

## Article

# *Blastocystis* sp. Infection: Prevalence and Clinical Aspects among Patients Attending to the Laboratory of Parasitology–Mycology of Fann University Hospital, Dakar, Senegal

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**Abstract:** Introduction: *Blastocystis* sp. is a unicellular obligate anaerobic protozoa found in the human intestinal tract. Its role in human health is widely discussed because of the high proportion of asymptomatic carriers. In sub-Saharan Africa, the prevalence of the disease is underestimated. This study was performed to describe the epidemiological, clinical, and parasitological aspects of *Blastocystis* sp. infection in patients attending to Fann University Hospital. Material and Methods: We carried out a retrospective and descriptive study among patients attending to the laboratory of Parasitology and Mycology of Fann University Hospital from January 2016 to December 2020. All stool samples collected were examined using direct examination, a formal ether concentration method, and a modified Zeilth–Nielsen staining method. A descriptive analysis was performed with Stata MP 16 software. The significance level was set at 5%. Results: Overall, 447 cases of *Blastocystis* sp. were reported in our study, representing a prevalence rate of 13.7% ((447/3264) (95% CI: 12.5–15.5)). The mean age of the patients was 26 ± 20.7 years. Subjects over 45 years of age were more affected, with a frequency of 14.7%. *Blastocystis* sp. carriage was more common in males, at 14.6%. The symptomatology was mainly represented by diarrhea, abdominal pain, and dyspeptic disorders. In asymptomatic patients, the frequency of *Blastocystis* sp. was 33.3%. Mono-infection was found in 78.6% of cases. In total, 96 patients were carriers of at least two parasites (21.5%). *Blastocystis* sp. was most associated with *Entamoeba coli* (8.1%) and *Endolimax nanus* (4.03%). The association with helminths was noted in 5 patients (3 *Ascaris lumbricoides*, *Trichuris trichiura*, and *Taenia*). Conclusion: These results show the frequency of *Blastocystis* sp. infection with a large proportion of asymptomatic carriers. The presence of the parasite in the stool, associated with digestive disorders or with the association of other intestinal parasites, could justify the initiation of an anti-parasitic treatment.

**Keywords:** *Blastocystis* sp.; epidemiology; clinical; Senegal

## 1. Introduction

*Blastocystis* sp., which is commonly found in the gastro-intestinal tract, has been recognized as a non-pathogenic organism for a few decades. The parasite has a worldwide distribution. A high prevalence has been reported in developing countries (22.1–100%). In industrialized countries, the prevalence ranges from 0.5% to 23.1% [1–5]. The high prevalence noted in developing countries is related to poor hygiene, low access to safe water and food, and a lack of standard sanitary facilities [6,7].



The pathogenicity of *Blastocystis* is controversial because most patients carrying this parasite are asymptomatic. Several studies have considered *Blastocystis* sp. as a commensal micro-organism, while other results have showed the pathogenicity of the parasite. In addition, several epidemiological studies have reported a high prevalence of *Blastocystis* in patients with irritable bowel syndrome (IBS) [8]. The presence of the parasite is often linked to symptoms such as nausea, fever, urticaria, vomiting, anorexia, diarrhea, cramps, flatulence, discomfort, and abdominal pain [9,10]. The illness may be acute or chronic, with symptoms persisting for several years [11].

However, most studies have reported that *Blastocystis* sp. was frequently found in immunocompromised patients such as kidney transplant patients, patients with hematologic malignancies, and HIV patients with a CD4 <200/mm<sup>3</sup> [12–14].

The data available in Africa concerning *Blastocystis* are still limited. In Zambia, 53.8% prevalence of *Blastocystis* sp. was observed by Thaddeus et al. [15]. The most common parasite noted in Gabon was *Blastocystis* sp., with 48.6% prevalence [16]. These results are similar with general distribution of *Blastocystis* in developing countries.

In Senegal, low prevalence rates of *Blastocystis* sp. were reported of 4.8% and 3.7% in Dakar (capital city) and rural areas, respectively [17,18].

