/24, 40-027 Katowice, Poland; e-mail: awiecek@sum.edu.pl

Introduction

Clostridium difficile is currently the most common identifiable pathogen of antibiotic-associated diarrhea [5,22,37] and also a major cause of nosocomial diarrhea [10,25]. This anaerobic, Gram-positive, spore-forming bacterium may produce toxins A and B, which trigger the mechanism of the intestinal mucous membrane damage [21]. The exposure to toxigenic strains of *Clostridium difficile* can lead to asymptomatic colonization of the intestine or to the development of *Clostridium difficile* infection (CDI). It is thought that 3% of the general population is colonized with this bacteria, and this proportion increases to 20-40% among hospitalized patients [33,50]. Moreover, it has been shown that the risk of asymptomatic colonization increases with the length of stay in hospital [44,47]. Clinical manifestations of CDI are very diverse. It may present as diarrhea with no signs of inflammation, inflammation of the intestine without membrane formation, pseudomembranous colitis and fulminant colitis followed by toxic megacolon, paralytic ileus or colon perforation [7,8]. A significant clinical and economic problem is the recurrence of CDI after an initial infection. That may be diagnosed in up to 20% of CDI patients [17,39].

The most important CDI risk factor is antibiotic exposure, especially to those agents with a broad antibacterial spectrum (penicillins, cephalosporins, fluoroquinolones and clindamycin) [22,31,36]. Greater predisposition to CDI was observed in elderly patients, recently hospitalized, with serious comorbidities such as malignant disease, chronic kidney disease, liver cirrhosis, diabetes mellitus or non-specific inflammatory bowel diseases. Additional risk factors include dysfunction of the immune system (HIV-positive patients, patients after solid organ or hematopoietic stem cell transplantation), previous gastrointestinal surgery or endoscopic procedure and the use of medication that suppresses gastric acid production such as proton pump inhibitors or histamine type 2 receptor antagonists [23,29,52].

Patients after solid organ transplantation are particularly predisposed to *Clostridium difficile* infection. It has been reported that the incidence of CDI among organ recipients is significantly higher than in the general population and is estimated to be 3.2 to 16% in kidney recipients, 2.7 to 7% in liver recipients and from 2 to 8% in kidney and pancreas recipients [1,4,15,37,48,49,51]. Such infection was diagnosed more often in the early period after transplantation, in males, in recipients of deceased-donor organs, with diagnosed leukopenia and after exposure to cephalosporins [46]. Limited data are available regarding the incidence and risk factors of CDI in patients after solid organ transplantation in the Polish population [37].

A significant increase in the incidence and the mortality due to *Clostridium difficile* infection has been observed in the past two decades [18,38,41,43]. According to the epidemiological data the number of CDI cases in Poland increased from 2409 in the year 2011 to 3293 cases in the year 2012 [12]. Moreover, higher rates of severe and severe-complicated infection, treatment failure, recurrence of CDI and a greater proportion of community-acquired disease were reported.

The aim of this study was to analyze *Clostridium difficile* infections among patients hospitalized in the Department of Nephrology, Transplantation and Internal Medicine, Medical University of Silesia in Katowice.

Material and methods

Thirty-seven patients with *Clostridium difficile* infection diagnosed between October 2011 and November 2013 (26 months), identified among 3728 patients hospitalized in this period, were included in this retrospective, single-center study. The CDI definition was based on the current recommendations of the European Society of Clinical Microbiology and Infectious Diseases (ESCMID) and was diagnosed in a two-step algorithm. The enzyme immunoassay Premier Toxins A&B test (Meridian Bioscience, Inc., Cincinnati, USA) was performed for *Clostridium difficile* toxin detection in stool samples obtained from patients with diarrhea. All stool samples with an initial negative toxin assay result were evaluated subsequently using the toxigenic culture with chromID *Clostridium difficile* culture media (bioMérieux S.A., Marcy L'Etoile, France) under anaerobic conditions. After 48 hours organisms cultured on selective media were tested for toxin production once again with the enzyme immunoassay Premier Toxins A&B test (Meridian Bioscience, Inc., Cincinnati, USA).

Recurrent CDI was defined as the recurrence of the infection within two months after the resolution of

the previous episode. Severe CDI was diagnosed in the following cases: hospitalization in case of community-acquired CDI, presence of toxic megacolon or perforation which may necessitate colectomy, admission to the intensive care unit or death secondary to CDI within 30 days from onset of symptoms.

Nosocomial infection was defined as infection occurring 48 hours or more after hospital admission. Community-acquired CDI was diagnosed in the case of onset of symptoms up to 48 hours after hospital admission or before admission, if the patient had healthcare exposure for more than the last 3 months.

The following risk factors for severe CDI were included in the analysis: age >70 years, ileus, computed tomographic findings suggestive of CDI, elevated white blood cell count >20 G/l, serum creatinine concentration >2 mg/dl, plasma albumin concentration <2.5 g/l, intravenous immunoglobulin treatment and previous surgery within the last 30 days.

Statistical analysis was performed using the STATISTICA 7.0 PL for Windows software package (StatSoft Polska, Kraków, Poland) and MedCalc 11.3.8. (Mariakerke, Gent, Belgium). Results are presented as mean values and standard deviations. Statistical significance of between-group differences was evaluated by analysis of the chi^2 test. In all statistical tests 'p' values below 0.05 were considered statistically significant.

