



Prevalence and Seasonal Distribution of Protozoal Infections— Entamoeba Histolytica and Giardia Lamblia—in Dir Lower

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ABSTRACT

Background: The research examines the prevalence and seasonal distribution of protozoal infections—*Entamoeba histolytica* and *Giardia lamblia*—in Dir Lower, 2021, highlighting distinct patterns.

Results: *E. histolytica* consistently recorded higher infection rates year-round, with a significant peak in August, likely tied to warmer temperatures that enhance both water contamination and protozoal survival. *G. lamblia* cases, although fewer, exhibited modest increases in late winter and summer, potentially due to post-rainy conditions

favorable to its transmission. Age and gender analysis revealed a higher prevalence of *G. lamblia* among male children in the 11–20 age group, possibly due to increased outdoor exposure and water contact, while females showed slightly lower infection rates.

Conclusion: The study underscores a need for age-specific interventions, particularly for boys under 15, to curb infections effectively. Emphasizing targeted strategies during summer and vacation periods, the research provides critical insights for designing more focused public health measures to control protozoal transmission in the Dir Lower community

Keywords: Protozoal Infections, Seasonal Distribution, Prevalence, Age and Gender Analysis, Targeted Interventions, Public Health Measures.

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INTRODUCTION

Zoology focuses on parasitology – intimate host-interaction systems where parasitic, mutualistic, or commensal associations emerge from coexistence and complex ecological entwinement. Among these, parasitism is where one organism the parasite derives some benefit from the other organism the host most often to the detriment of that organism (Solomon et al., 2014). Those parasites that exist in living organisms include ectoparasites, which exist on the outer surface of the host organism, and endoparasites which exist inside the host organism. Some common forms that infect humans and animals include They are common more so in tropical and subtropical areas where hygienic access is hard to come across; they are prevalent in low-income regions (Bush et al., 2001). Intestinal pathogenic parasites (IPP) are endo-organisms that live and reproduce in the small human digestive tract and of which some of the identifiable common variants are known to cause gastroenteritis. This type of infection has high health risks for children; particularly those in developing nations where several environmental and social and economic factors increase their risk exposure (Okyay et al., 2004). It is estimated that 3.5 billion individuals, specifically, are affected worldwide with morbidity associated with such factors as geographical environment, social and economic status, and hygiene and sanitation (Hotez et al., 2004; Pawar et al., 2016). The disease is communicated through food products, water, and contact with soil and animals that are affected by the disease. The ailment that is caused by intestinal parasites varies from fatal and deadly symptoms such as dysentery, and physical growth checks in children who are the most affected vulnerable groups (Rodriguez-Morales et al, 2006). When the condition rises to an acute attack, it goes by the name acute gastrointestinal illness (AGI), and this leads to the death of about 2.2 million people out of diarrheal diseases globally, most especially in the developing world (WHO, 2002). In Pakistan, the Percentage distribution of under-five mortality from diarrhea is ranged between 20% to 30% (Habib et al., 2013). In developed countries, mortality of AGI is insignificant, but morbidity of AGI often leads to complications that result in huge health costs (Fleury et al., 2009). Water born, soil born and food born are the most common sources of infection with IPPs (Mirdha and Samantray, 2002). Contamination of water highlighting IPP is

