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PREVALENCE OF INTESTINAL PROTOZOAN PARASITIC INFECTIONS AMONG PEOPLE ATTENDING SEBHA CENTRAL LABORATORY IN SEBHA, LIBYA: A RETROSPECTIVE STUDY

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Abstract:-

Intestinal d protozoan parasites are still major public health problems in the world, particularly in developing countries. This retrospective study was conducted using data obtained from the Department of Parasitology of Sebha Central Laboratory in Sebha, Libya. A total of 740 stool samples were recorded from January to December, 2017. All samples were examined by direct wet smears using normal saline and Lugol's iodine. The objective of this study was to determine the different types of intestinal protozoan in Sebha city. The results were analyzed using SPSS version 20.0, the Chi-square test was used to measure the statistical significant differences at level p<0.05. The overall prevalence rate of protozoan parasitic infections was 10.5% (78/740). Four species of protozoan parasites were detected as single infections: Blastocystis hominis (6.8%; 50/740), Entamoeba histolytica/Entamoeba dispar (2.3%; 17/740), Giardia lamblia (0.9%; 7/740), and Entamoeba coli (0.3%; 2/740). Two samples were obtained with mixed infections: the first by Blastocystis hominis with Entamoeba histolytica/Entamoeba dispar (0.1%; 1/740), and the second by Blastocystis hominis with Giardia lamblia (0.1%; 1/740). Prevalence of protozoan infections was insignificantly (p=0.740) higher in males, 10.9% (43/396) than females (10.2%; 35/344). The prevalence of protozoan infections was highest in the 10-19 years age group (14.1%, 14/99), and lowest in the 30-39 years age group (5.7%; 4/70), with no significant differences between the intestinal protozoan infection and age groups (p=0.641). According to the seasonal variations, the results revealed that the highest rate of infection was in November (20.8%, 11/53), followed by 19.6% in September (9/46), while the lowest infection was 4.3% in July (3/70). No significant differences were detected in the prevalence of infection between the different months (p=0.236).

Key words:-*Sebha; prevalence, intestinal protozoa.*

INTRODUCTION:

Intestinal parasitic infection is a major public health problem worldwide, particularly in the tropical and subtropical countries. World health organisation (WHO) estimated 3.5 billion individuals are affected in the world, with a morbidity of 450 million [1]. Numerous investigations in many parts of the world have indicated a direct correlation between the high prevalence of parasitic infections and lower socioeconomic status of the studied area. Furthermore, factors like poor hygienic environment, overcrowding of people, humid and hot tropical climate, lack of safe drinking water, and immigration of workers from the neighbouring countries are responsible for increasing the prevalence of these infectious agents [2-6]. Intestinal protozoa parasites are transmitted by the fecal-oral route and they cause intestinal signs such as abdominal pain, diarrhea, weight loss, anorexia, nausea, vomiting and anemia [7]. Many studies throughout the country of Libya have been performed and demonstrated a various rates of infection, although the infection with three intestinal protozoan infection (*Blastocystis hominis*, *Entamoeba histolytica* and *Giardia lamblia*) are still more prevalent than other intestinal parasites (*Balantidium coli*, *Entamoeba hartmani*, *Entamoeba dispar*, *Entamoeba coli*, *Endolimax nana*, *Iodamoeba bütschlii*, *Chilomastix mesnili*, *Trichomonas hominis* and *Cryptosporidium parvum*). Prevalence of intestinal parasites in different parts of Libya is varied from 10.6% in Zawia [8]; 12.9% [9] and 27.9% [10] in Benghazi; 14.8% in Sebha [11]; 15.4% in Al-Khoms [12]; 29.6% in Nalout [13]; to 31.0% in Derna [14]. The objective of of the current study is to describe the prevalence of intestinal protozoa infection in individuals referred to Sebha Central Laboratory in Sebha city, Libya.