Results

The characteristics of the study group are shown in Table 1. During the 26 months of observation *Clostridium difficile* infection was diagnosed in 37 patients and one patient suffered from recurrence of CDI. Nosocomial infection was identified in 70% of CDI cases and community-acquired CDI was diagnosed in 3 patients. The average time between the onset of symptoms and the diagnosis of CDI was 8.1 ± 7.5 days. Six (16%) patients developed severe infection and one of them died within 30 days after the onset of CDI symptoms (Table 2). Four cases of severe form of *Clostridium difficile* infection were diagnosed in patients after solid organ transplantation.

Between October 2011 and November 2013 the mean incidence rate of CDI was 9.9 per 1 000 patients hospitalized in the Department of Nephrology, Transplantation and Internal Medicine, Medical University of Silesia in Katowice. This observation period was divided into two 13-month intervals. There was increased incidence (of borderline significance) of *Clostridium difficile* infection in the second period compared to the first period (1.33% vs. 0.65% respectively, p=0.057). The number of incidents of CDI in particular quarters of a 26-month follow-up is shown in Figure 1.

In almost all patients with CDI (n=36, 97.3%) the main treatment was simultaneous use of oral metronidazole and oral vancomycin. One patient, who was in early pregnancy, was treated only with oral vancomycin. Six patients (16.2%) required intravenously administration of metronidazole because of the lack of effect of the oral treatment or the presence of severe CDI. Discontinuation of previous antimicrobial therapy was possible in less than half of the CDI group (43.2%).

Table 1. Characteristics of patients diagnosed with CDI (n=37)

Age [years]	**56 ± 14**
Sex [M/F]	20/17
BMI [kg/m^2]	25 ± 4.9
Diabetes [n/%]	[12/32%]
Chronic kidney disease [n/%]	[22/64%]
Dialysis [n/%]	[8/22%]
Systemic vasculitis [n/%]	[5/13%]
Liver cirrhosis [n/%]	[5/13%]
Inflammatory bowel disease [n/%]	[1/3%]
Cancer [n/%]	[7/19%]
Patients after organ transplantation [n/%]	[18/48%]

Table 2. Severe diseases associated with *Clostridium difficile* (n=6)

Admission to hospital for treatment of community-acquired CDI [n]	3
Toxic megacolon [n]	1
Colectomy [n]	0
Transfer to the intensive care unit due to CDI complications [n]	1
Death [n]	1

Diagnosis of *Clostridium difficile* infection

The main clinical symptom of CDI, observed in 37 (100%) patients, was diarrhea with various severity, sustained for 12.9 ± 8.4 days. The average number of stools per day was 6.7 ± 3.0. During the infection there was noted a reduction in body weight in 18 patients. In these patients mean reduction of body weight was 6.5 ± 4.3 kg.

Abdominal pain, vomiting and fever were other frequent accompanying symptoms, occurring in 22 (59.4%), 4 (10.8%) and 28 (77.7%) patients, respectively.

From patients with a clinical suspicion of having CDI in 35 cases the presence of toxins in the stool samples was detected using the enzyme immunoassay test. In the other two cases, with an initial negative toxin assay result, the toxigenic culture test was positive.

Leukocytosis (> 10 G/L) developed in 25 (67.6%) patients and leukopenia (<4 G/L) was observed in 3 (8.1%) cases. Most of the CDI patients (97.3%) had elevated serum concentrations of C-reactive protein (CRP). The ave-

