EPIDEMIOLOGICAL ASPECTS OF CLOSTRIDIUM DIFFICILE INFECTION IN THE SOUTHEAST REGION OF ROMANIA

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ABSTRACT

Clostridium difficile infection (CDI) in Galati county is an very important public health problem, the number of cases per 100 outpatients being in the first 5, nationally. The percentage of community-associated infection in this county is higher than the national and european ones. We performed a prospective observational study on 720 patients admitted between 1.01.2017-31.12.2018 in the Clinical Hospital of Infectious Diseases „St. Cuv. Parascheva“ Galati and we analyzed comparative demographic data, risk factors, disease evolution and prognosis of a batch of 565 (78.47%) patients with healthcare facility associated infection, with a batch of 144 patients (20%) with community-associated infection. The identified type of Clostridium difficile was ribotype 027 in 93.61% of the toxigenic strains studied. The study showed that CDI predominantly affects women in urban areas. Patients with community source are younger, with less comorbidities, have episodes of illness with lower severity and better prognosis both in terms of CDI recurrence and post-CDI death. Patients with a nosocomial source are older, with multiple comorbidities, with greater exposure to antibiotic, proton pump inhibitor and recent abdominal surgery, have severe episodes of the disease and have a poorer prognosis than those with a community source.

Keywords: *Clostridium difficile, epidemiology, community-associated infection.*

Introduction

Clostridium difficile infection has become an important public health issue following the outbreak of epidemics in Europe, Canada and United States (USA) in 2011. The emergence of these outbreaks, produced by a germ, initially known as normal intestinal commensal flora, of healthy newborns, was possible due to several genetic mutations that occurred in the historical strain, resulting in the emergence of strain BI / NAP1 / 027, a rapidly spreading strain, which produces an additional toxin (binary toxin), as well as an amount higher in toxins A and B, it has an increased capacity to generate spores and has

a higher resistance to fluoroquinolones than the historical strain (1-4).

The 2011 outbreak study of the US epidemic reported: the estimated number of incidente cases of CDI is 453000, with 83000 recurrent, 29300 deaths within 30 days from CDI and a cost, attributed to CDI, for acute care hospitals of 3427 USD - 9960 USD per episode, as well as a total annual cost of acute care, estimated at 1.2-5.9 billion USD. Also, it was found that 94% of the cases had a recent exposure to healthcare unit (5-8).

The most type of CD strain in the USA was ribotype 027 (28.4%), in a Centers for Disease Control and Prevention study in 2009 (9), and in

England, it had a prevalence of 55% in 2007 and 21% in 2010 (10). This decrease in prevalence of this strain, in England, was associated with great decreases in CDI incidence and related mortality, and was attributed to decreased use of fluoroquinolones, increased awareness of CDI, and improved control of CDI. The Netherlands reported a virulent strain, ribotype 078 whose prevalence increased between 2005 and 2008 and was related with ribotype 027 as severity, but was associated with a population more younger and more CA-CDI (11).

Some studies in Romania show that the main circulating CD is ribotype 027 (12, 13). The number of cases per 100 outpatients in Galati county was in 2018, in the first 5, nationally Figure 1 (14,15).

Figure 1. The incidence of 100 discharged CDI cases reported by hospitals, in Romania, in 2018 (14)

The source of the pathogen is the human and the wild or the domestic animals that host the *Clostridium difficile* (CD) in the gastrointestinal tract, (sick or healthy carriers). The transmission can be done in two ways. The direct mode is represented by transmitting the *Clostridium difficile* strain between the patients and between them and the medical personnel. The indirect mode is represented by the objects, including the clothing, the medical-sanitary equipment, the thermometers and the furniture, which constitute an important way of transmission. The hands can be contaminated directly (including by self-contamination) or indirectly. Patients' receptivity to infections caused by *Clostridium difficile* occurs when the host-bacterial relationship is imbalanced. The body is susceptible to infection with these microorganisms when there are gaps in the commensal microflora of the colon (antibiotic, immunodepression) or mucosal

barriers or the teguments are compromised by surgery, trauma, tumor, ischemia or necrosis, decreasing the potential for local oxide reduction. In addition, decreasing the body's overall resistance through age, alcoholism, diabetes, neoplasia, immunosuppression, angiopathy, results in increased responsiveness to CDI.