remarkable, through the consumption of water supply and use in agriculture and food preparation, interpersonal contact, and domestic practices. Contamination by parasites also comes about because of the consumption of fruits and vegetables without washing them (Munoz-Antoli et al., 2014). Additionally, the mode of transmission through food is often observable and salad vegetables and fruits if washed can act as agents of transmission. Thus, although being more extended in overweight individuals from low-income countries, intestinal parasites are certainly associated with significant morbidity in developed nations as well (Liu et al., 2012). Protozoa are irregularly shaped, Motile or non-motile organisms that may be free-living or parasites. They affect numerous categories of vertebrates and invertebrates depending on the host environment. Transmission may occur via the movement of fecal particles via food or water that has been ingested. *Entamoeba histolytica* and *Giardia lamblia* are two of the principal protozoan parasites. The parasite causing amebiasis is *Entamoeba histolytica* infects about fifty million people internationally, and has high mortality (Ham, 2020). Its life cycle stringently comprises pathogenic trophozoites as well as cysts by consuming water or foods contaminated with the cysts. *Giardia lamblia* is a zoonotic coccidian parasite and is one of the most common causes of diarrheal illness. A cycle includes its infectious forms referred to as cysts and motile forms called trophozoites. Both stand out as major diagnostic and public health problems, particularly in the Third World where hygiene infrastructure is lacking.

METHODS

The study was carried out around the District Headquarter Hospital Timergara in Dir Lower, Khyber Pakhtunkhwa, Pakistan. This area is dominated by mountains and villages; the majority population is settled in the Panjkora Valley. The climatic conditions add to the hardness of terrains combined with a stringer identification of rural settings that give the region different patterns of disease incidence and prevalence which also affect patterns of protozoan infections. Data collection for the study took place in several key laboratories: Such labs include Dr. Fazal Rahim's Lab in the Pathology Department of Timergara Teaching Hospital, Qazi Lab No. 1 and 2, and Malak Inam Lab, and six other private laboratories in the

area. These disparate data sources gave a quantitative synoptic view of protozoal infection in Dir Lower that is commensurate with the demographic and environmental influences that affect public health.

This was a cross-sectional study done about residents of Timergara and surrounding areas in January 2021. Respondents in the study were selected based on age, gender, and region within the Dir Lower district only. The target patients included those with diarrheal illness or other manifestations of gastrointestinal infection, and samples were mainly collected from public and private healthcare facilities in Timergara.

Altogether 1822 fecal samples were taken from the cases who had clinical symptoms. To avoid contamination each sample was collected in a pre-labeled sterile disposable plastic bottle. Equal numbers of samples were taken from each age group and sex and in winter, summer, autumn, and spring to assess the annual rhythm, seasonal fluctuations, and other patterns in infection rates in the general population. All samples were fixed in 5% formalin and were shipped immediately to DHQ Hospital laboratories in Timergara.

All the fecal samples were assessed according to the standard operating procedure used in this laboratory. These items were Pipes, gloves, normal saline solution, glass slides, cover slips, Microscopes, and other accessories. For every sample collected, floatation and sedimentation methods were employed to identify protozoan cysts, ova, and eggs. Floatation Technique Tiny amount of feces (2mg) was teased with a drop of saline on a clean glass slide and a cover slip was placed on it and left to settle for 15 minutes. The smear was then considered the scope under the microscope. Saline was used as the vehicle for parasite searching and microscopy mainly centered on the detection of cysts, ova, or eggs of any intestinal parasite according to their morphological appearance.

For sedimentation, three milliliters of the sample were diluted with 40 ml of water before straining. The filtrate was collected in a test tube and allowed to deposit for about 5 minutes before it was reluctantly redispersed in 5 ml of distilled water. A drop of saliva was placed on the slide, a cover slip was put on and it was

examined. For parasite determination, the direct slide smears with saline wet mount were followed by Lugol staining to increase the contrast and enable species to differentiate the morphology.

The data collected were analyzed using Statistical Package for the Social Science (SPSS) software. To test hypotheses, norms for percentile values such as yearly incidence rate of infection, seasonal fluctuation, and distribution of protozoan infection in terms of demographic characteristics were developed. The findings presented in the work gave an understanding of the specifics of protozoal infections, such as, their seasonal and demographic dependence in Dir Lower.

