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MEDICINE

PREVALENCE AND CLINICAL COURSE OF RELAPSES IN CLOSTRIDIUM DIFFICILE INFECTION IN A ROMANIAN HOSPITAL: A RETROSPECTIVE STUDY

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Introduction: The human intestinal tract is home to a diverse and complex microbial community, which plays a central role in human health. A set of microorganisms that form a true "forgotten organ" of our body which is called the human microbiome.

Clostridium difficile infection is acquired exogenously (spores handcrafted, including by hospital staff) or endogenous (intestinal flora).

The administration of antibiotics, proton pump inhibitors can lead to the imbalance of the intestinal flora, with the selection / favoring of the multiplication of *Clostridium difficile*.

Materials and Methods: We conducted a retrospective study of patients with relapses of CDI for a period of 3 years compared to the total number of cases in the first episode of the disease in patients admitted to a tertiary hospital in South East Romania.

Demographics, risk factors (age, history of antibiotics or proton pump inhibitors in the past, comorbidities, surgery) were noted.

The clinical course and antibiotics use, other than the basic treatment of the condition, were analyzed.

Results: Out of a total of 181 patients identified with CDI, 40 (22.09%) had relapses.

Compared to those in the first episode based on the statistical analysis, we identified as predisposing factors for the recurrence of the symptoms the value of the serum albumin ($p = 0.025 < \alpha = 0.05$) and the duration of hospitalization ($p = 0.025 < \alpha = 0.05$)

In ower study there is no association, no dependency relationship between the age variable and the recurrence variable ($p = 0.1505$).

There are no statistically significant differences regarding the comorbidities, surgeries, antibiotic treatments or proton pump inhibitors in the two groups-with or without recurrence ($p = 0.8695$, $p = 0.0939$, $p = 0.3112$, $p = 0.8687$).

From the point of view of biological characteristics, the value of white blood cells and the value of serum creatinine has no statistical significance ($p = 0.416 > \alpha = 0.05$; $p = 0.607 > \alpha = 0.05$).

Conclusions: Our study highlighted that serum albumin level and the duration of hospitalization were associated with recurrence of CDI.

Keywords: human microbiome, *Clostridium Difficile*, recurrence, risk factors

INTRODUCTION

The history of Clostridium Difficile infection is still being written. It started from the discovery in 1935 by Hall and O'Toole of the colonization of the digestive tract in healthy newborns, later, in 1978, citing the first case of postantibiotic therapy pseudomembranous colitis [1,2]. In the last 20 years, due to the irrational use of antibiotics, we have witnessed a real explosion of cases in North America and Europe [3,4].

Recurrences are probably the most challenging aspect in the management of CD infection. A rate of 10-25% of patients develop a second event of the disease within the next 60 days after treatment of the first episode (most commonly in the first 2

weeks) and then one or more relapses occur in 65% of cases [5,6].

Since 2013, the specialists from the Romanian Ministry of Health have developed a guide for the management of C. difficile infection.

In Romania, the centralized surveillance system started at national level in March 2014. The target population is made up of patients in both the state and private systems.

The prevalence has increased gradually in recent years, reaching recently 5.2 cases per 1000 patients/day compared to other gastroenteritis of viral origin which remained relatively constant [7,8,9].

In the case of recurrent CDI the main associated risk factor was hospitalization in the last year (94%), followed by surgery in the last 2 weeks (36%) and administration of gastric antisecretors (34%) [10].

MATERIALS AND METHODS

We conducted a retrospective study in which we introduced all patients diagnosed with Clostridium Difficile infection in the Clinical Hospital for Infectious Diseases Constanța for a period of 3 years.

The inclusion criteria were: patients over or 18 years of age, regardless of gender. With positive bacteriological tests for the culture of *Clostridium Difficult* being cultured in the hospital laboratory on selective medium with agar and cycloserine-cefoxitin-fructose (BioMerieux, CLO agar). Toxin production was confirmed by immunoenzymatic methods (VIDAS, ELFA A + B- Enzyme Linked Fluorescent Assay).

The case definition for CDI includes the existence of more than 3 diarrhea stools per day or clinical suspicion of toxic megacolon in the absence of diarrhea stools.

Data were collected on risk factors: age> 60 years, antibiotics or proton pump inhibitors previously administered, comorbidities, surgery. Laboratory data: leukocyte count, serum creatinine, serum albumin. Concomitant use of antibiotics other than those recommended for CDI, ALTAS score, the length of hospital stay.

The experimental data was processed using the IBM SPSS Statistics 23 Statistical Processing Program. The procedures used were: Descriptive statistics (for characterizing discrete and continuous variables defined at the database level), Graphs, Parametric statistical tests (t test for independent variables), Nonparametric statistical tests - addressed to categorical variables (test $\chi 2$ of the association, of the connection between two categorical variables, with the calculation of OR, Test $\chi 2$ for the comparison of two proportions).

RESULTS AND DISCUSSIONS

Over a period of 3 years, 181 patients diagnosed with Clostridium Difficile infection were hospitalized at the Infectious Diseases Clinical Hospital in Constanta. Of these, 40 (22.09%) had recurrence of symptoms.