The infection with *Clostridium difficile* was initially considered nosocomial, afterwards there were cases in which an epidemiological link with any medical services unit could not be established and the community infection was studied.

Europe - A recent multicenter study in 34 European countries, of 97 hospitals applied increased surveillance criteria for CDI cases and found that 70/506 (14%) of cases were classified as community acquired (16).

North America - A study, from 2010, in six North Carolina centers, reported a 20% prevalence of infections of community origin (CA-CDI) (17). In another US multi-center study in eight geographical areas with 10,342 cases, the prevalence of CA-CDI was 32% (18). Allard and colleagues analyzed 15 hospitals in Montreal 2005-2006 and, 599/2297 (27%) cases were CA-CDI (19).

Australia - A CDI study from three states, Tasmania, Western Australia and Victoria showed that CA-CDI accounted for 26% of all hospitalized CDI cases from 2010-2012 (20).

The CDI incidence in Romania has been steadily increasing since 2011. The CDI surveillance system was implemented at the national level in March 2014, the target population being represented by all patients admitted to the state or private health units in the country. In Romania, in 2018, the number of 10241 cases confirmed, with 2% more cases than in 2017 (10080 cases) entered in the CDI surveillance system. Of the total 10,241 cases confirmed by the CDI, 7743 (76%) were classified as healthcare associated infections (HFA-CDI), 1931 (19%) CA-CDI and 567 (6%) infections of indeterminate origin (21).

The incidence of 100 outpatients of CDI cases reported by hospitals in Galati county in 2017 and 2018 was between 0.44 and 0.66, the maximum value at national level.

Materials and methods

A prospective observational study was conducted that evaluated the demographic and epidemiological characteristics of 720 patients hospitalized with the diagnosis of CDI at the Clinical Hospital of Infectious Diseases „St. Cuv. Parascheva“ Galati, between 1.01.2017-31.12.2018.

The etiologic diagnosis for CD was done with coloured chromatographic immunoassay for the simultaneous qualitative detection of *Clostridium difficile* Glutamate Dehydrogenase (GDH), Toxin A and Toxin B. A subset of 50 patients benefited from a complex etiologic diagnosis that included real-time polymerase chain reaction (PCR) witch detects *tcdB* (CD toxin B encoding gene), *cdt* (binary toxin gene) and deletion of a nucleotide at position 117 of the *tcdC* gene, associated with ribotype 027 strain; and anaerobic culture in chromogenic media with Vitek identification and bacilli visualization in optical or transmission electron microscopy.

Of these 720 patients, 565 (78.47%) of them were hospitalized prior to the onset of the disease in the last 0-4 weeks and were considered HFA-CDI, in 11 (1.52%) patients were hospitalized in the last 4-8 weeks and were considered infections of indefinite origin and 144 (20%) patients either did not have continuous hospitalization during the last 8 weeks, or had them more than 8 weeks before the onset of the disease and were considered CA-CDI, Figure 2.

Figure 2. The number of cases with Healthcare facility - associated CDI and Community - associated CDI

The agreement of the manager and the legal commission of the county hospitals in the south-eastern region of Romania as well as of some hospitals with reported CDI was obtained, in order to obtain the number of CDI cases treated in those units during the period 1.01.2017-31.12.2018. The number of cases from 2017 and 2018, by the CDI treated in these units were: 734 in the Emergency County Clinical Hospital “St. Andrei” Galati, 1 in the Emergency Clinical Hospital for Children “St. Ioan” Galati, 140 in the Pneumophysiology Hospital of Galati, 252 in the Emergency County Clinical Hospital “St. Andrei” Constanta, 207 in the Clinical Hospital of Infectious Diseases Constanta, 40 in the Clinical Hospital of Pneumophysiology Constanta, 180 in the Emergency County Hospital of Tulcea, 364 in the Emergency County Hospital of Buzau, 341 in the Emergency County Hospital of Brăila and 359 in the Emergency County Hospital “St. Pantelimon” of Focsani, Figure 3.