RESULTS

A monthly distribution of protozoal infection, *Entamoeba histolytica*, and *Giardia lamblia* for the year 2021 has also shown the flow of patients for both infections in different months of the year in the year 2021, the flow of cases of *Entamoeba histolytica* is consistently higher than the cases of *Giardia lamblia* in all the months of the year. Epidemiological data of *Entamoeba histolytica* have a higher prevalence during summer, especially in August, which implies that hot climatic conditions may promote an increase in water activities that lead to the spread of the parasite. On the other hand, *Giardia lamblia* cases are comparatively low but slightly rise in late winter and summer maybe due to certain factors influencing the rate of transmission of *giardia lamblia*. These seasonal variations show that essentially both the protozoa have peak during summer, and the number drops in cooler months hinting at low parasite survivability and occurrence in such a climate. While comparing *Entamoeba histolytica* and *Giardia lamblia* ‘month by month,’ *Entamoeba histolytica* continues to be the most rampant protozoan infection throughout most of the period. The cross-comparison chart extends the focus on infected rates where *Entamoeba histolytica* remains the higher figure again, which underscores the need for more specific attention for this specific parasite. This pattern suggests that the protozoan infection rate probably rises because of conducive conditions brought about by increased water activities and warm temperatures that could enhance the transmission of *E. histolytica* to a larger

extent than GL. The Monthly case load prevalent total protozoan infections identify significant fevers in February, August and November. These increases in the rate of infected individuals may be linked to regional differences or societal practices like festive occasions, contact with infected relevant objects, or relating to another environmental attribute. This has raised the probability of transmission of *Giardia lamblia* in certain months, and this has in one way, may be useful in carrying out specific public health campaigns or extended surveillance in those months. This study of *Giardia lamblia* showed more prevalence in males, particularly in the age range of 11-20 and 21-30 years. These demographic characteristics might have been influenced by young males coming into close contact with the disease risk factors such as staying outdoors or engaging in water contact activities. Same as for the entire population the curve is similar for females, though slightly lower infection rates might be explained by the different levels of exposure or possible differences in immunity. The identified pattern becomes even more pronounced when the results are categorized by five-year age intervals: males of 11-15 and 16-20 age groups reveal increased sensitivity to *Giardia lamblia*. It clearly shows that there is an age and gender variation that affects vulnerable young groups especially males who may have higher exposure levels. Altogether, the outline presents a general dynamic of *Entamoeba histolytica* and *Giardia lamblia* in 2021 considering seasonal, demographic, and monthly features. The trends for the two protozoa are different; *Entamoeba histolytica* is significantly higher than the other protozoa; although *Giardia Lamblia* shows an increase at different times, but not as much as *Histolytica*, especially during warmer months. These research scenarios underscore important prevention and control opportunities including intensifying interventions in the high-risk months and pediatric and male populations to prevent and contain protozoal transmission in Dir Lower.

Table 1: Annual prevalence of *Entamoeba histolytica* *Giardia Lamblia*

Month	Total Cases	<i>Entamoeba histolytica</i>	<i>Giardia lamblia</i>
January	13	8	5
February	17	14	3
March	11	10	1
April	10	9	1
May	7	6	1
June	8	6	2
July	7	5	2
August	15	13	2
September	12	10	2
October	13	10	3
November	16	12	4
December	6	4	2