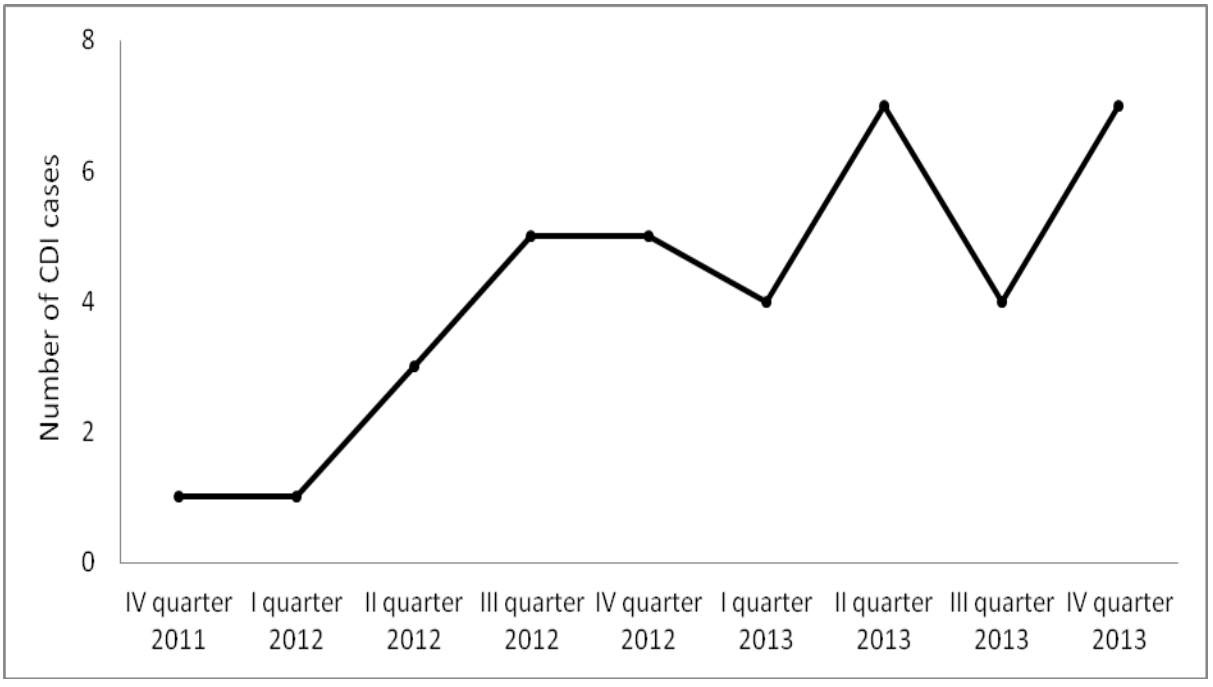

Fig. 1. Number of incidents of CDI in particular quarters of a 26-month observation period

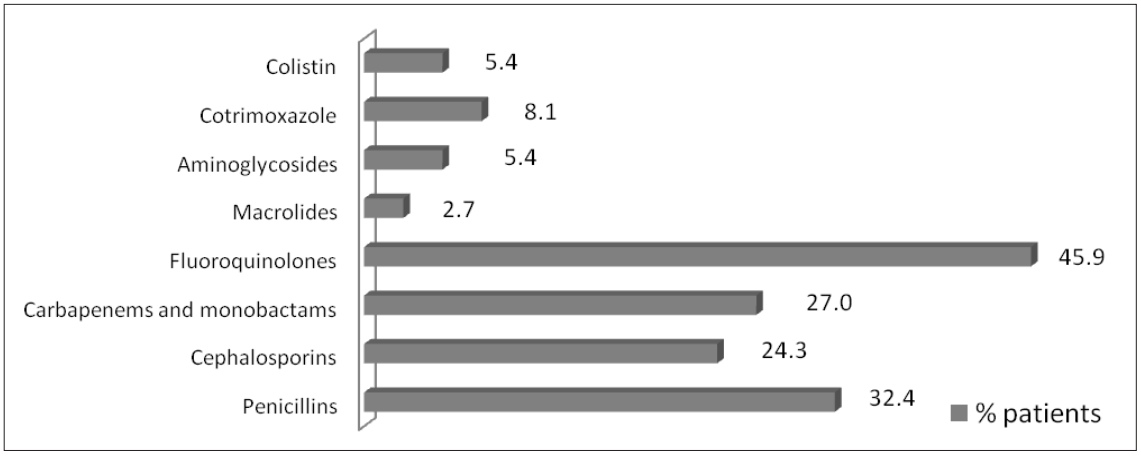

Fig. 2. Antibiotic exposure in the CDI patients

rage leukocyte count and CRP serum concentrations in the study group were 16.8 ± 13.9 G/L and 89.9 ± 80.9 mg/L, respectively. There was noted a decrease in hemoglobin concentration in 20 (54%) patients (by an average of 1.7 ± 1.5 g/dL), and hypokalemia (serum potassium concentration <3.5 mmol/l) was present in 10 (27%) patients.

Toxic megacolon was diagnosed in 1 patient. There was no perforation of the colon and none underwent colectomy.

The average length of stay in hospital and the time between the diagnosis of CDI and the discharge from the hospital in the study group were 21 ± 11 and 11 ± 8 days, respectively.

Risk factors

54% of the study group were male. The mean age of CDI patients was 58 ± 16 years. CDI occurred more frequently in elderly persons. This disease was found before age 40, between 40 and 50 and over age 50 in 6 (16.2%), 6 (16.2%) and 25 patients (67.6%), respectively.

As a risk factor, 29 (78.4%) patients had a history of antibiotic use in the previous 4 weeks before onset of CDI symptoms, and 19 patients (51.3%) were treated with more than one antibiotic. The clinical presentation of CDI was observed an average of 9.1±7.0 days after the beginning of antibiotic therapy, and fluoroquinolones were the most frequently used drugs. The antibiotic exposure in the study group is shown in Figure 2.

Table 3. Risk factors of severe course of CDI in the study group (n=37)

Age > 70 years [%]	16
WBC >20 G/l [%]	22
Serum creatinine > 2 mg/dL [%]	76
Albumin <2.5 g/dL [%]	32
Bowel obstruction [%]	3
Changes in the large intestine detected in CT [%]	8
History of surgery in the last 30 days [%]	13
Intravenous infusion of immunoglobulins [%]	8

Table 4. Immunosuppressive regimens applied in the CDI patients after solid organ transplantation (n=18) and mean blood concentration of calcineurin inhibitors noted before the diagnosis of CDI. CsA – cyclosporine A, Tc – tacrolimus, MMF – mycophenolate mofetil, MPA – mycophenolic acid, E – everolimus, CsA C0 – cyclosporine trough level, Tc C0 – tacrolimus trough level

CsA/Tc [n]	3/15
MMF/MPA [n]	4/5
E [n]	1
Glucocorticoids [n]	14
CsA dose [mg/kg per day]	1.6 ± 0.7
Tc dose [mg/kg per day]	0.1 ± 0.1
MMF dose [g per day]	0.7 ± 0.2
MPA dose [g per day]	0.6 ± 0.5
E dose [mg per day]	1.0 ± 0.0
Glucocorticoids dose [mg per day]	13.2 ± 9.2
CsA C0 [ng/mL] before the diagnosis of CDI	131.5 ± 7.8
Tc C0 [ng/mL] before the diagnosis of CDI	9.5 ± 6.4

Proton pump inhibitors were used in 29 (78.4%) patients with CDI. None of them was treated with histamine 2 receptor blockers.

