The effect of Saccharomyces boulardii primary prevention on risk of Hospital Onset Clostridioides difficile infection in hospitalized patients administered antibiotics frequently associated with Clostridioides difficile infection

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SUMMARY: Use of single probiotic strain, *Saccharomyces boulardii*, in a cohort of hospitalized patients administered antibiotics frequently associated with hospital onset *Clostridioides difficile* infection (HO-CDI), resulted in a significant HO-CDI risk reduction, especially when started early relative to antibiotics.

Abstract

BACKGROUND: Hospital Onset *Clostridioides difficile* infection (HO-CDI) is a costly problem leading to readmissions, morbidity and mortality. We evaluated the effect of a single probiotic strain, *Saccharomyces boulardii*, at a standardized dose on the risk of HO-CDI within hospitalized patients administered antibiotics frequently associated with HO-CDI.
METHODS: This retrospective cohort study merged hospital prescribing data with HO-CDI case data. The study assessed patients hospitalized from January 2016 through March 2017 that were administered at least one dose of an antibiotic frequently associated with HO-CDI during hospitalization. Associations between *S. boulardii* administration, including timing, and HO-CDI incidence were evaluated by multivariable logistic regression.
RESULTS: The study included 8,763 patients. HO-CDI incidence was 0.66% in the overall cohort. HO-CDI incidence was 0.56% and 0.82% among patients co-administered *S. boulardii* with antibiotics and not co-administered *S. boulardii*, respectively. In adjusted analysis, patients co-administered *S. boulardii* had a reduced risk of HO-CDI (OR=0.57, 95% CI 0.33–0.96, p=0.04) compared to patients not co-administered *S. boulardii*. Patients co-administered *S. boulardii* within 24-hours of antibiotic start demonstrated a reduced risk of HO-CDI (OR=0.47, 95% CI 0.23–0.97, p=0.04) compared to those co-administered *S. boulardii* after 24-hours of antibiotic start.
CONCLUSIONS: *S. boulardii* administered to hospitalized patients prescribed antibiotics frequently linked with HO-CDI was associated with a reduced risk of HO-CDI.

KEYWORDS: clostridium infections; saccharomyces; probiotics; nosocomial infection; infection control

Introduction

Clostridioides difficile infection (CDI) has become one of the most common healthcare-associated infections in the United States with the incidence nearly doubling between 2001 and 2010.[1,2] The Centers for Disease Control considers CDI an urgent threat requiring prevention and monitoring.[1] Risk factors for CDI include increasing age, proton-pump inhibitors, and most significantly, broad-spectrum antibiotics that are hypothesized to accelerate *Clostridioides difficile* (*C. difficile*) colonization by reducing levels of beneficial bacteria that serve as barriers to infection.[3] Co-administering probiotics with antibiotics may prevent CDI development by reinforcing the barrier of good bacteria lost through antibiotic administration.[4,5]

Beyond restoring altered intestinal microflora, administering probiotics may stimulate the immune system to prevent pathogen adhesion and invasion, and clear pathogens and toxins from the intestinal tract.[6] *Saccharomyces boulardii* (*S. boulardii*), a specific yeast-derived probiotic, may prevent CDI by inducing direct inhibitory actions against *C. difficile* toxins.[5,6] Two studies demonstrate upregulation of total and specific intestinal antibodies (IgA) to toxin A in response to *S. boulardii* exposure, consequently reducing *C. difficile* pathogenicity.[7,8] Other evidence demonstrates *S. boulardii* directly inhibits *C. difficile* toxin by releasing a protease to hydrolyze *C. difficile* toxins.[6,9]

Available literature examining the clinical outcomes of probiotics in preventing Hospital Onset *C. difficile* infection (HO-CDI) is contentious. Three randomized clinical trials[10-12] and one cohort study[13] demonstrate *S. boulardii's* primary or secondary prevention effects on HO-CDI. In contrast, the largest randomized trial of probiotics to date[14] and one cohort study of *S. boulardii*[15] show no protective associations. Overall these studies are small in scale and do not take into account the timing of probiotic initiation relative to antibiotic start.[5,11,15] A meta-analysis found administering probiotics more than 2 days after initiating

antibiotics significantly reduced the efficacy by half.[16] Given mixed evidence, research needs to assess the extent to which administrative timing impacts probiotics' beneficial effects.