In order to improve the management of *Blastocystis* infection, an update of the data is necessary. It was in this context that we conducted this study, with the objective of evaluating the prevalence of *Blastocystis* sp.

## 2. Results

### 2.1. Socio-Demographic Characteristics of Study Participants

A total of 3264 patients were included in the study. The mean age of the study participants was  $26.3 \pm 20.7$  years. Patients under 15 years of age were most represented (41.8%), followed by patients with ages ranging from 15 to 30 years (21.5%). Patients in the age group of 30–45 years and those over 45 years represented 16.7% and 20% of the total, respectively. According to gender, the study population was predominantly male (50.9%). The sex ratio was 1.04. The numbers of samples received were higher in 2018 and 2017 at 24.6% and 24.5%, respectively. Regarding the season, the number of samples was higher in the dry season at 78.8% compared to 24.2% in the rainy season (Table 1).

**Table 1.** Socio-demographic characteristics of study participants (N = 3264).

Variable	Frequency (n)	Percentage (%)	95% CI
<b>Year</b>			
2016	672	20.6	19.1–22.2
2017	801	24.5	22.8–26.3
2018	804	24.6	22.9–26.3
2019	616	18.8	17.4–20.4
2020	371	11.4	10.2–12.6
<b>Age group</b>			
<15 years	1365	41.8	39.6–44.1
[15–30]	703	21.5	19.9–23.2
[30–45]	545	16.7	15.3–18.2
≥45 years	651	20	18.4–21.5
<b>Gender</b>			
Female	1600	49.1	46.6–51.5
Male	1664	50.9	48.5–53.5
<b>Season</b>			
Rainy	2475	78.8	72.8–78.8
Dry	789	24.2	22.5–25.9
<b>Total</b>	3264	100	



## 2.2. Clinical Characteristics of Study Patients

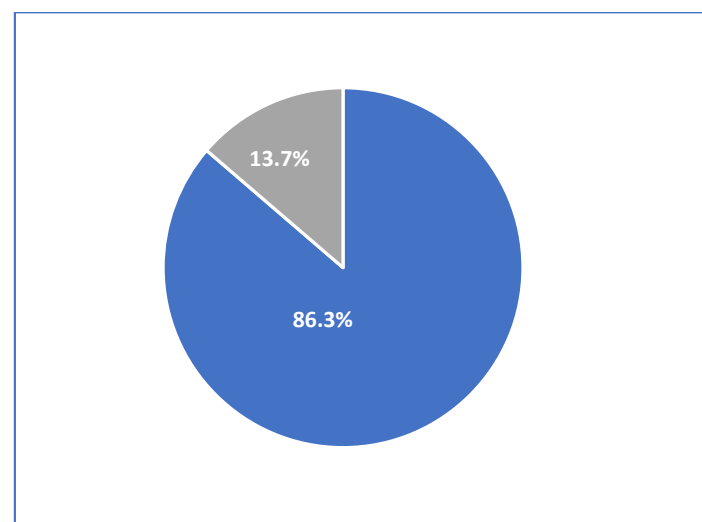
According to the symptoms, 53.2% (1737/3264) of the patients were symptomatic, while 46.8% of the study participants were asymptomatic. Among the symptomatic patients, diarrhea, abdominal pain, fever, and dyspeptic disorders were the most frequently noted symptoms. However, several types of diarrhea were observed, such as acute febrile diarrhea (4.7%), acute non-febrile diarrhea (16.4%), chronic diarrhea in HIV-positive patients (9.1%), and chronic diarrhea in HIV-negative patients (23%). The other symptoms were represented by abdominal pain (20.9%), fever (8.2%), dyspeptic disorders (8.2%), anal pruritus (0.8%), dysenteric syndrome (3.5%), urticaria (1.2%), and vomiting (0.8) (Table 2).