70.3% of CDI patients were hospitalized in the Department of Nephrology, Transplantation and Internal Medicine or in another hospital during the three months before symptoms of *Clostridium difficile* infection, and every third patient was hospitalized more than once.

The analysis of already known risk factors of severe CDI is shown in Table 3.

Patients after transplantation

Eighteen (48%) patients with CDI were after solid organ transplantation, including 11 renal recipients (i.e. 61% of patients after transplantation), 5 liver recipients (28%) and 2 patients after simultaneous kidney and pancreas transplantation (11%). All transplanted organs came from deceased donors. One-third of patients (33.3%) before the diagnosis of CDI received a triple immunosuppressive therapy, consisting of cyclosporine A (16.6%) or tacrolimus (83.3%), mycophenolate mofetil, mycophenolic acid (50%) or everolimus (5.5%) and steroids. Three patients (16.6%) received basiliximab (Simulect, Novartis, Basel, Switzerland) or antithymocyte globulin (ATG, Fresenius, Bad Homburg, Germany) as an induction therapy. In 3 patients elevated blood concentration of calcineurin inhibitors was noted before the diagnosis of *Clostridium difficile* infection (13 and 17 ng/mL blood concentration of tacrolimus at 2 months after renal transplantation and 26.8 ng/ml blood concentration of tacrolimus at 1 month after liver transplantation). The mean blood concentrations of tacrolimus and cyclosporine A (C0 – cyclosporine trough level) in the course of infection were respectively 10.5 ± 4.2 ng/mL and 101.5 ± 78.5 ng/mL. Immunosuppressive regimens applied in the CDI patients after solid organ transplantation are shown in Table 4.

Table 5. Antibiotic use indications among CDI patients after solid organ transplantation

Indications for antibiotic use	n
Urinary tract infection	10
Pneumonia	1
Upper respiratory tract infection	1
Sepsis	1
Other	3

In 9 recipients (50%) CDI symptoms occurred within the first 3 months after transplantation. 55.5% of transplanted patients were men, leukopenia was diagnosed in 2 recipients, and the most commonly used antibiotics were fluoroquinolones. The indications for antibiotic use among CDI patients after solid organ transplantation are shown in Table 5. Proton pump inhibitors were used in 17 (94.4%) recipients with CDI. This infection leads to deterioration of kidney graft function in 10 kidney or kidney and pancreas transplant recipients. In 5 (38%) patients the loss of graft function was irreversible. The individual serum concentrations of creatinine during *Clostridium difficile* infection among these patients are shown in Figure 3.

Eleven patients with *Clostridium difficile* infection were transplanted between October 2011 and November 2013. In this period there were 325 solid organ transplantations in our Clinical Hospital in Katowice, and the percentage of CDI patients diagnosed at this time among renal transplant recipients, kidney and pancreas and liver recipients was 2.7%, 7.7% and 5.9%, respectively. There was a greater incidence of CDI among patients after liver and simultaneous kidney and pancreas transplantation compared to renal transplant recipients, but this difference was not statistically significant (6.3 vs. 2.7, p= 0.28), probably due to the low number of patients enrolled in this analysis.