Given the limitations of previous studies, the Infectious Disease Society of America (IDSA) and Society of Healthcare Epidemiology of America (SHEA) state that "there [is] insufficient data at this time to recommend administration of probiotics for primary prevention of CDI".[17] The IDSA and SHEA guideline writers cite specific limitations such as studies including patients with abnormally high rates of CDI, using inconsistent probiotic formulations across studies, or assessing patient populations at low risk for CDI.[17] These limitations underscore the need for further research. The guideline recommendations from IDSA and SHEA conclude with suggested areas of further research including: 1) What preventive measures can be taken to reduce the incidence of CDI? 2) Can administration of probiotics effectively prevent CDI?

Our primary objective was to evaluate the effect of prescribing a single probiotic formulation *S. boulardii* on risk of developing HO-CDI in a large cohort of hospitalized patients receiving antibiotics frequently associated with HO-CDI. Our secondary objective was to evaluate the extent to which timing of *S. boulardii* initiation relative to antibiotic start affects HO-CDI risk.

Methods

This retrospective observational cohort study compares the risk of HO-CDI in hospitalized patients that only received antibiotics frequently associated with HO-CDI versus those who received antibiotics in combination with *S. boulardii*. The study merged *C. difficile* case data with medication administration records to evaluate associations between HO-CDI incidence and *S. boulardii* administration occurring between January 1, 2016 and March 31, 2017. The study setting is a 220-bed level-2 trauma center non-academic hospital.

In December 2015, our institution established *S. boulardii* at a dose of 250 mg twice daily as the only formulary probiotic. This decision resulted from an internally performed literature review of probiotic agents by the Pharmacy and Therapeutics Committee. The committee concluded from the literature reviewed that the use of *S. boulardii* at a dose of 250mg twice daily was more often associated with positive outcomes for reducing antibiotic associated diarrhea and CDI. Corresponding with the formulary change, an electronic pop-up box was added to the physician electronic order entry system with an option to order *S. boulardii*. The pop-up box appeared following the entry of an antibiotic order for beta-lactams, fluoroquinolones, or lincosamides. The *S. boulardii* order was not an automatic reflex order. The pop-up box also listed precautions for use of *S. boulardii*, including 1) a history of organ transplantation currently receiving anti-rejection medication; 2) concomitantly receiving chemotherapy or radiation; 3) low neutrophil count; 3) diagnosis of AIDS; or 4) active GI ulcer.

The cohort included adult patients 1) admitted to an inpatient medical unit with an average length of stay equaling 3 days or greater, and 2) having barcode administration evidence for reception of at least one dose of an antibiotic frequently associated with HO-CDI, the latter of which was defined as clindamycin, fluoroquinolones, third- and later-generation cephalosporins, carbapenems, and penicillins.[19-21] For the purpose of this study,

"antibiotic" will subsequently refer to these defined antibiotics frequently associated with HO-CDI. The unit of analysis was defined as the first hospitalization during the study period. Only the first hospitalization per unique patient was included in the analytic dataset to remove within-patient changes in *S. boulardii* exposure across separate hospitalizations.

Patients were classified as either having been or not having been administered *S. boulardii* during hospitalization, based upon 1) having barcode administration evidence for reception of at least 1 dose of *S.boulardii*; or 2) no evidence of barcode scan for administration, respectively. *S. boulardii* (Florastor Daily Probiotic Supplement®, Biocodex, Inc., Redwood City, CA, USA) administration consisted of two - 250mg capsules by mouth twice daily. Each 250mg capsule contains 5 billion CFUs for a total daily administration of 20 billion CFUs. The dose of 20 billion CFUs is consistent with previously published studies assessing CDI primary prevention.[22]

The study used *C. difficile* case data defined by and reported to the National Health Safety Network (NHSN) from our institution's infection control office. HO-CDI cases were defined as positive if an unformed stool specimen tested positive for *C. difficile* greater than or equal to 3 days after admission and greater than 8 weeks from a previous positive specimen result, consistent with the CDC definition for HO-CDI.[23] Cases were laboratory confirmed cases by polymerase chain-reaction for the gene encoding toxin B alone without a reflex algorithm. Patients required the following criteria prior to testing: 1) 3+ loose stools within 24 hours, 2) no laxative/stool softeners within 48 hours, 3) no positive *C. difficile* test in last 30 days, and 4) no negative *C. difficile* test in last 7 days. Samples were required to be watery and if the stool sample received by the lab was formed or semi-formed, the lab rejected the sample.