MATERIALS AND METHODS

Study area:

The study was conducted to determine the prevalence of intestinal protozoan parasitic infections in Sebha city, Libya. Sebha is located in the South-east of Libya between 26, 28 North, and 14,16 East. The climate in this city is generally hot and dry. Summertime temperatures routinely exceed 45°C, and in the winter, it can drop at nights to freezing point (0°C).

Study populations:

This study was carried out on all individuals who were referred to Sebha Central Laboratory for stool exam request. A total of 740 fresh stool samples (396 males and 344 females) were examined during the period from January to December, 2017. The ages of these cases ranged from one month to 92 years, and divided into 7 age groups.

Stool examination:

Fresh Stool specimens were collected in clean labelled plastic containers. Collection date and personal information (name, sex and age) were recorded for each sample. The macroscopic examination of each sample (coulor, consistency, and occurrence of blood and mucus) was also checked. Fecal specimens were examined by direct wet smears using both normal saline and Lugol's iodine solutions [15]. Stool samples were not examined for *Cryptosporidium* spp. The diagnosis of protozoan parasites was based on the morphological features of cysts and/or trophozoites found in the stools.

Statistical analysis:

The results were analyzed using SPSS version 20.0 The Chi-square test was used to measure the statistical significant differences at level $p<0.05$.

RESULTS

A total of 740 stool samples were collected from individuals referred to Sebha Central Laboratory from January to December, 2017. Of these, 396 (53.5%) were males and 344 (46.5%) were females. The ages of these people ranged from 0 to 92 years, and the majority of them aged between 0-9 years (34.9%; 258/740). The highest number of the samples was collected in months of May (17.0%; 126/740) and April (11.8%; 87/740). The overall prevalence rate of protozoan parasitic infections was 10.5% (78/740). Prevalence of protozoan infections was insignificantly (p=0.740) higher in males, 10.9% (43/396) than females (10.2%; 35/344) as indicated in Table1.

Table 1: Prevalence of intestinal protozoa according to gender.

Sex		The result		
		positive	negative	Total
male	No. samples	43	353	396
	Prevalence %	10.9%	89.1%	100.0%
female	No. samples	35	309	344
	Prevalence %	10.2%	89.8%	100.0%
Total	No. samples	78	662	740
	Prevalence %	10.5%	89.5%	100.0%

About 97.4% of infected samples contained a single parasite and 2.6% contained double parasites. Four species of protozoan parasites were detected *in this study (Table 2), B. hominis* being the most common parasite was present in 50 samples (6.8%) followed by *E. histolytica/E. dispar* present in 17 samples (2.3%), *G. lamblia* in 7 samples (0.9%) and *E.coli* in 2 samples (0.3%). However, a significant association was observed prevalence of infection between the detected

species of protozoan parasites (p<0.001). Two samples were obtained with mixed infections: the first by *B. hominis* with *E. histolytica/E. dispar* (0.1%; 1/740), and the second by *B. hominis* with *G. lamblia* (0.1%; 1/740). A High significant difference was detected between the single and mixed infections (p=0.000). The distribution of protozoan parasites among infected samples was presented in Figure 1.

Table 2: Frequency of detected species of parasites in infected male and female individuals.

Sex	Infected males		Infected females		Total	
	Single infection					
No. of examined	N= 396		N= 344		N= 740	
Parasite species	Frequency No.	Percentage %	Frequency No.	Percentage %	Frequency No.	Percentage %
B. hominis	27	6.8%	23	6.7%	50	6.8%
E. histolytica/E.dispar	11	2.8%	6	1.7%	17	2.3%
G. lamblia	3	0.8%	4	1.2%	7	0.9%
E. coli	0	0.0%	2	0.6	2	0.3%
	Mixed infection					
B. hominis & E. histolytica/dispar	1	0.25%	0	0.0%	1	0.1%
B. hominis & G. lamblia	1	0.25%	0	0.0%	1	0.1%
Total	43	10.9%	35	10.2%	78	10.5%