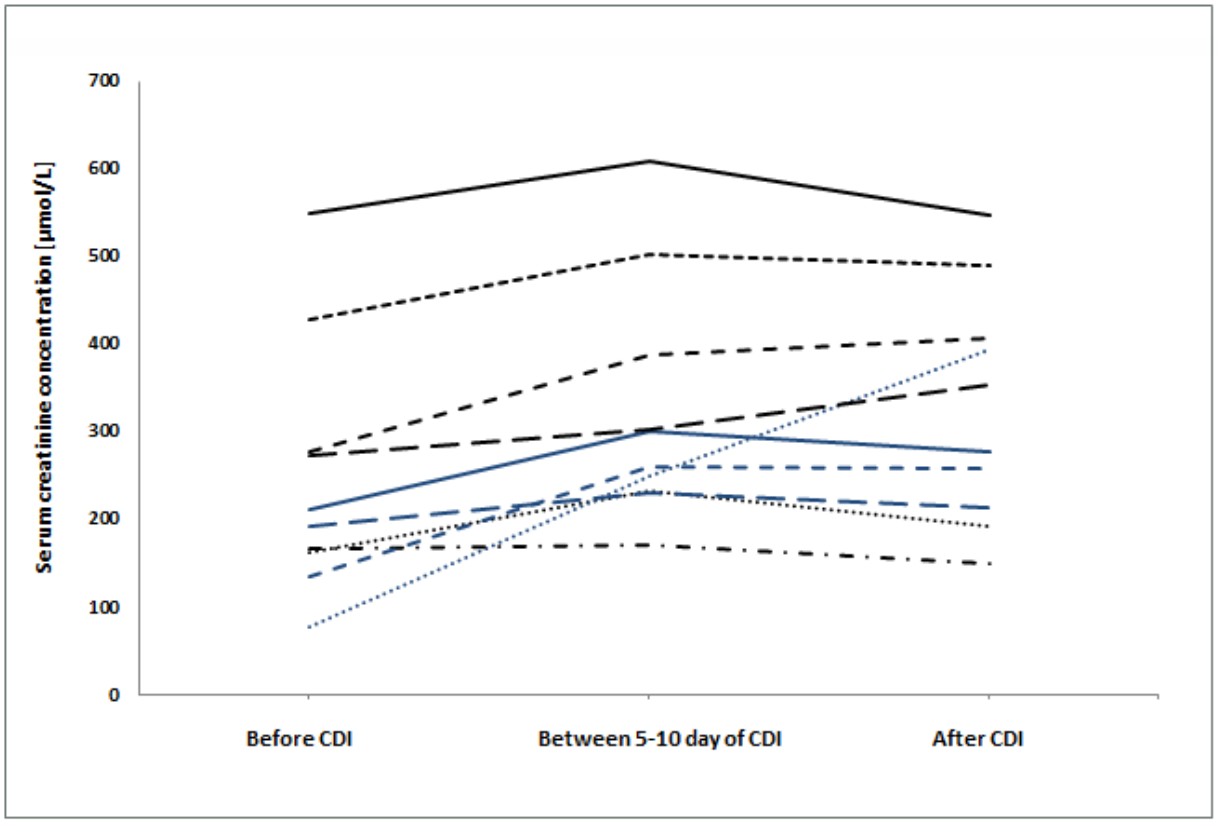

Fig. 3. Serum concentrations of creatinine during *Clostridium difficile* infection among 10 patients with deterioration of kidney graft function

Discussion

This study is one of the few publications so far concerning the incidence and risk factors of *Clostridium difficile* infections in the Polish population. The results showed an increase (of borderline significance) of the incidence rate of CDI in the last 2 years and relatively high severe CDI occurrence. This higher rate of infection observed during past years may be a consequence of increased prevalence of the hypervirulent endemic B1/NAP1/027 strain of *Clostridium difficile*. This ribotype is characterized by extensive production of A and B toxins, production of an additional binary toxin, greater ability to form spores and higher resistance to fluoroquinolones. Moreover, there have been reported associations between infection of the B1/NAP1/027 ribotype and more frequent occurrence of severe and complicated forms of CDI, higher percentage of disease recurrence and higher mortality rate [27,32,45]. According to the epidemiological data, ribotype 027 is detected in 5-75% of CDI cases [9,26] in European countries and in about 30-34% of CDI cases diagnosed in Poland [42]. Due to the retrospective nature of the present study, genotype analysis of *Clostridium difficile* strains was not possible.

The average incidence rate of CDI in the study group was 9.9 per 1000 hospitalizations and was comparable to data presented for the US population [19,30], but higher in comparison to data from studies of the European population [2]. The frequency of *Clostridium difficile* infections in Poland has not been fully explored yet. The preliminary results presented by Pituch et al. showed that in 2011 the mean incidence of CDI was 3.66 per 10 000 hospital admissions [42]. However, according to some authors, the actual CDI frequency remains underestimated by as much as 25%. This may be explained by the variety of diagnostic methods used in different medical centers [2,13].

One of the main risk factors of CDI is the use of broad-spectrum antibiotics [34,45]. This association was confirmed in the present study. The majority of our patients were taking the antibiotics in the previous 4 weeks before onset of CDI symptoms. Fluoroquinolones were the most frequently administered drugs, which may be explained by the fact that urinary tract infections were the main indications for antibiotic use in the study group.

A significant percentage of patients with CDI were treated with proton-pump inhibitors. Studies published so far have presented contradictory results about the correlation between the use of gastric acid secretion reducing drugs and frequency of *Clostridium difficile* infections [10,23]. Nevertheless, in 2012 the Food and Drug Administration made an announcement about the possibility of CDI developing as an adverse effect of proton-pump inhibitors use [16].

CDI incidence rate increases with age and the number of previous hospitalizations [24]. In this study patients aged over 50 were the majority of the CDI group. Moreover, every third patient was hospitalized more than once during the three months before the occurrence of *Clostridium difficile* infection symptoms.

Patients after solid organ transplantations have several unique risk factors that may lead to particularly high CDI predisposition [14]. Such factors are immunosuppressive therapy and frequent use of antibiotics, frequent hospitalizations and immune system dysfunction [14]. Moreover, in the last decades there has been a significant increase in the number of elderly patients after organ transplantation, with numerous accompanying diseases, which may also affect the CDI risk. In the present study, 48% of CDI patients were after organ transplantation and patients after liver transplantation and kidney with pancreas transplantation, characterized by a tendency for a higher CDI incidence rate in comparison to kidney recipients. Similarly, Ali et al. [3], Boutros et al. [11] and Stelzmueller et al. [48], estimating the frequency of CDI among patients after organ transplantations, reported a higher percentage of patients with diagnosed CDI in the group of liver recipients than in kidney recipients. Transplantation of the liver, an organ less immunogenic than kidneys, allows the use of a less aggressive immunosuppressive protocol. Nevertheless, patients with cirrhosis are most often cachectic, with immunodeficiency, and frequently require use of broad-spectrum antibiotics. These patients also stay longer in hospital after the transplantation. In this study, kidney and pancreas recipients had a higher (though without statistical significance) CDI incidence rate, which is consistent with data presented by other authors [22,35,46]. One can assume that using more intensive immunosuppressive therapy, including induction therapy, longer hospitalization after the operation and operating directly within the digestive tract may increase the risk of CDI among patients after kidney and pancreas transplantation.