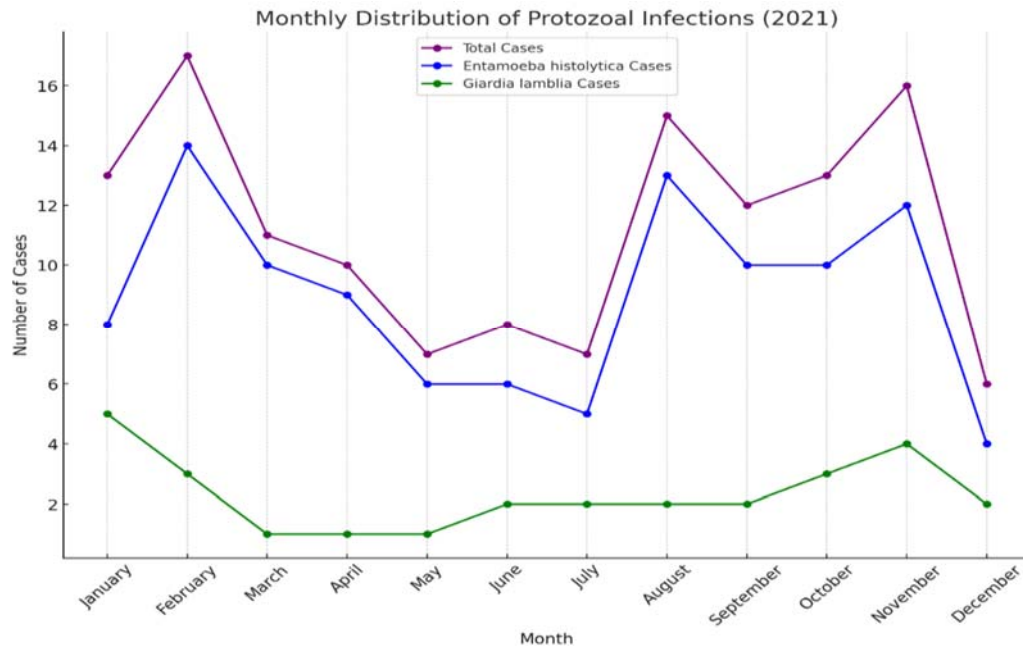
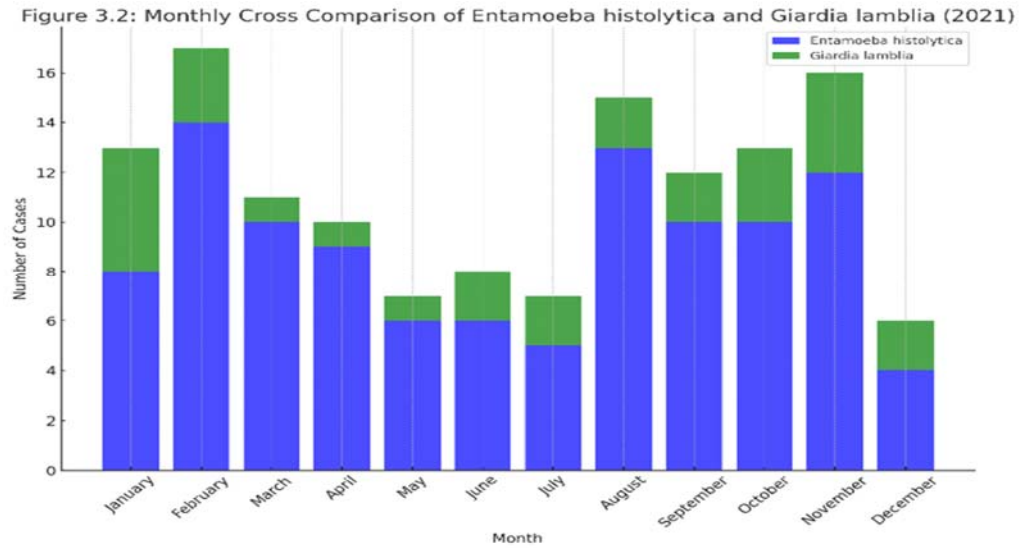


Figure 3.1: Annual Prevalence of Entamoeba histolytica and Giardia lamblia in 2021.

This figure displays the monthly case report for Entamoeba histolytica and Giardia lamblia for the whole calendar year of 2021. The mean frequency of Entamoeba histolytica always experiences a higher spike each month and has the highest rate in the summertime with August in particular. It also rises during a season when protozoal growth is likely to be boosted by environmental conditions like higher temperatures and contact with water. Giardia lamblia cases continue to be relatively low and show seasonal fluctuation and hint of rising during late winter and summer suggesting a feature of contagiousness in warmer and post-rainfall seasons.