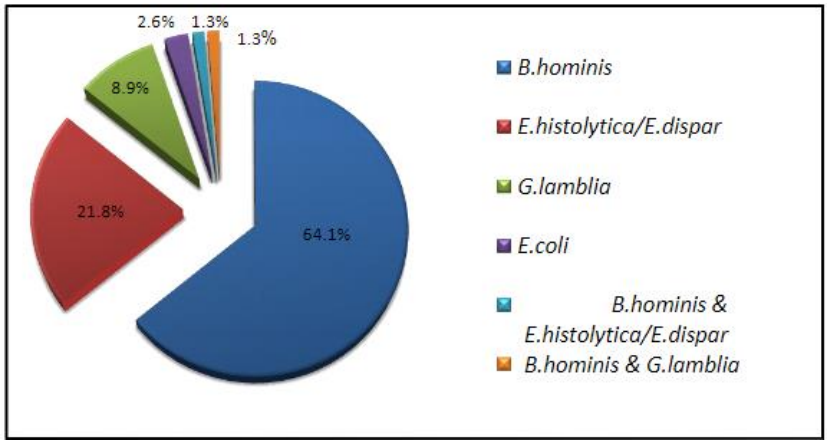

Figure 1: Prevalence of intestinal protozoan infections detected in infected stool samples.

The prevalence of protozoan infections was highest in the 10-19 years age group (14.1%, 14/99), and lowest in the 30-39 years age group (5.7%; 4/70) as indicated in Table 3. There was no statistically significant difference in the percentage of intestinal protozoan infections according to the age group (p=0.641).

Table 3: Prevalence of protozoan infections stratified by age.

Sex	Age group	0-9	10-19	20-29	30-39	40-49	50-59	≥ 60	Total
Male	No.of samples	151	53	31	39	46	37	39	396
	Frequency	16	5	6	3	3	5	5	43
	Percentage %	10.6%	9.4%	19.4%	7.7%	6.5%	13.5%	12.8%	10.9%
Female	No. of samples	107	46	54	31	38	32	36	344
	Frequency	11	9	3	1	4	3	4	35
	Percentage %	10.3%	19.6%	5.6%	3.2%	10.5%	9.4%	11.1%	10.2%
Total	No. of samples	258	99	85	70	84	69	75	740
	Frequency	27	14	9	4	7	8	9	78
	Percentage %	10.5%	14.1%	10.6%	5.7%	8.3%	11.6%	12.0%	10.5%

According to the seasonal variations, the results revealed that the highest rate of infection was detected in November (20.8%, 11/53), followed by 19.6% in September (9/46), as shown in Figure 2. While the lowest infection was 4.3% in July (3/70). The results indicated in table 4 showed that there was no significant differences in the prevalence of infection between the different months (p=0.236).

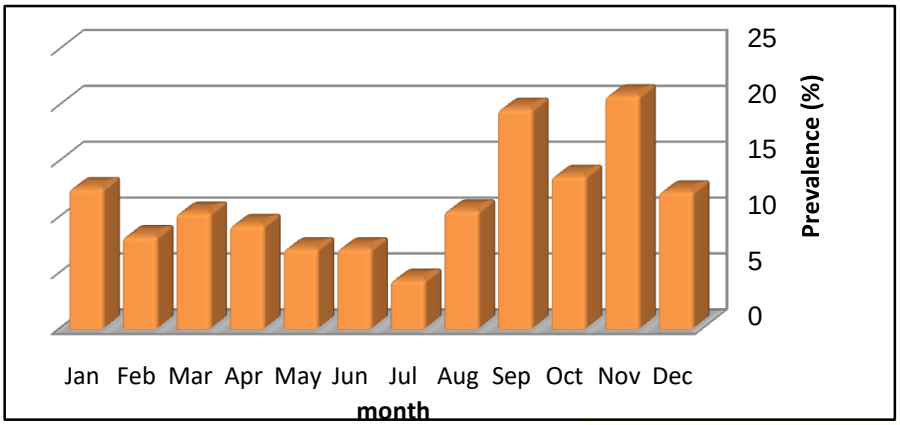

Figure 2: Prevalence of intestinal protozoan parasites in relation to months of the year.