Similarly to the results of earlier studies [11,35,46], in the present group the symptoms of *Clostridium difficile* infection appeared particularly often in the early period after transplantation, most likely as an effect of intensive immunosuppressive therapy used in this period. An important aspect of this study is to show an adverse effect of *Clostridium difficile* infection on transplanted kidney function. This observation was also confirmed in a case-control study among patients after kidney transplantation, in which poor outcomes, defined as graft loss and/or all-cause mortality, were more common among recipients with CDI (adjusted hazard ratio 5.69; p = 0.001) [40]. In our study no biopsy of the transplanted kidney was performed during the infection or in the early period after CDI. However, one can suppose that dehydration and even a slight degree of variation in blood concentration of immunosuppressive drugs during diarrhea can lead to irreversible damage of the transplanted organ.

American Society of Transplantation recommendations for CDI treatment are based on the results of the few observational and case-control studies [15]. Thus, it seems necessary to perform a large, randomized clinical study to estimate risk factors and diagnostic and therapeutic procedures in this group of patients.

The limitation of our study is that due to its retrospective nature genotype analysis of *Clostridium difficile* strains was not possible. We have found that this infection could lead to deterioration of kidney graft function. However, we did not perform biopsy of transplanted kidneys, so we could not exclude acute rejection and evaluate the histopathological features of the renal damage.

In conclusion, *Clostridium difficile* infection is particularly common among patients in the early period after solid organ transplantation. CDI may lead to irreversible deterioration of transplanted kidney function.

References

[1] Albright J.B., Bonatti H., Mendez J., Kramer D., Stauffer J., Hinder R., Michel J.A., Dickson R.C., Hughes C., Nguyen J., Chua H., Hellinger W.: Early and late onset *Clostridium difficile*-associated colitis following liver transplantation. Transpl. Int., 2007; 20: 856-866

[2] Alcala L., Martin A., Marin M., Sanchez-Somolinos M., Catalan P., Pelaez T., Bouza E., Spanish Clostridium difficile Study Group.: The undiagnosed cases of *Clostridium difficile* infection in a whole nation: where is the problem? Clin. Microbiol. Infect., 2012; 18: E204-E213

[3] Ali M., Ananthakrishnan A.N., Ahmad S., Kumar N., Kumar G., Saeian K.: *Clostridium difficile* infection in hospitalized liver transplant patients: a nationwide analysis. Liver Transpl., 2012; 18: 972-978

[4] Apaydin S., Altiparmak M.R., Saribas S., Ozturk R.: Prevalence of Clostridium difficile toxin in kidney transplant recipients. Scand. J. Infect. Dis., 1998; 30: 542

[5] Barbut F., Petit J.C.: Epidemiology of *Clostridium difficile*-associated infections. Clin. Microbiol. Infect., 2001; 7: 405-410

[6] Bartlett J.G.: Antibiotic-associated diarrhea. N. Engl. J. Med., 2002; 346: 334-339

[7] Bartlett J.G., Gerding D.N.: Clinical recognition and diagnosis of *Clostridium difficile* infection. Clin. Infect. Dis., 2008; 46, Supp. 1: 12-18

[8] Bauer M.P., Kuijper E.J., van Dissel J.T.: European Society of Clinical Microbiology and Infectious Diseases (ESCMID): treatment guidance document for *Clostridium difficile* infection (CDI). Clin. Microbiol. Infect., 2009; 15: 1067-1079

[9] Bauer M.P., Notermans D.W., van Benthem B.H., Brazier J.S., Wilcox M.H., Rupnik M., Monnet D.L., van Dissel J.T., Kuijper E.J., ECDIS Study Group: *Clostridium difficile* infection in Europe: a hospital-based survey. Lancet, 2011; 377: 63-73

[10] Biswal S.: Proton pump inhibitors and risk for *Clostridium difficile* associated diarrhea. Biomed J., 2014; 37: 178-183

[11] Boutros M., Al-Shaibi M., Chan G., Cantarovich M., Rahme E., Paraskevas S., Deschenes M., Ghali P., Wong P., Fernandez M., Giannetti N., Cecere R., Hassanain M., Chaudhury P., Metrakos P., Tchervenkov J., Barkun J.S.: *Clostridium difficile* colitis: increasing incidence, risk factors, and outcomes in solid organ transplant recipients. Transplantation, 2012; 93: 1051-1057

[12] Czarkowski M.P, Cielebak E., Kondej B., Staszewska E.: Choroby zakaźne i zatrucia w Polsce w 2012 roku. www.pzh.gov.pl (10.2013)

[13] Davies K.A., Longshaw C.M., Davis G.L., Bouza E., Barbut F., Barna Z., Delmée M., Fitzpatrick F., Ivanova K., Kuijper E., Macovei I.S., Mentula S., Mastrantonio P., von Müller L., Oleastro M., Petinaki E., Pituch H., et al.: Underdiagnosis of *Clostridium difficile* across Europe: the European, multicentre, prospective, biannual, point-prevalence study of *Clostridium difficile* infection in hospitalised patients with diarrhoea (EUCLID). Lancet Infect. Dis., 2014; 14: 1208-1219

