Prophylactic use of Saccharomyces boulardii probiotics in preventing antibiotic-associated diarrhea: a single center hospital-based case-control study in Serbia

Nikola Panic[1], Maja Tufegdzic[1,2], Stefania Boccia[3], Marija Stokic[1], Milutin Bulajic[1,4]

ABSTRACT

BACKGROUND: Antibiotic-associated diarrhea (AAD) develops through the loss of normal bacterial intestinal flora. We have conducted a case-control study in order to assess whether prophylactic administration of *Saccharomyces boulardii* (*S. boulardii*) prevents occurrence of AAD among adult hospitalized patients.

METHODS: Single-center hospital based case-control study was conducted in University Clinic "Dr Dragisa Misovic-Dedinje", Belgrade, Serbia. Hospital records were screened in order to identify all the patients developing AAD in period January 1. 2010 – August 31. 2015. For every case, one age and gender matched control was randomly selected among patients hospitalized at the same time at the same department who were administered with antibiotics and did not develop AAD. For both cases and controls data were extracted on demographics, medical history, indication for use of antibiotics, antibiotics used, and prophylactic use of *S. boulardii* probiotics. The relationship between occurrence of AAD and putative risk factors were measured using the odds ratios (ORs) and their 95% confidence interval (CI) derived from logistic regression analysis.

RESULTS: Number of 59 cases and 59 controls were included in the study. Most of AAD cases were associated with old age (mean age of 78.05), and almost half (49.15%) were hospitalized on geriatrics department. Most prescribed class of antibiotics among cases was III generation cephalosporins (50.85%), followed by fluoroquinolones (28.81%) and trimethoprim-sulfamethoxazole (20.34%). Significantly more cases than controls were treated with carbapenems (16.95% vs. 5.08% respectively, p=0.04). Significantly less cases were administered with prophylactic *S. boulardii* probiotics (18.64% vs. 42.37% p=0.005). We identified prophylactic use of *S. boulardii* to act protectively against development of AAD from both univariate (OR: 0.31, 95% CI: 0.14-0.72) and multivariate analysis (OR:0.36, 95% CI: 0.14-0.80). Use of carbapenems was borderline significant risk factor for development of AAD in univariate (OR: 3.81, 95% CI: 0.99-14.64) as well as multivariate analysis (OR: 3.82, 0.91-16.08) (Table 3).

CONCLUSION: Prophylactic use of probiotics containing *Saccharomyces boulardii* acts protectively against antibiotic-associated diarrhea among hospitalized patients.

Key words: antibiotic-associated diarrhea, Clostridium difficile infection, probiotics, prophylaxis, Saccharomyces boulardii

(1) University Clinical-Hospital Center "Dr Dragisa Misovic-Dedinje", Belgrade, Serbia

(2) General Hospital "Euromedic", Belgrade, Serbia

(3) Institute of Public Health, Section of Hygiene, Catholic University, Largo Agostino Gemelli 8, Rome, Italy

(4) Department of Gastroenterology, University Clinical Hospital 'Santa Maria della Misericordia'

CORRESPONDING AUTHOR: *Nikola Panic - Department for Digestive Endoscopy, University Clinical-Hospital Center "Dr Dragisa Misovic-Dedinje", Milana Tepica 1, 11000 Belgrade, Serbia Tel: 381-64-3509133; Fax: 381-11- 3672025; E-mail: nikola.panicmail@gmail.com*

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INTRODUCTION

Antibiotic-associated diarrhea (AAD) describes any unexplained diarrhea associated with the use of an antibiotic, including infection caused by *Clostridium difficile* (CDI). A rise in the use of antibiotics has resulted in an increase in AAD. Nowadays AAD occurs in 5-35% of patients treated with antibiotics [1]. It ranges from mild forms to severe conditions, some being life-threatening. Most severe such as sepsis and septic shock, toxic megacolon requiring colectomy, and death are mainly associated with CDI [2]. Therefore, conduction of measures aimed at limiting the in-hospital spread of CDI is of highest importance [3].