The monthly cross-sectional of 2021, presenting *Entamoeba histolytica* and *Giardia lamblia* cases in Figure 3.2. The stacked bar chart enables the direct comparison of the monthly distribution of each protozoon with at least *Entamoeba histolytica* being significantly higher in each of the months depicted. This figure successfully explains the possibilities of getting infected in the two species.

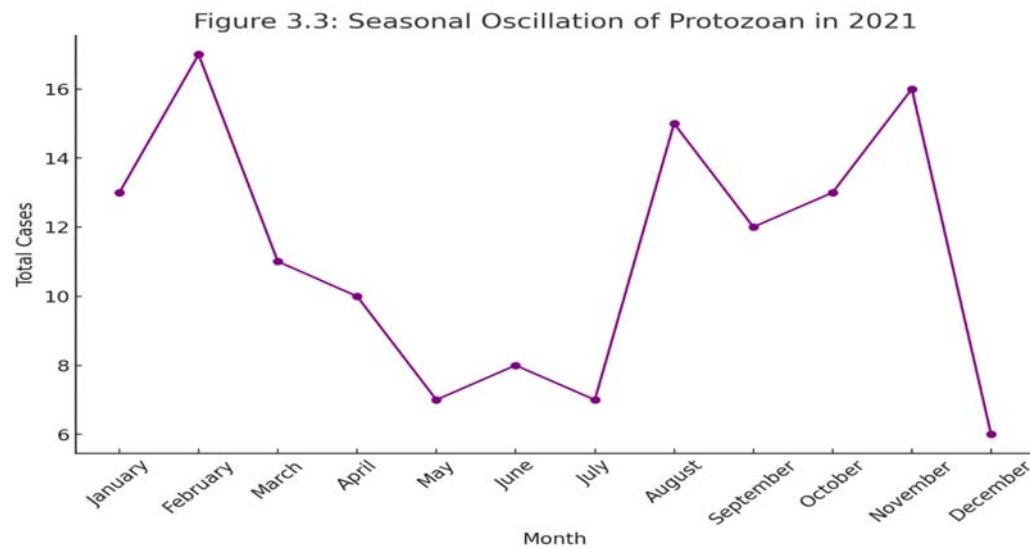


Figure 3.3: Seasonal Oscillation of Protozoan Cases in 2021

These results also depict that, rather than using the absolute number, raw or total number of monthly protozoan infections are preferred to establish seasonality. Here again way more cases were noted particularly in summer, especially in August, which further s Thus, illustrates the seasonality of *Entamoeba histolytica* and *Giardia lamblia*. Others include increased water exposure and often poor sanitation during the hot months of the year to this increase. It must be noted that the result of these pathogens is very low during winter since the pathogens do not survive and cannot spread easily during this season.