[14] Dubberke E.R., Burdette S.D.: *Clostridium difficile* infections in solid organ transplantation. Am. J. Transpl., 2013; 13, Suppl. 4: 42-49

[15] Dubberke E.R., Riddle D.J.: *Clostridium difficile* in solid organ transplant recipients. Am. J. Transplant., 2009; 9, Suppl. 4: 35-40

[16] FDA Drug Safety Communication: *Clostridium difficile*-associated diarrhea can be associated with stomach acid drugs known as proton pump inhibitors (PPIs). http://www.fda.gov/drugs/drugsafety/ucm290510.htm (10.2013)

[17] Fekety R., McFarland L.V., Surawicz C.M., Greenberg R.N., Elmer G.W., Mulligan M.E.: Recurrent *Clostridium difficile* diarrhea: characteristics of and risk factors for patients enrolled in a prospective, randomized, double-blinded trial. Clin. Infect. Dis., 1997; 24: 324-333

[18] Gerding D.N., Lessa F.C.: The epidemiology of *Clostridium difficile* infection inside and outside health care institutions. Infect. Dis. Clin. North. Am., 2015; 29: 37-50

[19] Ho M., Yang D., Wyle F.A., Mulligan M.E.: Increased incidence of *Clostridium difficile*-associated diarrhea following decreased restriction of antibiotic use. Clin. Infect. Dis., 1996; 23: 102-106

[20] Hubert B., Loo V.G., Bourgault A.M., Poirier L., Dascal A., Fortin E., Dionne M., Lorange M.: A portrait of the geographic dissemination of the *Clostridium difficile* North American pulsed-field type 1 strain and the epidemiology of *C. difficile*-associated disease in Québec. Clin. Infect. Dis., 2007; 44: 238-244

[21] Kelly C.P., LaMont J.T.: *Clostridium difficile* infection. Annu. Rev. Med., 1998; 49: 375-390

[22] Keven K., Basu A., Re L., Tan H., Marcos A., Fung J.J., Starzl T.E., Simmons R.L.: *Clostridium difficile* colitis in patients after kidney and pancreas-kidney transplantation. Transpl. Infect. Dis., 2004; 6: 10-14

[23] Khanna S., Pardi D.S.: *Clostridium difficile* infection: new insights into management. Mayo Clin. Proc., 2012; 87: 1106-1117

[24] Khanna S., Pardi D.S.: *Clostridium difficile* infection: management strategies for a difficult disease. Therap. Adv. Gastroenterol., 2014; 7: 72-86

[25] Khanna S., Pardi D.S.: The growing incidence and severity of *Clostridium difficile* infection in inpatient and outpatient settings. Expert Rev. Gastroenterol. Hepatol., 2010; 4: 409-416

[26] Kuijper E.J., Barbut F., Brazier J.S., Kleinkauf N., Eckmanns T., Lambert M.L., Drudy D., Fitzpatrick F., Wiuff C., Brown D.J., Coia J.E., Pituch H., Reichert P., Even J., Mossong J., et.al: Update of *Clostridium difficile* infection due to PCR ribotype 027 in Europe, 2008. Euro Surveill., 2008; 13: 1-7

[27] Kuijper E.J., Coignard B., Brazier J.S., Suetens C., Drudy D., Wiuff C., Pituch H., Reichert P., Schneider F., Widmer A.F., Olsen K.E., Allerberger F., Notermans D.W., Barbut F., Delmée M., et.al.: Update of *Clostridium difficile*-associated disease due to PCR ribotype 027 in Europe. Euro Surveill., 2007; 12: E1-E2

[28] Kuijper E.J., Coignard B., Tüll P.; ESCMID Study Group for Clostridium difficile; EU Member States; European Centre for Disease Prevention and Control: Emergence of *Clostridium difficile*-associated disease in North America and Europe. Clin. Microbiol. Infect., 2006; 12, Suppl. 6: 2-18

[29] Leonard J., Marshall J.K., Moayyedi P.: Systematic review of the risk of enteric infection in patients taking acid suppression. Am. J. Gastroeneterol., 2007; 102: 2047-2056

[30] Lucado J., Gould C., Elixhauser A.: *Clostridium difficile* infections (CDI) in hospital stays, 2009. Statistical brief #124. Healthcare Cost and Utilization Project (HCUP) Statistical Briefs, [Internet]. Rockville (MD): Agency for Health Care Policy and Research (US); 2006-2012 Jan

[31] Lyerly D.M., Barroso L.A., Wilkins T.D., Depitre C., Corthier G.: Characterization of a toxin A-negative, toxin B-positive strain of *Clostridium difficile*. Infect. Immun., 1992; 60: 4633-4639

[32] McDonald L.C., Killgore G.E., Thompson A., Owens R.C. Jr., Kazakova S.V., Sambol S.P., Johnson S., Gerding D.N.: An epidemic, toxin gene-variant strain of *Clostridium difficile*. N. Engl. J. Med., 2005; 353: 2433-2441

[33] McFarland L.V., Mulligan M.E., Kwok R.Y., Stamm W.E.: Nosocomial acquisition of *Clostridium difficile* infection. N. Engl. J. Med., 1989; 320: 204-210

[34] Mertz D., Frei R., Plagge H., Battegay M., Widmer A.F.: Stronger correlation between antibiotic use and the incidence of *Clostridium difficile* determined by culture results instead of faecal toxin detection only. Eur. J. Clin. Microbiol. Infect. Dis., 2010; 29: 1575-1578