AAD develops through the loss of normal bacterial intestinal flora. Probiotics are live organisms which improve the microbial balance of the host, counteract disturbances in intestinal flora, and reduce the risk of colonization by pathogenic bacteria [4]. They maintain or restore gut microecology through receptor competition, competition for nutrients, inhibition of epithelial and mucosal adherence of pathogens, introduction of lower colonic pH favoring the growth of nonpathogenic species, stimulation of immunity, or production of antimicrobial substances [5, 6]. As probiotics are associated with low incidence of adverse events [7] and low costs and increased availability, there is an increasing interest in their use of probiotics for the prevention of AAD.

Probiotics include bacteria as well as yeasts. The yeast *Saccharomyces boulardii* (*S. boulardii*) has been reported to have several potentially beneficial properties in the intestinal lumen, including a direct inhibitory action against toxins produced by *C. difficile* [8]. As serious complications of AAD are mainly associated with CDI, *S. boulardii* could be a good candidate for primary prevention of AAD, especially among hospitalized patients.

We have conducted a single-centre hospital based case-control study in order to assess whether prophylactic administration of *S. boulardii* prevents occurrence of AAD among adult hospitalized patients.

METHODS

Single-center hospital based case-control study was conducted in University Clinic "Dr Dragisa Misovic-Dedinje", Belgrade, Serbia in period October-November 2015. Hospital records were screened in order to identify all the patients developing AAD in period January 1. 2010 – August 31. 2015. The inclusion criteria were adult patients aged >18 years who received oral or injected antibiotics, consequentially developing AAD. AAD was defined as diarrhea (≥3 loose stools/day for at least 2 days or ≥5 loose stools/48 h) following in-hospital administration of antibiotics. We did not include patients with basal diarrheal disease (acute enteritis, inflammatory bowel disease, radiation enteritis, ischemic colitis), those diagnosed with *C. difficile* colitis within the previous 3 months and those treated with antidiarrheal, antispasmodic or motility agents for other diseases.

For every case, one age and gender matched control was randomly selected among patients hospitalized at the same time at the same department who were administered with antibiotics and did not develop AAD. For both cases and controls data were extracted on age, gender, department where were hospitalized, medical history, indication for use of antibiotics, antibiotics used, and prophylactic use of S.boulardii probiotics. Prophylactic use of *S. boulardii* was defined as administration of at least 5 billion of living cells of *S. boulardii* one time daily (Bulardi plus, AbelaPharm d.o.o. Belgrade), initiated concomitant with antibiotics and continued at least two weeks after antibiotics are discontinued.

Statistical analysis

Descriptive analysis using proportion and means ± standard deviation was computed for categorical and quantitative variables, respectively. Differences between groups were calculated using a chi-squared, two-group t-tests or Fisher's exact tests, as appropriate.

The relationship between occurrence of AAD and putative risk factors were measured using the odds ratios (ORs) and their 95% confidence interval (CI) derived from logistic regression analysis. Final set of variables for multivariate analysis included significant variables from univariate analysis as well as those being clinically relevant. Statistical analyses were performed using Stata software (StataCorp. 2013. Stata Statistical Software: Release 13. College Station, TX: StataCorp LP).

RESULTS

Number of 59 cases and 59 controls were included in the study. Demographic characteristics and medical history of cases and controls are reported in Table 1. Most of AAD cases were associated with old age (mean age of 78.05), and almost half (49.15%) were hospitalized on geriatrics department (Table 1).

Table 2 reports use of antibiotics and prophylactic *S. boulardii* probiotics in cases and controls included in the study. Majority of AAD cases were treated with antibiotics because of urinary infection (47.46%), followed by respiratory infection, including pneumonia (28.81%) and sepsis (22.03%) (Table 2). Most prescribed class of antibiotics among cases were III generation cephalosporins (50.85%), followed by fluoroquinolones (28.81%) and trimethoprim-sulfamethoxazole (20.34%) (Table 2). Although majority of patients subsequently developing AAD were treated with only one class of antibiotic (61.02%) significant number were treated with two (27.12%), three (6.78%) and even four (5.08%) different classes (Table 2). Significantly more cases than controls were treated with carbapenems (16.95% vs. 5.08% respectively, p=0.04) (Table 2). However, significantly less cases were administered with prophylactic *S. boulardii* probiotics in compare to controls who did not develop AAD (18.64% vs. 42.37% respectively, p=0.005).