Table 4: The distribution of intestinal parasites in infected people stratified by month.

Month	sex						Total		
	Male			Female					
	No. of samples	infected	Percentage (%)	No. of samples	infected	Percentage (%)	No. of samples	infected	Percentage (%)
January	10	3	30%	14	0	0.0%	24	3	12.5%
February	34	3	8.8%	39	3	7.7%	73	6	8.2%
March	29	3	10.3%	29	3	10.3%	58	6	10.3%
April	51	5	9.8%	36	3	8.3%	87	8	9.2%
May	72	5	6.9%	54	4	7.4%	126	9	7.1%
June	16	2	12.5%	12	0	0.0%	28	2	7.1%
July	43	2	4.8%	27	1	3.7%	70	3	4.3%
August	28	2	7.1%	29	4	13.8%	57	6	10.5%
September	26	5	19.2%	20	4	20.0%	46	9	19.6%
October	24	3	12.5%	20	3	15.0%	44	6	13.6%
November	24	4	16.7%	29	7	24.0%	53	11	20.8%
December	39	6	15.4%	35	3	8.6%	74	9	12.2%
Total	**396**	**43**	**10.9%**	**344**	**35**	**10.2%**	**740**	**78**	**10.5%**

DISCUSSION

This study resulted that 78 individuals out of 740 were infected with one or two species of protozoan parasites namely: *B. hominis*, E. *histolytica/E. dispar*, G. *lamblia*, and *E. coli*. *E. histolytica* and *G. lamblia* are pathogenic parasites associated with diarrhea and responsible for casing Amebiasis and Giardiasis, respectively [16]. Both can be transmitted orally by drinking or eating the infective stage with contaminated water or food. *E. histolytica* is considered to be the third most leading cause of death in the world after Malaria and Schistosomiasis [17]. *B. hominis* is still not clear whether it is a pathogenic parasite or commensal [18, 19]. Whereas, *E. dispar* and *E. coli* are non-pathogenic protozoan parasites.

Studies on the prevalence of intestinal parasites in the south of Libya are still very rare. The total number of stool samples collected from people who attending Sebha Central Laboratory in the current study was 740 and, it is very low in comparison with 1,526 in the similar study was conducted in 2014 [11]. From the previous studies, the epidemiology of intestinal protozoan infections indicates that these parasites are found in both sexes and all age groups. However, the infection incidence is high in some places in the world, in children and in patients with Acquired Immunodeficiency Syndrome (AIDS) [20-23]. The present study indicated that the overall prevalence of intestinal protozoan infection was 10.5% which is similar to previous studies in Zawia (10.6%) [8], Al-Baida (12.0%) [24], Houn (12.7%) [25] And in Benghazi (12.9%) [9]. this finding is lower in comparison to studies carried in Libya, Sebha (14.8%, 27.6%) [11, 26], Al-Khoms (15.4%) [12], Derna (31.0%) [14], and Nalout (29.6%) [13]; and other countries, 16.6% in Ethiopia [27]; 18.0% in Kashmir [28]; 27.8% in Saudi Arabia [29]; 52.0% in Pakistan [30]; and 53.0% in Yemen [31].

The finding of this study indicated that the rate of intestinal protozoa in males (10.9%) was slightly higher than in females (10.2%), with no statistically significant difference between them (p=0.740). This result is similar to that obtained by Elsaid et al.,[8], Zaed [25] and Mathuria1 and Singh [32]. In contrast, the higher rates of infections in females than males were reported by Ibrahim [11] and Okyay et al., [33]. This can be attributed to the fact that males like to eat outside fast foods more than females.