[35] Mitu-Pretorian O.M, Forgacs B., Qumruddin A., Tavakoli A., Augustine T., Pararajasingam R.: Outcomes of patients who develop symptomatic *Clostridium difficile* infection after solid organ transplantation. Transplant. Proc., 2010; 42: 2631-2633

[36] Moyenuddin M., Williamson J.C., Ohl C.A.: *Clostridium difficile*-associated diarrhea: current strategies for diagnosis and therapy. Curr. Gastroenetrol. Rep., 2002; 4: 279-286

[37] Niemczyk M., Leszczyński P., Wyzgał J., Paczek L., Krawczyk M., Luczak M.: Infections caused by *Clostridium difficile* in kidney or liver graft recipients. Ann. Transplant., 2005; 10: 70-74

[38] Noblett S.E., Welfare M., Seymour K.: The role of surgery in *Clostridium difficile* colitis. Br. Med. J., 2009; 338: 1563

[39] Olsen M.A., Yan Y., Reske K.A., Zilberberg M.D., Dubberke E.R.: Recurrent *Clostridium difficile* infection is associated with increased mortality. Clin. Microbiol. Infect., 2015; 21: 164-170

[40] Origüen J., Fernández-Ruiz M., Lumbreras C., Orellana M.Á., López-Medrano F., Ruiz-Merlo T., San Juan R., García-Reyne A., González E., Polanco N., Paz-Artal E., Andrés A., Aguado J.M.: Potential role of post-transplant hypogammaglobulinemia in the risk of *Clostridium difficile* infection after kidney transplantation: a case-control study. Infection, 2015; 43: 413-422

[41] Pépin J., Valiquette L., Alary M.E., Villemure P., Pelletier A., Forget K., Pepin K., Chouinard D.: *Clostridium difficile* associated diarrhea in a region of Quebec from 1991 to 2003: a changing pattern of disease severity. CMAJ, 2004; 171: 466-472

[42] Pituch H., Obuch-Woszczatyński P., Lachowicz D., Młynarczyk G., Wultańska D., Kuijper E.: First results of a polish surveillance programme to *Clostridium difficile* infections. High prevalence of *Clostridium difficile* PCR ribotype 027. 23rd European Congress of Clinical Microbiology and Infectious Diseases (ECCMID): Abstract. Presented April 30, 2013

[43] Reveles K.R., Lee G.C., Boyd N.K., Frei C.R.: The rise in *Clostridium difficile* infection incidence among hospitalized adults in the United States: 2001-2010. Am. J. Infect. Control., 2014; 42: 1028-1032

[44] Riggs M.M., Sethi A.K., Zabarsky T.F., Eckstein E.C., Jump R.L., Donskey C.J.: Asymptomatic carriers are a potential source for trans-

mission of epidemic and nonepidemic *Clostridium difficile* strains among long-term care facility residents. Clin. Infect. Dis., 2007; 45: 992-998

[45] Shah K., Pass L.A., Cox M., Lanham M., Arnold F.W.: Evaluating contemporary antibiotics as a risk factor for *Clostridium difficile* infection in surgical trauma patients. Trauma Acute Care Surg., 2012; 72: 691-695

[46] Shah S.A., Tsapepas D.S., Kubin C.J., Martin S.T., Mohan S., Ratner L.E., Pereira M., Kapur S., Dadhania D., Walker-McDermott J.K.: Risk factors associated with *Clostridium difficile* infection after kidney and pancreas transplantation. Transpl. Infect. Dis., 2013; 15: 502-509

[47] Simor A.E, Bradley S.F., Strausbaugh L.J., Crossley K., Nicolle L.E.: *Clostridium difficile* in long term care facilities for the elderly. Infect. Control Hosp. Epidemiol., 2002; 23: 696-703

[48] Stelzmueller I., Goegele H., Biebl M., Wiesmayr S., Berger N., Tabarelli W., Ruttmann E., Albright J., Margreiter R., Fille M., Bonatti H.: *Clostridium difficile* colitis in solid organ transplantation- a single-center experience. Dig. Dis. Sci., 2007; 52: 3231-3236

[49] Tsapepas D.S., Martin S.T., Miao J., Shah S.A., Scheffert J., Fester K., Ma K., Lat A., Egan R., McKeen J.T.: *Clostridium difficile* infection, a descriptive analysis of solid organ transplant recipients at a single center. Diagn. Microbiol. Infect. Dis., 2015; 81: 299-304

[50] Viscidi R., Willey S., Bartlett J.G.: Isolation rates and toxigenic potential of *Clostridium difficile* isolates from various patient populations. Gastroenterology, 1981; 81: 5-9

[51] West M., Pirenne J., Chavers B., Gillingham K., Sutherland D., Dunn D., Matas A.: *Clostridium difficile* colitis after kidney and kidney-pancreas transplantation. Clin. Transplant., 1999; 13: 318-323

[52] Wistrom J., Norrby S.R., Myhre E.B., Eriksson S., Granstrom G., Lagergren L., Englund G., Nord C.E., Svenungsson B.: Frequency of antibiotic associated diarrhoea in 2462 antibiotic-treated hospitalized patients: a prospective study. J. Antimicrob. Chemother., 2001; 47: 43-50

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