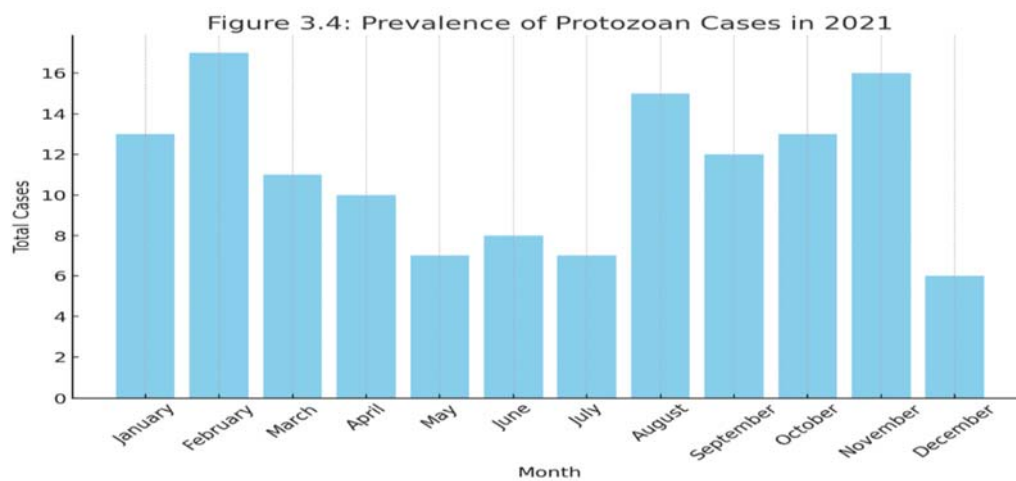


Figure 3.4: Therefore, we compared the 2021 protozoan case frequency with the total percentage.

That is why the monthly point prevalence of total protozoal cases is depicted in the following bar chart, which gives a better vision of the monthly infection load. The chart shows fluctuations with high possible associations with local environmental change and population interactions and contaminated sources in February, August, and November.

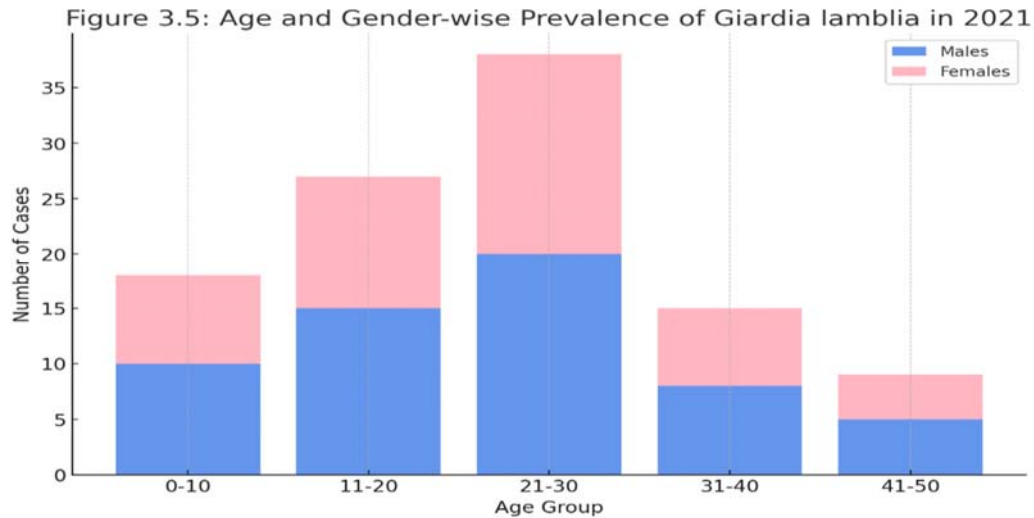


Figure 3.5: Age and Gender-wise Prevalence of Giardia lamblia in 2021.

This figure was hypothesized to illustrate the prevalence rate across age and gender, which is why such distribution is assigned. As a result, the 11-20 and 21-30 age male groups are identified as having the highest prevalence of Giardia lamblia; this could be in connection with outdoor activity and water contamination. Likewise, female dominance is noticed in similar age subgroups, although the general number of cases does not almost reach the figures in males probably because of various rates of exposure to the virus and immune response.