**Table 2.** Clinical symptoms of study participants (N = 1737).

Clinical Symptom	Frequency (n)	Percentage (%)
Constipation	16	0.9
Acute febrile diarrhea	82	4.7
Acute non-febrile diarrhea	285	16.4
Chronic diarrhea (HIV-positive)	157	9.1
Chronic diarrhea (HIV-negative)	400	23
Abdominal pain	364	20.9
Hypereosinophily	5	0.3
Malnutrition	6	0.3
Clinical anemia	25	1.4
Anal pruritus	15	0.8
Dysenteric syndrome	60	3.5
Fever	143	8.2
Dyspeptic disorders	145	8.2
Urticaria	20	1.2
Vomiting	14	0.8
<b>Total</b>	<b>1737</b>	<b>100</b>

## 2.3. Prevalence of *Blastocystis* sp. Infection

Overall, 13.7% (447/3264) of the study participants were found to be infected by *Blastocystis* sp. (Figure 1). Mono-infection was noted in 351 patients (78.6%). Di-parasitism (presence of two parasites) and poly-parasitism (more than two parasites) were found in 21.5% and 3.6%, respectively. In 98.6% of cases the protozoa were isolated alone, and in 1.6% of cases they were associated with helminths. The vacuolar form was the most common form, representing 87.7% of *Blastocystis* sp. isolates, while the granular form was noted in 12.3% of cases (Table 3).



**Figure 1.** Prevalence of *Blastocystis* sp. infection.



The main associations of the species were *Blastocystis* sp. + Cyst *Entamoeba coli* (8.1%), *Blastocystis* sp. + Cyst *Endolimax Nana* (4%), *Blastocystis* sp. + Trophozoite *Entamoeba coli* (2.9%), and *Blastocystis* sp. + *Ascaris lumbricoides* (0.8%) (Table 4).

**Table 3.** Parasitological aspects of *Blastocystis* sp. infection.

	Frequency (n)	Percentage (%)	95% CI
<b>Parasitism</b>			
Mono-parasitism	351	78.6	70.5–87.2
Di-parasitism	96	21.5	17.4–26.2
Poly-parasitism	16	3.6	2.1–5.8
<b>Type of parasite</b>			
Protozoa	441	98.6	89.6–99.9
Protozoa—Helminth	6	1.6	0.4–2.9
<b>Parasite form</b>			
Vacuolar	392	87.7	79.2–96.8
Granular	55	12.3	9.3–16