The age of study population varied from 1 month to 92 years. Maximum prevalence of infection was found in the age group of 10–19 years (14.1%), which is also reported in previous studies [32, 34-36]. The high prevalence in this group of age may be referred to the activity and life style of these individuals and the poor hygiene among these individuals at these ages. No statistical differences were found in the prevalence of infection between the seven age groups (p=0.641), which is similar to studies done by Dar et al, [10], Sadaga and Kassem [14], and Elsaid et al., [8].

The highest prevalence of parasitic infection was detected in November (20.8%), followed by 19.6% in September, while the lowest prevalence was recorded in July (4.3%), with no significant differences in the prevalence of infection between the different months (p=0.236). On average the autumn months (September-October) showed the highest incidence rate of infection (16.6%), followed by winter months (November–February; 13.4%), in comparison with spring (March-April; 9.8%) and Summer (May-August; 7.3%) months. Dry climate and the high temperature (>45◦C) may be responsible of decreasing the incidence rate in summer months and affecting on the viability protozoan parasites cysts. Furthermore, the differences in the rate of infection during the year months in the previous studies may be attributed to the weather and the socioeconomic status for the subjected persons in the study.

The data of the present study showed that *B. hominis* was the most common parasite discovered in this study with infection rate of 6.8% (50/740). This parasite represented 64.1% from the infected people (50/78). This finding is nearly similar to those previously reported in Derna [14] and Sebha [11] with infection rates of 6.7% and 9.8%, respectively.

The level of *B. hominis* infection was also reported in other studies in Libya with rates ranged from 18.6% to 29.6% [26, 37-39].

Entamoeba histolytica/ Entamoeba dispar and *G. lamblia* are the second and third most frequently identified intestinal parasites detected in this study, with infection rates of 2.3% (17/740) and 0.9% (7/740) respectively. *E. histolytica* and *G. lamblia* are pathogenic protozoan parasites causing Amoebiasis and Giardiasis, respectively. The lower rate of *E. histolytica/E dispar* infection is almost similar to those reported in Sebha 3.1% [11] and 4.0% [40]; Houn 4.8% [25]; Wadi Al-Shati 1.2% [41]; Zawia 3.1% [8]; 4.4% in Benghazi [42] and Tripoli 3.0% [43]. However, higher rates of *E. histolytica/ E. dispar* were also reported: 6.6% in Derna [14]; 11.8% in Zliten [44]; 12.1% in Alkhoms [12]; 16.3% in Sirt [45]; and 21% in Nalout [13].

The current study shows that 0.9% of subjected people were infected by *G.lamblia*. This finding was consistent with reports in Zliten (1.2%) [44]; 1.3% in Tripoli [46] and Brack [47]; 1.5% [11], 1.6% [26] in Sebha; 1.8% in Zawia [8] and Wadi Al-Shati [41]. The higher rates were reported and varied from 3.2% in Sebha [48]; 11.4% in Benghazi [10]; 12.7% in Derna [14]; 17.0% in Tripoli [43]; to 28.8% in Sirt [45].

The fourth parasite detected in this study was *Entamoeba coli*. This non-pathogenic protozoan was only found in two females with a prevalence rate of 0.3%. Similar results have been recorded in difrent places in Libya, 0.3% [11] and 0.4% [26] in Sebha, 0.4% in Nalout [13], 0.9% in Wadi Al-Shati region [41]. The rate of *E. coli* infection is low in comparison with other rates were reported in Benghazi 2.6% [9]; Derna 3.2% [14]; Zawia 3.6% [8]; and Tripoli 4.0% [49].

Two samples were obtained with mixed infections (*B.hominis* with *E.histolytica/E.dispar*; and *B.hominis* with *G.lamblia*) with a prevalence rate of 0.3%.
In conclusion, intestinal parasitic infections are still a major public health problem in African countries. In the present study, low prevalence of intestinal protozoan infections was recorded in comparison with the previous studies in Libya. We suggest that the source of drinking water and environmental sanitation are the main factors affecting the prevalence of these infections in Sebha city. Therefore, more future investigations including risk factors are required.

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