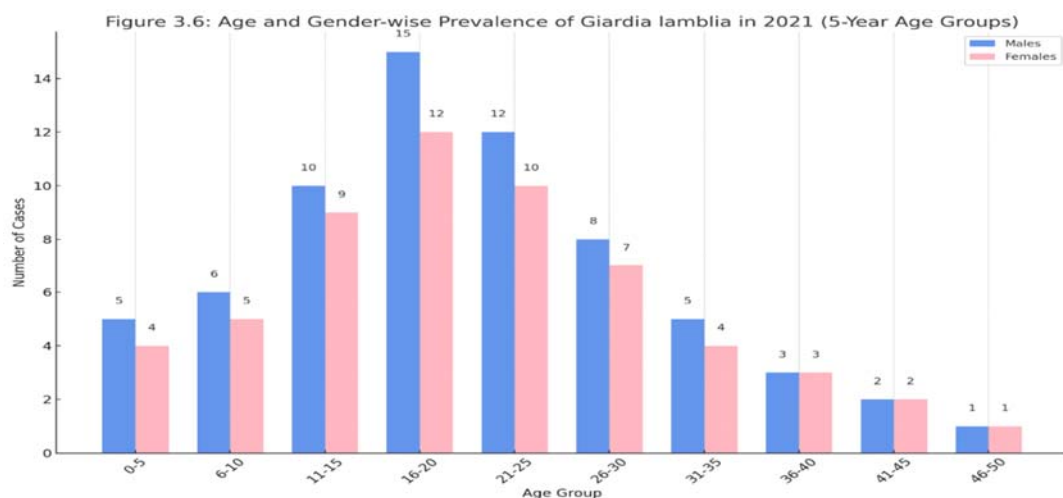


Figure 3.6: Age and Gender-wise Prevalence of Giardia lamblia in 2021

**Mapping Vulnerability Age, Gender, and Geographic Analysis of Protozoal Infections
in Dir Lower 2021**

Lab	Age Group	Gender	Assigned Patients	Area
Teaching Hospital Pathology Dept. -Late Dr. Fazal Rahim	0-10	Male	116	Muhajir Camp
	0-10	Female	69	Main City Timergara
	20-Nov	Male	46	Christian Colony
	20-Nov	Female	46	Bagha Dushil
	21-30	Male	69	Baroon
	21-30	Female	46	Arang
	31-40	Male	46	Muajir Camp
	31-40	Female	23	Main City Timergara
Qazi Lab No. 1	0-10	Male	72	Muajir Camp
	0-10	Female	40	Main City Timergara
	20-Nov	Male	27	Christian

				Colony
	20-Nov	Female	27	Bagha Dushil
	21-30	Male	40	Baroon
	21-30	Female	27	Arang
	31-40	Male	27	Muajir Camp
	31-40	Female	13	Main City Timergara
	0-10	Male	63	Muajir Camp
	0-10	Female	35	Main City Timergara
	20-Nov	Male	23	Christian Colony
	20-Nov	Female	23	Bagha Dushil
	21-30	Male	35	Baroon
	21-30	Female	23	Arang
	31-40	Male	23	Muajir Camp
Malak Inam Lab	31-40	Female	11	Main City Timergara

	0-10	Male	60	Muajir Camp
	0-10	Female	32	Main City Timergara
	20-Nov	Male	21	Christian Colony
	20-Nov	Female	21	Bagha Dushil
	21-30	Male	32	Baroon
	21-30	Female	21	Arang
	31-40	Male	21	Muajir Camp
	31-40	Female	10	Main City Timergara
Private Lab 1	0-10	Male	43	Muhajir Camp
	0-10	Female	24	Main City Timergara
	20-Nov	Male	16	Christian Colony
	20-Nov	Female	16	Bagha Dushil
	21-30	Male	24	Baroon

	21-30	Female	16	Arang
	31-40	Male	16	Muajir Camp
	31-40	Female	8	Main City Timergara
Private Lab 2	0-10	Male	40	Muhajir Camp
	0-10	Female	21	Main City Timergara
	20-Nov	Male	14	Christian Colony
	20-Nov	Female	14	Bagha Dushil
	21-30	Male	21	Baroon
	21-30	Female	14	Arang
	31-40	Male	14	Muajir Camp
	31-40	Female	7	Main City Timergara
Private Lab 3	0-10	Male	32	Muhajir Camp
	0-10	Female	16	Main City Timergara