TABLE 1

DEMOGRAPHIC CHARACTERISTICS AND MEDICAL HISTORY OF 59 HOSPITAL CASES OF ANTIBIOTIC ASSOCIATED DIARRHEA (AAD) AND 59 CONTROLS INCLUDED IN THE STUDY

	AAD* CASES	CONTROLS	
Age	78.05±9.16	78.45±9.80	p=0.83
GENDER			
Female	31 (52.54%)	32 (54.24%)	p=0.85
Male	28 (47.46%)	27 (45.76%)	
HOSPITAL DEPARTMENT			
Geriatrics	29 (49.15%)	30 (50.85%)	p=1.00
Cardiology	8 (13.56%)	8 (13.56%)	
Endocrinology	7 (11.86%)	7 (11.86%)	
Pulmology	6 (10.17%)	6 (10.17%)	
Gastroenterology	5 (8.47%)	4 (6.78%)	
Hematology	4 (6.78%)	4 (6.78%)	
MEDICAL HISTORY			
Arterial hypertension	31 (50.85%)	29 (49.15%)	p=0.71
Atrial fibrillation	15 (25.42%)	14 (23.73%)	p=0.83
Angina pectoris	13 (22.03%)	16 (27.12%)	p=0.52
Congestive heart failure	18 (30.51%)	22 (37.29%)	p=0.44
Other cardio-vascular condition	17 (28.81%)	25 (42.37%)	p=0.12
Anemia	22 (37.93%)	26 (44.07%)	p=0.50
Diabetes mellitus type I	9 (15.25%)	7 (11.86%)	p=0.59
Diabetes mellitus type II	13 (22.03%)	13 (22.03%)	p=1.00
Chronic obstructive pulmonary disease	9 (15.52%)	12 (20.69%)	p=0.47
Chronic renal insufficiency	21 (35.59%)	19(32.20%)	p=0.70
Malignancy	6 (10.17%)	7 (11.86%)	p=0.77

*AAD: antibiotic associated diarrhea

TABLE 2

USE OF ANTIBIOTICS AND PROPHYLACTIC SACCHAROMYCES BOULARDII (S. BOULARDII†) PROBIOTICS AMONG 59 HOSPITAL CASES OF ANTIBIOTIC ASSOCIATED DIARRHEA (AAD) AND 59 CONTROLS INCLUDED IN THE STUDY

	AAD* CASES	CONTROLS	
INDICATION FOR ANTIBIOTIC			
Urinary infection	28 (47.46%)	30 (50.85%)	p=0.75
Respiratory infection	17 (28.81%)	15 (25.42%)	
Sepsis	13 (22.03%)	14 (23.73%)	
Other	1 (1.59%)	0 (0.0%)	
ANTIBIOTIC CLASS			
Cephalosporin III generation	30 (50.85%)	38 (64.41%)	p=0.14
Fluoroquinolone	17 (28.81%)	19 (32.76%)	p=0.65
Trimethoprim-sulfamethoxazole	12 (20.34%)	7 (11.86%)	p=0.21
Aminoglycosides	11 (18.64%)	12 (18.64%)	p=1.00
Carbapenem	10(16.95%)	3 (5.08%)	p=0.04
Penicillin	7 (11.86%)	5 (8.47%)	p=0.54
Glycylcycline	3 (5.08%)	4 (6.78%)	p=0.70
Macrolide	2 (3.39%)	3 (5.08%)	p=0.65
Cephalosporin II generation	0 (0.0%)	1 (1.59%)	p=0.31
CLASSES OF ANTIBIOTICS USED			
One	36 (61.02%)	37 (62.71%)	p=0.95
Two	16 (27.12)	15 (25.42%)	
Three	4 (6.78%)	5 (8.47%)	
Four	3 (5.08%)	2 (3.39%)	
PROPHYLACTIC PROBIOTICS (S.BOULARDII†)			
Yes	11 (18.64%)	25 (42.37%)	p=0.005
No	48 (81.36%)	34 (57.63%)	

*AAD: antibiotic associated diarrhea; † S.Boulardii: Saccharomyces boulardii

We identified prophylactic use of *S. boulardii* probiotics to act protectively against development of AAD from both univariate [odds ratio (OR): 0.31, 95% confidence interval (CI): 0.14-0.72] and multivariate analysis (OR:0.36, 95% CI: 0.14-0.80) (Table 3). Use of carbapenems was borderline significant risk factor for development of AAD in univariate (OR: 3.81, 95% CI: 0.99-14.64) as well as multivariate analysis (OR: 3.82, 0.91-16.08) (Table 3).