**Table 4.** Intestinal parasite species associated with *Blastocystis* sp.

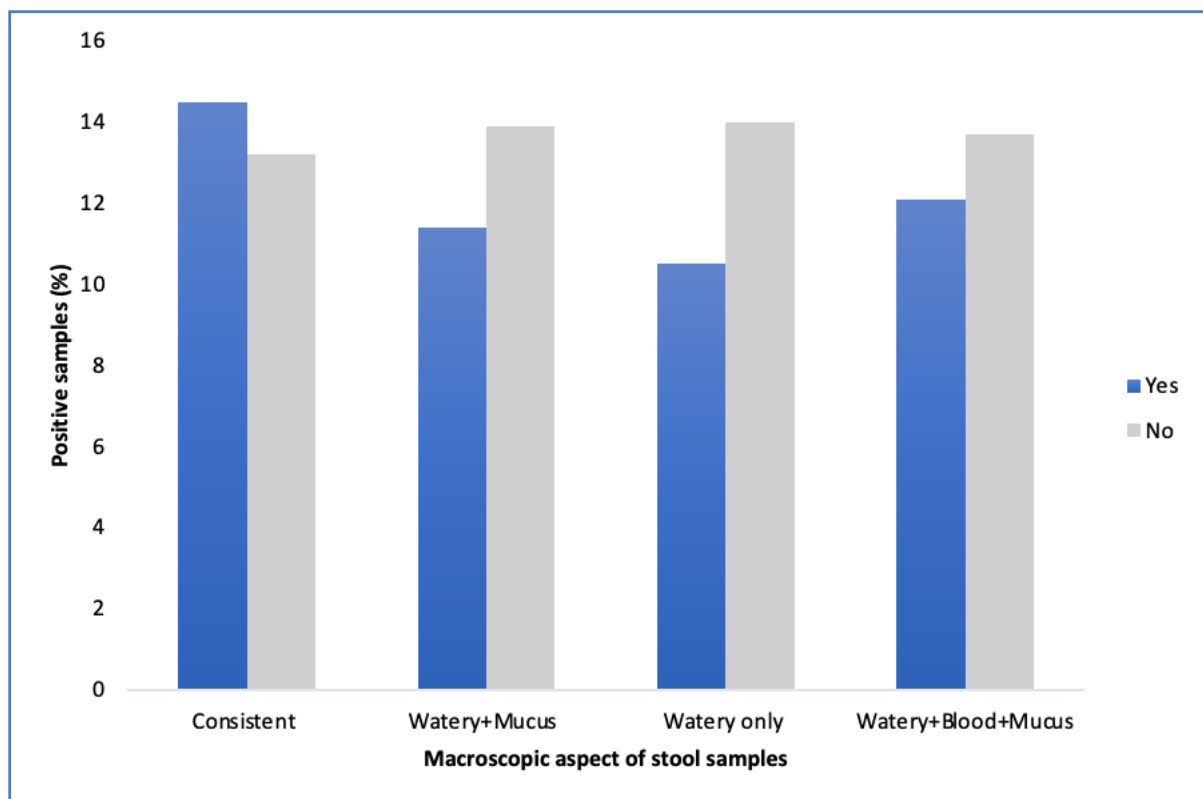
Parasites Species	Frequency (n)	Percentage (%)
<b>Di-parasitism</b>		
<i>Blastocystis</i> sp. + Cyst <i>Entamoeba coli</i>	36	8.1
<i>Blastocystis</i> sp. + Cyst <i>Endolimax nana</i>	18	4
<i>Blastocystis</i> sp. + Trophozoite <i>Entamoeba coli</i>	13	2.9
<i>Blastocystis</i> sp. + Cyst <i>Giardia intestinalis</i>	12	2.7
<i>Blastocystis</i> sp. + <i>Ascaris lumbricoides</i>	3	0.8
<i>Blastocystis</i> sp. + <i>Trichuris trichiura</i>	1	0.2
<i>Blastocystis</i> sp. + <i>Teniasis</i>	1	0.2
<b>Polyparasitism</b>		
<i>Blastocystis</i> sp. + Trophozoite <i>E. coli</i> + Cyst <i>E. coli</i>	6	1.3
<i>Blastocystis</i> sp. + Cyst <i>E. nana</i> + Cyst <i>E. coli</i>	2	0.4
<i>Blastocystis</i> sp. + <i>Pseudolimax Butschli</i> + Cyst <i>E. coli</i>	2	0.5
<i>Blastocystis</i> sp. + Trophozoite <i>E. coli</i> + Cyst <i>E. coli</i> + <i>Trichomonas. intestinalis</i>	1	0.2
<i>Blastocystis</i> sp. + Cyst <i>G. intestinalis</i> + Cyst <i>E. coli</i>	1	0.2
<i>Blastocystis</i> sp. + <i>T. intestinalis</i> + Cyst <i>E. coli</i>	1	0.2

According to the macroscopic aspect of the stool specimens, in patients with consistent stools, the *Blastocystis* sp. prevalence was 14.7%. In patients who had watery stools, it was 14.5%. The prevalence of *Blastocystis* sp. infection was 12.1% in patients with watery stools with blood and mucus (Figure 2).