Figure 3. The number of cases from the hospitals in the south-eastern region of Romania that reported CDI treated in these units during the period 1.01.2017-31.12.2018

Results

The three groups of patients: the total group, the HFA-CDI group and the CA-CDI group were analyzed compared to the Medcalc statistical analysis program and the results were summarized in Table 1,2.

In all groups, the female gender and the urban environment predominated, the most significant share being in the CA-CDI group.

In the HFA-CDI group we analyzed the percentage of the hospitals and the source sections,

Table 1. The characteristics of the patients with Healthcare facility - associated CDI and Community - associated CDI, statistical correlation

%	Healthcare facility - associated CDI	Community - associated CDI	Total CDI	Statistical correlation Community / Healthcare facility - associated CDI (Difference)
Female Gender	52.74%	66.66%	55.55%	14.12% (p=0.0032)
Urban living area	65.30%	74.30%	67.22%	9.00% (p=0.0508)
Charlson score				
0-4	45.48%	77.77%	51.94%	31.99% (p<0.0001)
5-6	24.77%	14.58%	22.77%	10.19% (p=0.0126)
7-14	29.73%	7.63%	25.27%	22.10% (p<0.0001)
Consumption of antibiotics before the beginning of CDI	66.72%	42.36%	61.66%	24.36% (p<0.0001)
Consumption of proton pump inhibitors before the beginning of CDI	32.38%	10.41%	27.91%	21.97% (p<0.0001)
Abdominal surgery in the last 8 weeks before the onset of the disease	11.85%	0.69%	9.58%	11.16% (p=0.0001)
ATLAS score				
0-2	60.70%	84.72%	65.83%	24.02% (p<0.0001)
3-5	36.81%	13.88%	32.08%	23.93% (p<0.0001)
6-7	3.83%	1.38%	2.36%	2.45% (p=0.2293)
Relapses	19.29%	10.41%	17.63%	8.88% (p=0.0172)
Reinfections	4.42%	4.86%	4.58%	0.44% (p=0.9982)
Deaths	11.68%	2.77%	10.27%	8.91% (p=0.0023)

in which the patients had a hospitalization between 0-4 weeks before the onset of CDI. The hospitals with the most source cases were the Emergency County Clinical Hospital “St. Andrei” from Galati - 68.84%, the Clinical Hospital of Infectious Diseases „St. Parascheva” Galati - 8.67%, the Pneumophysiology Hospital Galati - 8.31%, Figure 4.

Figure 4. The main source hospitals for CDI

Table 2. Statistical correlations of age, hospitalization and duration of treatment

Arithmetic average /lowest value-highest value/median	Healthcare facility - associated CDI	Community - associated CDI	Total CDI	Statistical correlation Community / Healthcare facility - associated CDI (Difference)
Age (years old)	67.56 3-95 70	58.95 1-91 61	65.73 1-95 68	8.60 (p<0.0001)
Hospitalization(days)	8.19 1-43 8	7.63 1-25 7	8.12 1-43 8	0.56 (p=0.1080)
Duration of treatment (days)	10.23 1-65 10	10.25 1-25 10	10.25 1-65 10	0.01(p=0.9752)

The main source sections were internal medicine - 20%, general surgery - 15.04%, infectious diseases - 8.49%, pneumology - 6.52% gastroenterology - 6.38%, Figure 5.

Figure 5. The main source departments for CDI

A batch of 50 patients who had positive chromatographic immunoassay for GDH and at least one toxin A or B, also did a PCR test to determine the ribotype of the strain (027 or not). The results were: 48 valid tests (2 errors) of which one test with non-toxygen strain, 3 tests with toxigenic strains other than 027, 44 tests with 027 ribotype (93.61% of the toxigenic strains).