To evaluate the effect of *S. boulardii* administration on HO-CDI risk, we conducted our first multivariable logistic regression to estimate the risk of HO-CDI during a

hospitalization conditional upon the co-administration of *S. boulardii* with antibiotic(s) versus antibiotic(s) without *S. boulardii*. To account for both potential confounders and selection bias, the model included propensity scores generated by a separate multivariable logistic model testing the likelihood of patients receiving *S. boulardii* conditional upon: 1) antibiotic(s) administered during hospital admission; 2) metronidazole administration during hospitalization but greater than 48-hours prior to CDI diagnosis; 3) proton-pump inhibitor (PPI) administration at any time during hospitalization prior to CDI diagnosis; 4) intensive care unit (ICU) admission; 5) patient age of 65-years or greater, and 6) gender. Each hospitalization was assigned a propensity score used as a covariate in the final multivariate model.

To evaluate the extent to which the timing of *S. boulardii* initiation relative to the first dose of antibiotic affected HO-CDI risk, we conducted a second multivariable logistic regression amongst only the subgroup of patients receiving *S. boulardii*. This model tested the risk of HO-CDI conditional upon early versus late co-administration of *S. boulardii* with antibiotics. "Early" was defined as *S. boulardii* administered within 24 hours of first antibiotic dose administration; "Late" was defined as *S. boulardii* administered 24 hours or more after first antibiotic dose. Similar to the model run in the full cohort, propensity scores for this second analysis were generated with a multivariable logistic model testing the likelihood of patients receiving Early versus Late *S. boulardii* conditional upon the same baseline covariates included in the primary model run in the full cohort. Each hospitalization was assigned a propensity score used as a covariate in the final multivariable model. Receiver operating characteristic (ROC) curves were used to calculate C-statistics to estimate the overall global fit for all multivariable logistic regressions, including those used to generate propensity scores. All statistical analysis were conducted using Statistical Package for the Social Sciences (SPSS 24.0, IBM-SPSS Inc., Chicago, IL).

Results

A total of 8,763 patients administered at least one dose of an antibiotic frequently associated with HO-CDI were assessed. The cohort was 39% male and averaged 64 years of age. Patients co-administered *S. boulardii* and antibiotics were more often male ($p<0.0001$) and older than 65 years of age ($p<0.0001$) compared to patients only administered antibiotics. Ceftriaxone, piperacillin-tazobactam, levofloxacin, and ciprofloxacin were the most frequently administered antibiotics, administered in 44%, 32%, 25% and 21% of patients, respectively. Carbapenem, fluoroquinolone and cephalosporin administration was significantly higher in patients co-administered *S. boulardii* versus not co-administered *S. boulardii* ($p<0.0001$). PPIs were administered in 46% of patients. PPI administration was significantly higher in patients co-administered *S. boulardii* and antibiotics (50%) compared to patients not co-administered *S. boulardii* (41%) ($p<0.0001$) (Table 1).

The overall incidence of HO-CDI during the study period was 0.66%. Patients admitted to the ICU and patients administered PPIs demonstrated a higher incidence of and risk for HO-CDI (Table 2 and Table 3). The incidence of HO-CDI was lower in patients co-administered *S. boulardii* and antibiotics (0.56%) compared to patients administered antibiotics without *S. boulardii* (0.82%). With respect to administration timing, the incidence of HO-CDI in patients receiving early *S. boulardii* was less than half of those patients administered antibiotics alone without *S. boulardii* (0.38% vs. 0.82%) (Table 2).

When adjusting for possible confounders and selection bias using the propensity score, the risk for HO-CDI was significantly less in patients administered *S. boulardii* and antibiotics (OR=0.57, 95% CI 0.33 – 0.96) (Table 4) compared to patients administered antibiotics alone. Early *S. boulardii* administration displayed a stronger HO-CDI

preventative effect compared to late *S. boulardii* administration (OR=0.47, 95% CI 0.23 – 0.97) (Table 5).

Discussion

When adjusted for potential confounders, patients administered *S. boulardii* in conjunction with antibiotics frequently associated with HO-CDI had a lower risk of developing HO-CDI compared with patients not administered *S. boulardii* in this single center study. The protective effect was more pronounced in the early *S. boulardii* sub-group versus the late *S. boulardii* subgroup, suggesting early administration might offer greater HO-CDI risk reduction compared to late administration. A recent meta-analysis on the use of probiotics for primary prevention of HO-CDI revealed similar findings alluding to the importance of probiotic initiation timing relative to the first dose of antibiotic.[16] The meta-analysis observed the preventative effect was limited to probiotic initiation within 1-2 days of antibiotic start and a lack of benefit when administered outside of 2-days.

