



Small Intestinal Bacterial Overgrowth in Irritable Bowel Syndrome: Frequency and Microbiological Insights from Duodenal Aspirate Analysis

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Abstract

Background: Small Intestinal Bacterial Overgrowth (SIBO) has been implicated in the pathophysiology of Irritable Bowel Syndrome (IBS), particularly in diarrhea-predominant (IBS-D) and mixed-type (IBS-M) subtypes. This study aimed to determine the frequency of SIBO and analyze the microbiological profile of duodenal aspirate cultures in IBS patients.

Methods: This observational cross-sectional study conducted at department of Gastroenterology, Dhaka medical college hospital, Dhaka; from September 2018 to August 2019 and included 104 IBS patients diagnosed based on Rome IV criteria. Duodenal aspirate samples were collected and cultured, with SIBO defined as $\geq 10^5$ CFU/mL of bacterial growth. The frequency of SIBO was compared between IBS subtypes, and the isolated bacterial species were identified.

Results: SIBO was detected in 38 (36.5%) of IBS patients, with a higher prevalence in IBS-D (53.3%) compared to IBS-M (13.7%). Culture positivity was observed in 60.6% of cases. *Pseudomonas* (78.9%) was the most frequently isolated organism in SIBO-positive patients, followed by *E. coli* (21.1%). In contrast, *Klebsiella* and *Citrobacter* were only found in SIBO-negative cases.

Conclusion: SIBO was significantly associated with IBS-D, suggesting a potential pathogenic role of *Pseudomonas* and *E. coli* in symptom development. The findings emphasize the importance of screening for SIBO in IBS-D patients for targeted therapeutic interventions. Further large-scale studies are needed to validate these observations.

Keywords: IBS; SIBO; Duodenal Aspirate Culture; *Pseudomonas*; *E. coli*; Gut Dysbiosis.

Introduction

Irritable bowel syndrome (IBS) is one of the most common functional gastrointestinal disorders, characterized by recurrent abdominal pain associated with altered bowel habits in the absence of any identifiable organic pathology [1]. According to the Rome IV criteria, IBS is classified into different subtypes, including diarrhea-predominant IBS (IBS-D), constipation-predominant IBS (IBS-C), and mixed-type IBS (IBS-M) [2]. Despite its high prevalence worldwide, the exact pathophysiology of IBS remains incompletely understood, with proposed mechanisms including altered gut motility, visceral hypersensitivity, low-grade inflammation, gut-brain axis dysfunction, and dysbiosis of the gut microbiota [3]. In the past few years, small intestinal bacterial overgrowth (SIBO) has been advocated as a

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potential contributor to the pathogenesis of IBS, particularly IBS-D [4]. SIBO is a pathological increase in the quantity and/or diversity of bacteria in the small intestine, typically more than 10^5 colony-forming units per milliliter (CFU/ml) [5]. This bacterial overgrowth disrupts normal digestion and leads to symptoms such as bloating, abdominal pain, diarrhea, and malabsorption, many of which overlap with IBS symptoms [6]. The relationship between IBS and SIBO has been examined in numerous studies with varying results, depending in large part on variations in diagnostic techniques, study populations, and geographic regions [7].

Diagnosis of SIBO remains challenging, and the diagnostic techniques available vary from non-invasive breath tests to direct culture of small bowel aspirates [3]. Although breath tests, namely lactulose and glucose breath tests, are in vogue due to their ease and non-invasiveness, they are limited by low sensitivity and specificity [8]. Duodenal aspirate culture, on the other hand, is considered the gold standard for SIBO diagnosis since it provides direct microbial identification and quantitative assessment of bacterial load [9]. However, it is rarely performed in routine clinical practice due to its invasiveness and logistic challenges. In Bangladesh, data on the prevalence and microbiological characteristics of SIBO in patients with IBS are extremely limited. Information on the prevalence and microbial spectrum of SIBO in IBS patients from this region is crucial for improving diagnostic accuracy and formulating region-specific effective management strategies [10]. Identification of specific bacterial species implicated in SIBO in IBS also has the potential to guide the development of future therapeutic approaches, i.e., the use of targeted antibiotics or probiotics [7].

The present study was carried out to determine the prevalence of SIBO among IBS-D and IBS-M patients by duodenal aspirate culture and examine the microbiological pattern of isolated bacteria. By analyzing demographic, clinical, and microbial characteristics of IBS patients, this study aimed to contribute to the understanding of the role of SIBO in IBS pathogenesis, particularly in a Bangladeshi population. The findings could pave the way for improved diagnosis and more personalized treatment approaches for IBS patients, especially those with evidence of SIBO.