#### 2.4. Prevalence of *Blastocystis* sp. according to Socio-Demographic and Clinical Characteristics of Study Participants

Stratifying through the year of sample collection, the *Blastocystis* sp. infection rates were more frequent in 2017 and 2020 at 18.7% and 16.9%, respectively. A low positive rate was noted in 2016 (8.9%). The frequency of *Blastocystis* sp. was more important in patients aged over 45 years (14.7%). In patients under 15 years of age, the *Blastocystis* sp. prevalence was 13.9%. This prevalence was lower in the 30–45-year-old age group (12.5%). However, the differences were not significant between age groups ( $p = 0.65$ ). Infection with *Blastocystis* sp. was more frequent in male patients (14.6%) than women (12.7) ( $p$  value = 0.12). According to the season, *Blastocystis* sp. was more common in the rainy season (15.7%) than in the dry season (13.9%) ( $p = 0.05$ ). In total, 16.1% of asymptomatic patients had *Blastocystis* sp. in their stool samples versus 11.6% of symptomatic patients, with a significant difference ( $p < 10^{-3}$ ) (Table 5).





**Figure 2.** Prevalence of *Blastocystis* sp. according to the macroscopic aspect of stool specimens.

**Table 5.** Prevalence rates of *Blastocystis* sp. according to socio-demographic and clinical characteristics of study participants (N = 447).

Variable	Frequency (n)	Percentage (%)	95% CI	p Value
<b>Year</b>				
2016	80	8.9	6.8–11.5	<10 <sup>−3</sup>
2017	150	18.7	15.8–21.9	
2018	89	11.1	8.8–13.6	
2019	86	13.9	11.2–17.2	
2020	62	16.7	12.8–21.4	
<b>Age group</b>				
<15 years	191	13.9	12.1–16.1	0.65
[15–30]	92	13.1	10.5–16.1	
[30–45]	68	12.5	9.6–15.8	
≥45 years	96	14.7	11.9–18	
<b>Gender</b>				
Female	204	12.7	11.1–14.6	0.12
Male	243	14.6	12.8–15.5	
<b>Season</b>				
Rainy	124	15.7	13.1–18.7	0.06
Dry	323	13.1	11.6–15.5	
<b>Symptom</b>				
Asymptomatic	246	16.1	14.2–18.2	<10 <sup>−3</sup>
Symptomatic	201	11.6	10–13.2	

In symptomatic patients, the prevalence of *Blastocystis* sp. was 9.76% in patients with acute febrile diarrhea and 8.71% in patients with acute non-febrile diarrhea. In patients with chronic diarrhea, *Blastocystis* sp. was observed in HIV-positive and -negative patients at rates of 9.75% and 5.1%, respectively. High positivity rates were noted in patients with



abdominal pain (15.9%) and those with dyspeptic disorders (17.2). In patients with fever and dysenteric syndrome, the prevalence rates were 14.7% and 8.3%, respectively.

### 3. Discussion

*Blastocystis* sp. is the most common anaerobic parasite to colonize the gastrointestinal tracts of humans as well as many animals. It can be associated with gastro-intestinal disorders that are not specific to it and seems to be involved in some functional disorders, especially in patients suffering from irritable bowel syndrome and in immunocompromised subjects. In order to improve the management of patients and to update the epidemiology of the disease, this study was conducted among patients attending to the Laboratory of Parasitology of Fann University Hospital.

The results of this study showed 13.7% prevalence of *Blastocystis* sp. infection. Previous studies conducted in Senegal have described the frequency of blastocystosis. Sow et al., when assessing the prevalence rates of *Blastocystis* sp. infection from 2011 to 2013 in the same laboratory, showed 4.8% prevalence [17]. The same trends were described by Sylla et al. when evaluating the epidemiological profile of intestinal parasitic infection among preschool and schoolchildren living in a rural community, where 3.7% prevalence was observed [16]. These lower frequencies compared to our study could be explained by the long period of our study and the sample size. Low prevalence was also noted in Tunisia by Trabelsi et al., who showed a prevalence rate of *Blastocystis* of 7.2% [19].

Other studies conducted at the community level have noted high prevalence rates of *Blastocystis* infection. Niang et al., when evaluating the performance of the KOPCOLOR kit for the detection of intestinal parasites in November 2012 in 117 children living in the Thiès region (Central part of Senegal), found a prevalence rate of 87.1% for intestinal parasites, with *Blastocystis* sp (58.1%) as the main species observed [20]. El Safadi et al., in evaluating *Blastocystis* sp. via molecular method in 93 children aged 6–10 years in 2011 in the Senegal River Basin, showed a prevalence rate of 100% for *Blastocystis* sp. [21].