DISCUSSION

We report prophylactic use of probiotics containing *S. boulardii* to act protectively against AAD among hospitalized patients. Use of carbapenems seems to be a risk factor for development of AAD.

Number of mechanisms has been suggested in order to explain prophylactic role of *S. boulardii* in preventing AAD and CDI [8, 9]. *S. boulardii* acts within the intestinal lumen through luminal action, trophic action and mucosal-anti-inflammatory signaling effects [10-12]. It interferes with pathogenic toxins, preserve cellular physiology, interfere with pathogen attachment, interact with normal microbiota and assist in reestablishing short chain fatty acid levels [8]. It shows anti-toxin effect by direct destruction of the pathogenic toxin, blocking pathogen toxin receptor sites [13], or acting as a decoy receptor for the pathogenic toxin [14]. *S. boulardii* has been reported to stimulate intestinal immunoglobulin A immune

TABLE 3

RISK FACTORS FOR THE DEVELOPMENT OF ANTIBIOTIC-ASSOCIATED DIARRHEA (AAD) FROM UNIVARIATE AND MULTIVARIATE ANALYSES

	OR	CI 95%	ADJ OR*	CI 95%
Probiotics (*S.Boulardii*†) use	0.31	0.14-0.72	0.36	0.14-0.80
Carbapenems	3.81	0.99-14.64	3.82	0.91-16.08

† *S.Boulardii: Saccharomyces boulardii*
*adjusted by age, gender, indication for antibiotic use
OR= odds ratio; CI=confidence interval

response to *C. difficile* toxin A in mice models [15]. Furthermore it produces a 54 kDa serine protease that inactivates a receptor for toxin A of *C. difficile* and degrades *C. difficile* toxins A and B [16]. Finally *S. boulardii* can reduce mucositis [17], restore fluid transport pathways [18], and stimulate protein and energy production [19] in enterocytes.

Because of the characteristics of *S. boulardii*, in recent years there was a lot of interest in its prophylactic use in preventing AAD and CDI. Recent meta-analysis of randomized controlled trials reported *S. boulardii* to be effective in reducing the risk of AAD in children and adults, with risk reduction of 18.7% to 8.5% compared to no treatment or placebo [20]. Meta-analysis included 21 studies from all over the world. However none of the studies were conducted in region of south-eastern Europe, including Serbia. Recent study reported CDI to be the most important cause of hospital-acquired diarrhea in Serbia, mainly affecting elderly hospitalized patients with co-morbidities [21]. Furthermore CDI PCR-ribotype 027 has been reported to be predominant in acute care hospitals in Serbia [22]. As the ribotype 027 is associated with more severe type for CDI, more refractory to standard therapy, and more likely to relapse [23], prophylaxis is becoming increasingly important. Nevertheless, no research has been conducted on this important topic in Serbia so far. Therefore, we find our results, suggesting the prophylactic use of *S. boulardii* probiotics in preventing AAD among hospitalized patients in Serbia, very important.

Carbapenems have been previously reported to increase the risk of AAD, and especially CDI [24-26]. As the carbapenems are β-lactam antibiotics with the broadest spectrum of activity it is reasonable to assume that they have a strong propensity to disrupt intestinal microflora.

Our study has some limitations. First of all we were not able to conduct RCT, thus we needed to rely on result on case-control study. Second we conducted a single-center study, and therefore our sample was limited. Nevertheless we find our results important as this is first study in Serbia addressing the issue of prophylactic use of probiotics in preventing AAD.

CONCLUSION

In conclusion, prophylactic use of probiotics containing *Saccharomyces boulardii* acts protectively against antibiotic-associated diarrhea among hospitalized patients. Use of carbapenems represents risk factor for development of antibiotic-associated diarrhea.

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