To explore this idea further, we conducted a post-hoc multivariable logistic regression conditional upon the co-administration of early *S. boulardii* versus no *S. boulardii* with antibiotics. The analysis resulted in a reduced odds risk of HO-CDI for early *S. boulardii* vs no *S. boulardii* (OR = 0.44; 95% CI, 0.23 – 0.84, p = 0.013; C-statistic = 0.601), demonstrating a consistent stronger effect for early *S. boulardii* and HO-CDI risk reduction. Presumptively, an earlier initiation of a preventative therapy would have a greater effect than a preventative therapy started further from the inciting event. Here we provide data to support that assumption with *S. boulardii* administration relative to antibiotic initiation. The secondary analysis included a small number of events and therefore considered a provisional finding requiring further investigation.

Our study has several limitations. First, our sample originates from a single medical center and decreases the generalizability of the findings. Second, we were unable to assess pre-existing conditions, such as immunosuppression and CDI history, or patient severity status as risk factors for HO-CDI within the study or provider-level preventative practices. To mitigate this limitation, we included intensive care unit admission as a confounder to account for patients at a higher acuity level with severe illness. Third, providers were cautioned to avoid prescribing probiotics in patients 1) with a history of organ transplantation currently receiving anti-rejection medication; 2) receiving chemotherapy or radiation; 3) with low neutrophil count; 4) diagnosed with AIDS; or 5) having an active GI ulcer.[24-26] There was no system in place to ensure *S. boulardii* was not prescribed to a patient who met these criteria. Consequently, we cannot make conclusions regarding the probiotic effectiveness in these patient groups.

This study has several strengths. First, including propensity scores in our models helped us account for both confounding and selection bias. The propensity score helps account for observable risk factors either associated with an increase or decreased risk of HO-CDI. In our analysis, patients administered *S. boulardii* could be considered higher-risk for HO-CDI as they were more often over 65-years of age, administered a PPI during hospitalization at a higher rate, and more likely to receive a third- or fourth-generation cephalosporin, carbepenem, and fluoroquinolone than non-*S. boulardii* recipients. Adding age, PPI and antibiotic class utilization as covariates in the multivariable model used to derive the propensity score allows the propensity score to account for all of these factors simultaneously. Furthermore, the propensity score helps account for underlying factors driving selection bias that could not be measured in our dataset, including, but not necessarily limited to, patient history of HO-CDI or provider-level behaviors. Second, our observational study design enables us to measure real-world effectiveness based upon daily clinical

encounters in contrast to clinical trials that may not reflect outcomes or patients seen in everyday practice. Evidence from real-world practice can better inform changes in clinical guidelines or formulary policies. Third, isolating our study to a single-agent at a set dose improves our ability to determine the effects of uniform doses and inform specific dose recommendations. In contrast, previous studies included multiple probiotics in the same treatment group and could not tease out the effects of isolated agents or dosages. Fourth, we conducted a subgroup analysis to explore the importance of *S. boulardii* administration timing relative to antibiotic start and resultant HO-CDI incidence. This sub-group analysis suggests earlier probiotic administration timing improves probiotic primary prevention efficacy. Finally, our point estimate of OR=0.57 is similar with those found in meta-analyses by Johnston et al. (RR=0.34; 95% CI, 0.24-0.49)[27], Goldenberg et al. (RR= 0.36; 95% CI, 0.26-0.51)[28], Lau et al. (RR=0.40; 95% CI, 0.29-0.53)[29] and Shen et al. (RR=0.42; 95% CI, 0.30-0.57).[16] Similar results across multiple studies further strengthens confidence in our estimates.

We observed a number needed to treat (NNT) for early *S. boulardii* administration of 228, amounting to 17 cases of HO-CDI that may have been prevented amongst the 3,936 patients administered *S. boulardii* within 24 hours of antibiotic start during the study period. Given the magnitude of benefit and the relatively low cost of probiotics compared to the high cost of treating CDIs during hospitalization and post-discharge, adding *S. boulardii* probiotic as a primary preventative strategy may be a valuable strategy to reduce HO-CDI. Furthermore, results from a cost-effectiveness analysis suggest probiotic use is a cost-effective strategy in preventing CDI in hospitalized adults receiving antibiotics.[30]

Conclusions

This study provides evidence to support prescribing a uniform probiotic formulation as primary prophylaxis of HO-CDI in a high-risk population, a noted weaknesses in the existing literature.[17] We observed co-administration of probiotic *S. boulardii* at a dose of 20 billion CFUs per day with antibiotic therapy frequently associated with HO-CDI in an older hospitalized patient population significantly reduced the incidence of HO-CDI. Furthermore, initiating probiotics within 24-hours of antibiotic start appears to offer a more pronounced barrier to HO-CDI development. Our findings in conjunction with evidence from previous studies support continued exploration of probiotics for primary prevention of HO-CDI, with particular consideration of earlier probiotic administration timing.