In Zambia, 53.8% prevalence of *Blastocystis* sp. was observed by Thaddeus et al. [15]. The most common parasite noted in Gabon was *Blastocystis* sp. with 48.6% prevalence [16].

Several studies using molecular biology or other techniques as diagnostic tools have shown higher frequencies. For example, in a study comparing the Flotac dual 400 technique with the conventional concentration technique in 108 patients between May and June 2009, Becker et al. observed a prevalence of 20.4% [22]. Poulsen et al. showed 84% prevalence of *Blastocystis* sp. in a molecular biology study in 199 Nigerian children in 2013 [23].

In Tanzania, Forsell et al., when studying the distribution of *Blastocystis* sp. subtypes and the prevalence of intestinal parasites using molecular methods and microscopy, showed 61% prevalence of *Blastocystis* via PCR. The prevalence revealed via microscopy was lower (20%) [24].

Other studies have demonstrated that *Blastocystis* is an emergent pathogen, with frequency rates ranging from 3% to 14.5% in France, and from 0.5% in Japan to 60% in Malaysia [2,25–27].

The results from this study showed that the frequency of *Blastocystis* sp. was more important in patients under 15 years old and those over 45 years (13.9% and 14.7%, respectively). Sow et al. found similar results, with higher prevalence in subjects under 15 years and in the age group of 5–30 years [18]. A high prevalence (84%) of *Blastocystis* sp. infection was noted in children aged 2–14 years in Nigeria [23]. In Libya, Abdul Salam et al. found that adults (aged  $\geq 18$  years) were more affected than other age groups ( $< 18$  years) (29.4% vs 9.9%;  $p < 0.001$ ) [28]. A study conducted in Angola by Dacal et al. in Benguela province in 2015 showed that *Blastocystis* sp. is one the most common protozoa parasites detected (25.6%; 95% CI: 21.18–30.2%). Children of all ages are affected [29]. The last two studies used molecular methods for the detection of the parasite.

According to gender, men were more affected (14.6%) by *Blastocystis* sp. than women (12.7%) in our study. Our results are in line with what were found by Angolo and Malaysia [1,30]. In the study conducted by Sow et al. in Senegal, *Blastocystis* sp. in-



fection was more frequent in women [17]. However, several studies have noted that gender has no significant influence on the carriage of *Blastocystis* sp. [31,32].

The pathogenicity of *Blastocystis* sp. remains highly controversial due to the presence of asymptomatic carriers. In our study, the prevalence rates of *Blastocystis* sp. in asymptomatic patients were 16.1% and 11.6% in symptomatic patients ( $p < 10^{-3}$ ). Similar results were previously described. In the study performed by Sow et al., 50% of patients were asymptomatic and 32.1% were symptomatic [18]. Otherwise, in several studies, the presence of gastro-intestinal symptoms has been related to the presence of *Blastocystis* sp. This was demonstrated by Abdulsalam et al., who showed that the prevalence of gastrointestinal symptoms is higher in subjects with *Blastocystis* infection compared to those without *Blastocystis* infection (35.3% vs. 13.2%,  $p$  value  $< 0.001$ ) [28]. The same trends were observed in another previous study conducted in Italia [33].

The main symptoms noted in our study were diarrhea, abdominal pain, and dyspeptic disorders (flatulence). This is in line with results found by Abdulsalam et al., who noted that the most common symptoms in their study were abdominal pain (76.4%), flatulence (41.1%), and diarrhea (21.5%) [29]. Sebaa et al., in their study, noted that the most frequent clinical signs were abdominal pain (39.1%), diarrhea (6.5%), and nausea (3.3%) [34]. Other authors have noted that *Blastocystis* sp. is directly responsible for gastrointestinal disorders and is considered the etiological agent responsible for a certain type of persistent diarrhea, abdominal pain, and vomiting [2,21,35–37]. In immunocompetent subjects, the presence of *Blastocystis* is not correlated with gastrointestinal symptoms. This was previously demonstrated by Leder et al. [38]. Otherwise, immunocompromised subjects (cancer, HIV) seem to be more receptive to the parasite [39–41].