The difference between the proportion of patients aged >60 years and ≤60 years who had recurrence of the symptoms is 7.5% with no statistical significance: 95% CI = -2.8418% to 17.3064%, Chi-squared = 2,068, df = 1, p = 0.1505.

A number of 11 male patients (13.8%) and 29 female patients (20.6%) had relapses. The risk of finding a patient with relapse in the group of men is equal to the risk of finding a patient with relapse in the group of women: OR = 0.616 ≈ 1; 95% IC for OR = (0.289, 1.311).

From 168 patients with comorbidities, 30 patients (17.9%) and, respectively, out of 53 patients without comorbidities, 10 patients (18.9%) had relapses. The difference between the proportion of patients who experienced relapses is 1%, a difference that is not considered to have statistical significance: 95% CI = -9.5314% to 14.4797%, Chi-squared = 0.027, df = 1, p = 0.8695.

The difference between the two proportions of patients in terms of history of surgery (8.7%), use of antibiotics (6.2%) and use of proton pump inhibitors (1%) who experienced relapses is not considered statistically significant (95% CI = -1.5023% to 18.9349%, Chi-squared = 2.807, df = 1, p = 0.0939); (95% CI = -6.6767% to 16.3278%, Chi-squared = 1.026, df = 1, p = 0.3112); (95% CI = -11.7422% to 11.7357%, Chi-squared = 0.027, df = 1, p = 0.8687).

The mean value of leukocytes (white blood cell) level was 10791 celule/mmc for patients who relapsed and 11719.56 celule/mmc for patients who did not relapse.

There are no significant differences between the mean leukocyte value recorded in patients who relapsed and in those who did not relapse ($p = 0.416 > \alpha = 0.05$).

Table 1

Demographic and epidemiological characteristics in patients without recurrence *versus* with recurrence

Characteristics of patients with CDI	**Without recurrence n=181**	**Whith recurrence n=40**	**Chi squere tests p**
Age group:			
<60 years	86(86%)	14(14%)	0.1505
>60 years	95(78.5%)	26(21.5%)	
Sex:			
Women	112(79.4%)	29(20.6%)	0.2084
Men	69(86.3%)	11(13.8%)	
Comorbidities:			
Yes	138(82.1%)	30(17.9%)	0.8695
No	43(81.1%)	10(18.9%)	
Previous surgery:			
No	99(86.1%)	16(13.9%)	0.0939
Yes	82(77.4%)	24(22.6%)	
Previous antibiotics:			
No	46(88.5%)	6(11.5%)	0.3112
Yes	93(82.3%)	20(17.7%)	
Previous proton pump inhibitors:			
Yes	51(81%)	12(19%)	0.8687
No	112(80%)	28(20%)	

Table 2

Therapeutic characteristics in patients without recurrence *versus* with recurrence

Characteristics of patients with CDI	**Without recurrence n=181**	**Whith recurrence n=40**	**Chi squere tests p**
Associated antibiotics:			
No	136(81%)	32(19%)	0.5209
Yes	45(84.9%)	8(15.1%)	
ATLAS score			
1–5	17(77,3%)	5(22,7%)	0.5435
6–11	7(87,5%)	1(12,5%)	
Incalculable	16	175	
Duration of hospitalization:			
Days	7,95	9,67	0.025

When we take in the consideration mean value of serum creatinine (mg/dL) and compare the two groups we find that there is no statistical significance (1.10 mg/dL compared with 1.04 mg/dL) ($p = 0.607 > \alpha = 0.05$).

There are significant differences between the mean serum albumin value in patients who experienced relapses MDA = 3.5 mg / dL and the mean serum albumin value in patients who did not relapse MNU = 2.73 mg / dL: $t = 2.376$, $df = 28$, $p = 0.025 < \alpha = 0.05$, Mdif = 0.766 mg / dL, 95% CI of the Difference = (0.10; 1.42).

Of the 53 patients who used associated antibiotics, other than the etiological treatment of

CDI, 8 patients (15.1%) respectively, out of 168 patients who did not use associated antibiotics, 32 patients (19.0%) had relapses. The difference between the proportion of patients who experienced relapses is 3.9%, having no statistical significance: 95% CI = -9.1476% to 13.6990%, Chi-squared = 0.412, df = 1, p = 0.5209.

The risk of finding a patient with relapse in the ATLAS score patient group 1–5 is equal to the risk of finding a relapse patient in the ATLAS score patient group 6-11: OR= 2,059; 95% IC for OR = (0.202, 20,959). Since $p = 0.392 > \alpha = 0.05$, this means that there is no significant differences between the mean values of ATLAS score.

There are significant differences between the mean value of hospitalization duration in patients who relapsed MDA = 9.67 days and the mean value of hospitalization duration in patients who did not relapse MNU = 7.95 days: t = -2.257, df = 219, $p = 0.025 < \alpha = 0.05$, Mdif = -1,724 days, 95% CI of the Difference = (-3.22; -0.21).

CONCLUSIONS

Our study showed that the risk of finding a patient with recurrence is higher in those with prolonged hospital stay and in those with lower serum albumin levels.

It is very important to know the main risk factors that lead to CDI relapses for better management of these cases.

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