	20-Nov	Male	10	Christian Colony
	20-Nov	Female	10	Bagha Dushil
	21-30	Male	16	Baroon
	21-30	Female	10	Arang
	31-40	Male	10	Muajir Camp
	31-40	Female	5	Main City Timergara
	0-10	Male	25	Muhajir Camp
	0-10	Female	13	Main City Timergara
	20-Nov	Male	9	Christian Colony
	20-Nov	Female	9	Bagha Dushil
	21-30	Male	13	Baroon
	21-30	Female	9	Arang
	31-40	Male	9	Muajir Camp
Private Lab 4	31-40	Female	4	Main City

				Timergara
Private Lab 5	0-10	Male	21	Muhajir Camp
	0-10	Female	10	Main City Timergara
	20-Nov	Male	7	Christian Colony
	20-Nov	Female	7	Bagha Dushil
	21-30	Male	10	Baroon
	21-30	Female	7	Arang
	31-40	Male	7	Muajir Camp
	31-40	Female	3	Main City Timergara
Private Lab 6	0-10	Male	16	Muhajir Camp
	0-10	Female	8	Main City Timergara
	20-Nov	Male	5	Christian Colony
	20-Nov	Female	5	Bagha Dushil
	21-30	Male	8	Baroon

	21-30	Female	5	Arang
	31-40	Male	5	Muajir Camp
	31-40	Female	2	Main City Timergara

DISCUSSION

The protozoal infections in Dir Lower have their own patterns in the year 2021 with a drastic reduction in number probably due to COVID-19. A total of only 135 clients received the reported treatment, accounting for 41.5 percent of the total funding for the multi-year study period. Perhaps a low prevalence of infections due to COVID-19 strict measures which led to limited chances of hospital visits, light physical contact with other people, and restricted access to medical tests. Perhaps true numbers of infections were accordingly higher, this could be because of underreporting. Distribution per gender in 2021 identified that males were more affected 68.2% for *Entamoeba histolytica*, and 67.9% for *Giardia lamblia*. This trend is like previous similar studies, for instance, Arshad et al. (2019). Seasonality concerns show that the disease is most prevalent during the summer season and very low during the winter season. These seasonal trends suggest that factors such as heat, and increased water activity may also play a role in protozoa transmission. Finally, the overall rates of infection were significantly higher in children and young adults, and in low-income families, perhaps because of poor access to clean water and relatively low standards of hygiene.

To address the challenges observed in 2021, the following recommendations are proposed:

Increase Public Awareness: Organize focused health promotion interventions regarding personal and food hygiene and risks of protozoal infections mainly targeted at facilities in low socioeconomic neighborhoods. Of these, schools and community centers should be the focal areas of educational outreach.

Enhance Water Treatment and Sanitation: Communities should be encouraged to upgrade their water treatment centers and other sanitation facilities, especially in the most affected areas. This step is most important during some of the peak

times of infection in summer. Strengthen Surveillance and Reporting Systems: For protozoal disease surveillance, put in place effective means for gathering and reporting protozoal infection data regularly so as not to be compounded by restricted movement during an outbreak to produce an underestimation of the disease rates. Seasonal Intervention Measures: Promote awareness during high-risk periods with hand washing and water safety and sanitation literature to avoid the high spread of infections

CONCLUSION

From the work done in 2021, it was discovered that protozoal infection prevalence in Dir Lower was convinced through such external factors as the COVID-19 pandemic. Social distancing measures by avoiding health care services and limiting contact during lock-ins probably led to fewer reported cases. As would be expected, *Entamoeba histolytica* was the most prevalent of all species where the males had a higher infection rate than the females. The young, the poor, or individuals with poor backgrounds were most affected, and the cases' seasonal patterns have shown high levels in summer, which may be due to environmental factors.

FUNDING

None

CONFLICT OF INTEREST

None

ETHICAL APPROVAL

This study was approved by MTI- Hayatabad Medical Complex Ethical Review Board (Approval No: 731 A).

AUTHORS' CONTRIBUTIONS

All authors contributed equally as per ICMJE policy

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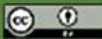
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