Even if the previous studies could not draw conclusions regarding the pathogenicity of *Blastocystis* sp., it is important to look for it and to report it to the clinicians, who will decide whether to treat the patient or not according to the clinical symptoms.

According to the results of this study, *Blastocystis* sp. prevalence was higher in the rainy season (15.7%) compared to dry season (13.1%). This is consistent with the data from literature showing that *Blastocystis* sp. infestation is more common in summer. Suresh et al., when comparing the methods for detecting *Blastocystis* sp., showed that infections were more prevalent in summer than in winter or spring [42].

## 4. Materials and Methods

### 4.1. Study Design and Population

We carried out a retrospective and descriptive study at the Parasitology and Mycology Laboratory at Fann University Hospital in Dakar. From January 2016 to December 2020, all patients attending to the laboratory for a parasitological examination of stool samples were included in the study.

### 4.2. Data Collection

Data on the sociodemographic, clinical, and biological aspects from patients were collected using the laboratory records. The following variables were collected: age, gender, year, season, clinical indication, macroscopic aspect of sample, and parasitological results. Age was defined as 4 categories: less than 15 years, [15–30 years], [30–45 years], and more than 45 years. The season was defined as either the dry season (October to June) or the rainy season (July to September).

### 4.3. Parasitological Examination

Fresh fecal specimens were collected into a wide-mouth screw-cap clean containers. The stool samples were examined macroscopically for color, consistency, blood, mucus, pus, and large worms. A portion of each of the stool samples was processed, with a direct examination by light microscopy to detect cysts, trophozoites, eggs, and larvae. The remaining parts of the stool samples were examined using a modified Ritchie technique and modified Ziehl–Neelsen technique.



#### 4.4. Data Analysis

After data collection, the data were entered in Excel software and the analysis was performed using Stata software version 16. A descriptive analysis was performed. The quantitative variables were described in terms of the means and standard deviations. For the descriptive data, a description in terms of the frequency with a 95% confidence interval was used. The significance level of the different tests was set to 5% for two sides.

#### 4.5. Ethics Considerations

This study was conducted according to the Declaration of Helsinki and existing national legal and regulatory requirements. To respect their confidentiality, an identification code was given to each participant

### 5. Conclusions

The results of this study have demonstrated the frequency of *Blastocystis* sp. infection in patients attending to Fann Hospital in Dakar (Senegal). Children under 15 years and adults over 45 years old were more affected. Gender has no influence in *Blastocystis* sp. carriage. *Blastocystis* was found in symptomatic and asymptomatic patients. The main clinical symptoms were diarrhea, abdominal pain, and dyspeptic disorders. The presence of the parasite in the stool, associated with digestive disorders or with the association of other intestinal parasites, could justify the initiation of an anti-parasitic treatment. Other studies assessing risk factors and using molecular methods will be necessary to better understand the dynamics of the transmission and to better characterize the parasite.

**Author Contributions:** K.S. conceived and designed the study. K.S. supervised the data collection. K.S. analyzed the data and wrote the first draft of the manuscript. D.S., S.L., T.D., R.C.T., B.F. read and approved the final manuscript. All authors have read and agreed to the published version of the manuscript.

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**Institutional Review Board Statement:** Data are routinely collected from patients who attend to the hospital for biomedical testing. Permission to use the data for publication was requested from the administration of Fann University Hospital, which is a national reference hospital. A code was given to each enrolled participant and data on their sociodemographic characteristics, clinical aspects, and parasitological findings were collected from the laboratory records.

**Informed Consent Statement:** Informed consent was not required.

**Data Availability Statement:** The data used for this research article are available from the corresponding author upon request.

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**Conflicts of Interest:** The authors declare that they have no competing interests.

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