Methodology and Materials

This observational cross-sectional study conducted at department of Gastroenterology, Dhaka medical college hospital, Dhaka; from September 2018 to August 2019, involving 104 adult patients diagnosed with irritable bowel syndrome (IBS) based on Rome IV criteria. Eligible participants included patients aged 18 years or older with IBS-D (diarrhea-predominant) or IBS-M (mixed-type) subtypes. Patients with IBS-C, organic gastrointestinal disorders, recent use of antibiotics, PPIs, probiotics, or

motility-altering drugs within the last eight weeks, history of major abdominal surgery, or pregnancy were excluded from the study. After obtaining informed consent, detailed demographic and clinical data were collected using a structured questionnaire. Upper gastrointestinal endoscopy was performed under standard aseptic precautions, and approximately 3-5 ml of duodenal aspirate was collected from the second part of the duodenum using a sterile catheter. The aspirate was immediately transported to the microbiology laboratory for culture and colony count analysis. Quantitative aerobic culture was performed using blood agar and MacConkey agar plates, incubated at 37°C for 24 to 48 hours. The presence of $\geq 10^5$ colony-forming units per milliliter (CFU/ml) was considered diagnostic for small intestinal bacterial overgrowth (SIBO). Identification of bacterial isolates was performed using standard biochemical tests. All collected data, including demographic variables, IBS subtype, smoking status, and culture results, were recorded and analyzed using SPSS software (version 25). Categorical variables were expressed as frequencies and percentages, and continuous variables were presented as mean \pm standard deviation. Comparisons between SIBO and non-SIBO groups were made using chi-square tests for categorical variables and independent t-tests for continuous variables. A p-value of less than 0.05 was considered statistically significant.

Results

Table I: Demographic, Clinical and microbiological profile of study participants.

Variables	IBS patients (n =104)
Age (Mean \pm SD)	31.69 \pm 10.01
Sex (Male)	78(75%)
Occupation (service holder)	47(45%)
Urban	61(58.6%)
Nonsmoker	77(74%)
IBS-D	60(57%)
IBS-M	44(42%)
Culture positivity	63(60.6%)
SIBO	38 (36.5%)

Table II: Distribution of the participants by the presence of SIBO (n=104).

SIBO	Frequency (n)	Percentage (%)
Present	38	36.5
Absent	66	63.5
Total	104	100

Table I shows mean age of the study population was 31 years and male were predominant participants. Almost half of the participants were service holders. Most of the participants from urban areas and 3/4th of the participants were nonsmokers. IBS-D participants were more than IBS-M and SIBO, which was positive in 36.5% cases.

Table II shows total 38/104 (36.5%) patients with IBS-D and IBS-M had SIBO (colony count $\geq 10^5$ CFU/ml) on duodenal aspirate culture.

Table III showed the Frequency of SIBO is more in IBS-D patients than IBS-M patients.

Table III: Frequency of SIBO in different types of IBS (n=104)

Types of IBS	No. of patients (104)	SIBO n (%)
IBS-D	60(57.7%)	32(53.3%)
IBS-M	44(42.3%)	6(13.7%)

Table IV: Isolated bacteria on culture of duodenal aspirate among participants (n=104)

Spectrum of bacteria isolated on culture	SIBO (n=38)	Non SIBO (n=66)
No growth	0 (0.0)	41 (62.1%)
Pseudomonas	30 (78.9%)	20 (30.3%)
E. Coli	8 (21.1%)	8 (12.1%)
Klebsiella	0 (0.0%)	1 (1.5%)
Citrobacter	0 (0.0%)	1 (1.5%)

In Table IV, Culture (duodenal aspirate) of 104 respondents showed colonies of several gram-negative organisms. Pseudomonas was the most prevalent organism and klebsiella, Citrobacter are only found in non SIBO patients.

Discussion

The mean age of the patients was 31.69 ± 10.01 years, which indicates that IBS is usually presented in young adults. The preponderance of male (75%) is consistent with the results of previous studies by Thompson et al., where they documented a higher prevalence of IBS-D among men [11]. Almost half of the patients were service holders (45%), and a majority resided in urban areas (58.6%), showing that lifestyle and dietary factors might be accountable for IBS prevalence, as revealed by Lu et al [12]. It was also observed that the majority of the participants were nonsmokers (74%), while there have been research studies suggesting smoking as a risk factor for gut dysbiosis [13].