NOTES

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Table 1. Demographic and Baseline Characteristics, by Cohort

N (%)	Total Patients (N=8763)	Patients Not Co-administered *S. boulardii* (N=3276)	Patients Co-administered *S. boulardii* (N=5487)	p-value
Demographics				
Age, mean ± SD, y	64.3 ± 18.4	62.3 ± 19.2	65.4 ± 17.7	< 0.001
Age >65	4631 (52.8)	1606 (49.0)	3025 (55.1)	< 0.001
Male Sex	3390 (38.7)	1259 (38.4)	2131 (38.8)	< 0.001
Intensive Care Unit	467 (5.3)	223 (6.8)	244 (4.4)	< 0.001
Concomitant Medications				
Metronidazole	1211 (13.8)	483 (14.7)	728 (13.3)	0.05
Proton-Pump Inhibitor	4050 (46.2)	1331 (40.6)	2719 (49.6)	< 0.001
Antibiotic(s) Administered				

During Hospitalization				
Penicillins	3173 (36.2)	1231 (37.6)	1942 (35.4)	0.04
Amoxicillin	69 (0.8)	45 (1.4)	24 (0.4)	< 0.001
Amoxicillin-Clavulanate	292 (3.3)	82 (2.5)	210 (3.8)	0.001
Ampicillin	66 (0.8)	41 (1.3)	25 (0.5)	<0.001
Ampicillin-Sulbactam	182 (2.1)	84 (2.6)	98 (1.8)	0.02
Piperacillin-Tazobactam	2780 (31.7)	1030 (31.4)	1750 (31.9)	0.7
Cephalosporins	4228 (48.2)	1407 (42.9)	2821 (51.4)	< 0.001
Ceftriaxone	3824 (43.6)	1278 (39.0)	2546 (46.4)	< 0.001
Cefdinir	134 (1.5)	27 (0.8)	107 (2.0)	< 0.001
Cefepime	408 (4.7)	123 (3.8)	285 (5.2)	0.002
Ceftaroline	58 (0.7)	12 (0.4)	46 (0.8)	0.008
Ceftolozane-Tazobactam	4 (0.0)	1 (0.0)	3 (0.1)	1.0
Ceftazidime-Avibactam	1 (0.0)	0 (0.0)	1 (0.0)	1.0
Carbapenems	187 (2.1)	40 (1.2)	147 (2.7)	< 0.001
Ertapenem	120 (1.4)	20 (0.6)	100 (1.8)	<0.001

Meropenem	83 (0.9)	24 (0.7)	59 (1.1)	0.1
Fluoroquinolones	3768 (43.0)	1280 (39.1)	2488 (45.3)	< 0.001
Ciprofloxacin	1863 (21.3)	661 (20.2)	1202 (21.9)	0.06
Levofloxacin	2168 (24.7)	695 (21.2)	1473 (26.8)	< 0.001
Lincosamides	369 (4.2)	155 (4.7)	214 (3.9)	0.06
Clindamycin	369 (4.2)	155 (4.7)	214 (3.9)	0.06

Abbreviation: SD, standard deviation

Chi-square statistical analysis of differences between groups

Table 2. Incidence of Hospital Onset *Clostridioides difficile* Infection for Baseline Characteristics