Interpretation of the CDI risk factors

The average age in the total group and in the HFA-CDI group was over 65 years, considered a risk factor for CDI and falling in the first category of severity of the episode of the disease quantified by the Atlas score, the patients in the HFA-CDI group being those older. The patients in the CA-CDI group were the youngest, with the average age below the risk threshold for CDI, with the median at the beginning of the first category of severity of the episode of the disease quantified by the Atlas score (60 years). Patients with CDI associate with multiple comorbidities, most often being the cause of the risk factors for CDI, either by consecutive immunosuppression or by nosocomial contact, or by the need for antibiotic or proton pump inhibitors (PPI). We appreciated the cumulative comorbidities of our patients through the Charlson score (22).

In the HFA-CDI group, the highest values of this score were registered, 24.77% of patients with a survival expectancy of 2-21% at 10 years and 29.73% with a survival expectancy at 10 years equal to 0. The CA-CDI patients group presented a much smaller cumulative score, with 77.77% of patients with 10-year survival expectancy over 50%. Antibiotic use in the last 8 weeks prior to the onset of CDI was statistically significantly higher in the HFA-CDI group. In this group aminopenicillins were given in 4.77% of patients receiving antibiotics, cephalosporins in 32.62%, fluoroquinolones in 22.81%, carbapenems in 1.59%, clindamycin in 0.53%. In the community group aminopenicillins were given in 21.31% of patients receiving antibiotics, cephalosporins in 31.14%, fluoroquinolones in 22.95%, carbapenems in 1.63%. With the exception of aminopenicillins, which have a higher percentage in the CA-CDI group, the other antibiotics were used in similar way in the two groups, which leads to the idea of the necessity of revising the protocols for the administration of cephalosporins and fluoroquinolones both in the ambulatory and in the hospital. Recent abdominal surgery and PPI use were statistically significantly higher in the HFA-CDI group.

Interpretation of the severity of the episode of CDI

The ATLAS score (23) was commonly used to assess the severity of CDI for choosing specific antibiotic treatment has recently been correlated, in some studies, with the risk of death by CDI (24) and in the HFA-CDI group has statistically significantly higher severity scores than in the CA-CDI group. The duration of the hospitalization was slightly higher in the group with HFA-CDI, and the duration of the specific antibiotic treatment summing both the period of hospitalization and the prescription at home was similar in the three groups studied.

CDI recurrences, defined as new episodes of disease occurring in the range 0-8 weeks after a CDI episode with a clinical resolution, are statistically significantly more in the HFA-CDI group than in the CA-CDI group.

Reinfections with CD, defined as new episodes of disease occurring over 8 weeks after

an episode of CDI with clinical resolution, are similar in the three groups studied.

The deaths occurring during the hospitalization for CDI and within 30 days from the discharge for CDI, are significantly statistically more in the HFA-CDI group than in the CA-CDI group.

Discussions

In Romania, the percentage of CA-CDI cases is higher than the European one. Galati County is, in 2018, in the top 5 counties with the highest number of CDI cases at 100 discharges. In the south-east region, Galati county has the highest number of CDI cases. The study presented is performed on about half of the cases of the county and shows a higher percentage of CA-CDI cases than the national one.

The CA-CDI group included younger patients with fewer comorbidities who developed episodes of disease with lower severity and with a better prognosis in both CDI recurrence and post-CDI death than those with nosocomial source.

The HFA-CDI group included older patients with multiple comorbidities, with higher exposure to antibiotic use, PPI and recent abdominal surgery, who developed more severe episodes of the disease with a poorer prognosis than those with community source.

Conclusions

The main source of CDI is nosocomial, associated with the intensive and / or inadequate use of antibiotics, but we observed the spread of infection in the community. The increase in the number of community cases, especially in young people, can be correlated with the easy access to the use of antibiotics and proton pump inhibitors, even without a prescription.

Prolonged hospitalization, antibiotic therapy errors and inadequate hygiene are associated with the onset of nosocomial CDI.

Promoting the rules for preventing the transmission of CDI and „antibiotic stewardship” strategies are the key to limiting CDI.

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