Among the 104 IBS patients in this study, 38 (36.5%) were diagnosed with SIBO based on quantitative culture of duodenal aspirate, where a bacterial load of $\geq 10^5$ CFU/

mL was considered positive. This is in line with the work of Pimentel et al., who reported SIBO prevalence of 30-40% among IBS patients [14]. The majority of SIBO-positive patients were IBS-D (32/60; 53.3%), whereas few IBS-M patients (6/44; 13.7%) were SIBO-positive. These results highlight a close association between SIBO and IBS-D, favoring the hypothesis that bacterial overgrowth may be involved in diarrhea-predominant IBS symptoms, as previously hypothesized by Ghoshal et al [15]. Sixty percent (60.6%) of high IBS patient percentages were positive in culture, indicating the development of bacterial growth in duodenal aspirate samples. Every culture-positive case did not hit the SIBO criteria $\geq 10^5$ CFU/mL, suggesting there may be alternative gut microbiota in some IBS patients but not hitting SIBO criteria. This is in keeping with findings by Rezaie et al., who emphasized that dysbiosis in IBS is not always associated with SIBO but can still contribute to symptom induction [16].

Microbiological analysis isolated Pseudomonas as the most frequent isolated pathogen, isolated in 78.9% of samples positive for SIBO. This suggests that Pseudomonas are likely a player in SIBO pathophysiology in the IBS patient population, consistent with findings of Sabaté et al., in which a high duodenal aspirate percentage of Pseudomonas among SIBO patients was observed [17]. Notably, Pseudomonas was found in 30.3% of SIBO-negative patients, potentially reflective of subclinical bacterial overgrowth or colonization below the diagnostic threshold. The second most common isolate was E. coli, in 21.1% of SIBO-positive and 12.1% of SIBO-negative patients, as with previous results by Ford et al., linking E. coli overgrowth to IBS symptoms [10]. Klebsiella and Citrobacter were isolated only in SIBO-negative patients, suggesting that these organisms are unlikely to be the primary causes of SIBO in IBS. The absence of bacterial overgrowth in a high proportion of SIBO-negative patients (41/66; 62.1%) suggests that most IBS symptoms might not be the direct result of bacterial overgrowth but could be caused by other factors such as gut motility disorders, visceral hypersensitivity, or immune dysfunction [18]. This also highlights the limitation of duodenal aspirate culture to capture the spectrum of gut dysbiosis because standard culture methods are not likely to detect anaerobic bacteria that are known to be resident in the small intestine [16].

Our results reinforce the link between SIBO and IBS-D as indicated by the significantly higher prevalence of SIBO in IBS-D compared to IBS-M. Prevalence of Pseudomonas and E. coli among SIBO patients suggests that gram-negative bacteria are most likely to be the key pathogenic players in IBS-related SIBO. The results agree with other research that has detected similar patterns of bacteria among SIBO patients, though differences exist due to geographic, food, and methodological differences [19]. Generally, this study

indicates the heavy burden of SIBO among IBS patients, and even more so IBS-D. The high prevalence of *Pseudomonas* and *E. coli* in SIBO-positive patients suggests an implication of a potential pathogenic function of these bacteria in symptom generation. Further studies with more numbers, inclusion of anaerobic culture techniques, and use of non-invasive breath tests might further clarify SIBO in IBS patients. Furthermore, evaluating the impact of antibiotic or probiotic treatment against SIBO-related organisms can offer new therapeutic avenues for the management of IBS symptoms in patients [4].

Limitations of the study

This study was conducted at a single center, limiting the generalizability of findings to the broader population. Additionally, the small sample size may not fully capture the diversity of IBS patients, and the culture-based method used may have underestimated the presence of anaerobic bacteria involved in SIBO.

Recommendations

Future large-scale, multicenter studies are needed to assess the true prevalence and risk factors of SIBO in IBS, particularly IBS-D. The use of advanced diagnostic techniques, such as breath tests or molecular methods, may improve detection rates. Clinicians should consider routine SIBO screening in IBS-D patients to facilitate early intervention and improve symptom management.

Conclusion

This study highlights a significant association between SIBO and IBS-D, notably with *Pseudomonas* and *E. coli* as the predominant bacterial isolates. Given the potential impact of bacterial overgrowth on symptom severity, targeted diagnostic and therapeutic strategies should be prioritized to improve patient outcomes.

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