N (%)	Total Patients (N=8763)	HO-CDI Event (N=58)	No HO-CDI (N=8705)	p-value
Demographics				
Age >65	4631 (52.8)	34 (0.7)	4597 (99.3)	0.4
Male Sex	3390 (38.7)	25 (0.7)	3365 (99.3)	0.5
Intensive Care Unit	467 (5.3)	9 (1.9)	458 (98.1)	0.001
Concomitant Medications				
Metronidazole	1211 (13.8)	8 (0.7)	1203 (99.3)	1.0
Proton-Pump Inhibitor	4050 (46.2)	35 (0.9)	4015 (99.1)	0.03
Antibiotic(s) Administered				
Penicillins	3173 (36.2)	36 (1.1)	3137 (98.9)	< 0.001
Amoxicillin	69 (0.8)	0 (0.0)	69 (100.0)	0.5
Amoxicillin-Clavulanate	292 (3.3)	1 (0.3)	291 (99.7)	0.5
Ampicillin	66 (0.8)	0 (0.0)	66 (100.0)	0.5
Ampicillin-Sulbactam	182 (2.1)	4 (2.2)	182 (97.8)	0.01
Piperacillin-Tazobactam	2780 (31.7)	34 (1.2)	2746 (98.8)	< 0.001
Cephalosporins	4228 (48.2)	31 (0.7)	4197 (99.3)	0.4
Ceftriaxone	3824 (43.6)	24 (0.6)	3800 (99.4)	0.7
Cefdinir	134 (1.5)	1 (0.7)	133 (99.3)	0.9
Cefepime	408 (4.7)	8 (2.0)	400 (98.0)	0.001
Ceftaroline	58 (0.7)	2 (3.4)	56 (96.6)	0.009
Ceftolozane-Tazobactam	4 (0.0)	1 (25.0)	3 (75.0)	1.0

Ceftazidime-Avibactam	1 (0.0)	0 (0.0)	1 (100.0)	0.9
Carbapenems	187 (2.1)	5 (2.7)	182 (97.3)	0.001
Ertapenem	120 (1.4)	2 (1.7)	118 (98.3)	0.2
Meropenem	83 (0.9)	5 (6.0)	78 (94.0)	< 0.001
Fluoroquinolones	3768 (43.0)	28 (0.7)	3740 (99.3)	0.4
Ciprofloxacin	1863 (21.3)	14 (0.8)	1849 (99.2)	0.6
Levofloxacin	2168 (24.7)	15 (0.7)	2153 (99.3)	0.8
Lincosamides	369 (4.2)	0 (0)	369 (100.0)	0.1
Clindamycin	369 (4.2)	0 (0)	369 (100.0)	0.1
S. boulardii	5487 (62.6)	31 (0.56)	5456 (99.4)	0.2
Early *S. boulardii*	3936 (44.9)	15 (0.38)	3921 (99.6)	0.01
Late *S. boulardii*	1551 (17.7)	16 (1.03)	1535 (99.0)	0.5

Abbreviations: HO-CDI, hospital onset *Clostridioides difficile* infection

Chi-square statistical analysis for development of hospital onset *Clostridioides difficile* infection

Table 3. Risk Associations of Hospital Onset *Clostridioides difficile* Infection Determined by Unadjusted Bivariate Analysis (N=8763)

	OR (95% CI)	p-value
Demographics		
Age >65	1.27 (0.75 - 2.14)	0.4
Male	1.20 (0.71 - 2.03)	0.5
Intensive Care Unit	3.31 (1.62 - 6.77)	0.001
Concomitant Medications		
Metronidazole	1.00 (0.47 - 2.1)	1.0
Proton-Pump Inhibitor	1.78 (1.05 - 3.01)	0.03
Intervention		
S. boulardii & Antibiotic	0.68 (0.41 - 1.1)	0.2

Abbreviations: HO-CDI, hospital onset *Clostridioides difficile* infection; OR, odds risk; CI, confidence interval

Table 4. Adjusted Risk Associations of Hospital Onset *C. difficile* Infection with *S. boulardii* Using Propensity Scores in Multivariable Regression (N=8763)

	HO-CDI Event Rate N (%)	Adjusted Risk OR (95% CI)	p-value
No *S. boulardii* (antibiotic only)	27/3276 (0.82)	*Ref.*	*Ref.*
S. boulardii & Antibiotic	31/5487 (0.56)	0.57 (0.33 – 0.96)	0.035

Abbreviations: HO-CDI, hospital onset *Clostridioides difficile* infection; OR, odds risk; CI, confidence interval

Adjusted multivariable logistic regression included propensity score.

C-statistic from model used to derive propensity score = 0.620; C-statistic for adjusted risk association = 0.634

Table 5. Adjusted Risk Association of Hospital Onset *C. difficile* Infection with Early versus Late *S. boulardii* (N=8763)

	HO-CDI Event Rate N (%)	Adjusted Risk OR (95% CI)	p-value
Early *S. boulardii*	15/3936 (0.38)	0.47 (0.23 – 0.97)	0.041
Late *S. boulardii*	16/1551 (1.0)	*Ref.*	*Ref.*

Abbreviations: HO-CDI, hospital onset *Clostridioides difficile* infection; OR, odds risk; CI, confidence interval

Adjusted multivariable logistic regression included propensity score.

C-statistic from model used to derive propensity score = 0.616; C-statistic for adjusted risk association = 0.717