Available online at sjuoz.uoz.edu.krd

journals.uoz.edu.krd

Science Journal of University of Zakho

Vol. 10, No. 4, pp. 153 –158, October-December 2022

p-ISSN: 2663-628X
e-ISSN: 2663-6298

PREVALENCE OF CRYPTOSPORIDIOSIS AND ITSASSOCIATED RISK FACTORS AMONG HUMAN POPULATION IN ZAKHO DISTRICT, DUHOK PROVINCE, KURDISTAN REGION, IRAQ

Hamdea Ismail Ahmed Shahoi [a, *], Wijdan Mohammed Salih Mero[a,b], Ahmed Basheer Mohammed[a]

[a] Department of Biology, Faculty of Science, Zakho University, Zakho, Duhok Province, Kurdistan Region, Iraq (hamdeaismailahmed@gmail.com).
[b] Scientific Research Center, Nawroz University, Duhok Province, Kurdistan Region, Iraq.

Received: *06 Jul., 2022* / ***Accepted:*** *18 Sep., 2022* / ***Published:*** *09 Oct., 2022* ***https://doi.org/10.25271/sjuoz.2022.10.4.970***

ABSTRACT:

Cryptosporidiosis caused by species belongs to genus *Cryptosporidium*. Cryptosporidiosis is a zoonotic disease that affects both animals and humans, making it a major public health and veterinary concern. A cross sectional study was performed during the period from August 2021 to March2022, to estimate the prevalence of *Cryptosporidium* spp. in the human population in order to determine the effect of associated risk factors on the rate of infection in Zakho district, Duhok. Seven hundred Fecal specimens were collected from Zakho General Hospital and two primary schools located in suburban area of Zakho city. The collected stool specimens were examined macroscopically and microscopically using a modified Ziehl–Neelsen staining method. *Cryptosporidium* infection was found in 87.57% (613/700) of the examined specimens. Males showed slightly a higher infection rate than females (88.4% vs. 85.5%) with non-significant difference ($P > 0.05$). age group (31-41 years) showed highest rates, with significant difference ($P<0.05$). The highest rates of infection were reported among families with 6-10 members, people drinking unfiltered municipal tap water, suburban residents, those with close contacts with domestic animals, and illiterate people. The control of this disease necessitates the improvement of living conditions, providing potable clean water and disseminating health education programs among the community.

KEYWORDS: Cryptosporidium, prevalence, humans, risk factors, Zakho, Iraq.

1. INTRODUCTION

Cryptosporidium species are obligate intracellular protozoan parasites. These parasites complete their life cycles in a single host (a monoxenous life cycle) (Ghazy *et al*., 2015). Cryptosporidiosis is the fifth most important food-borne parasite and the most important parasites causing watery diarrhea in the world (Aniesona and Bamaiyi, 2014; Ryan *et al*., 2014; Bodager *et al*., 2015). Thick-walled oocysts are produced by this coccidian parasite that is found throughout the environment, which are consumed with contaminated water or food and infect the host's gastrointestinal tract (Smith *et al*., 2007; Ghoshal *et al*., 2018).
Several *Cryptosporidium* species have been described and classified into 44 viable species containing nearly 120 genotypes (Ryan *et al.,* 2021). Cryptosporidium species cause infections in a wide range of animals, including fish, birds, reptiles, mammals, and humans (Ryan *et al*., 2014; Ryan and Hijjawi, 2015). Epidemiologic investigations have observed that the species *C. hominis* and *C. parvum* cause the overwhelming majority of human diseases caused by members of this genus (Ryan *et al*., 2014;Thomson *et al*., 2017; Feng *et al.,* 2018). Such studies assume that *C. hominis* is only transmitted between humans, whereas the major reservoirs for *C. parvum* are domestic livestock, primarily cattle, and the parasite is transmitted to humans either directly or indirectly through contact with infected cattle and drinking contaminated water respectively (Krumkamp *et al.,* 2021). Recent epidemiological studies from African and Asian countries considered *Cryptosporidium* spp. as the second most common parasite in children causing severe diarrhea and significant morbidity (Kotloff *et al.,* 2013; Kalantari *et al.,* 2018; Gerace *et al.,* 2019). In Asia, according to a study carried out by Kahn and his colleagues (2019), low-income nations have a high frequency of *Cryptosporidium* (29.88 %), particularly among young individuals. Regarding to the prevalence of cryptosporidiosis in Kurdistan Region and Iraq, most studies on cryptosporidiosis found variable rates of infection. In Dohuk, a rate of 66.95% was reported among children (Al-Saeed *et al.,* 2020), 40.4 % among diarrheal patients in Wasit (Alkhanaq and Thamer, 2022);14.6% in Erbil (Khoshnaw *et al.,* 2017), 47.3% among children in Baghdad (Merdaw *et al*., 2018), and 58% among diarrheal patients in Al-Najaf (Sayal, 2019).
Due to the lack of information on the prevalence of cryptosporidiosis in Zakho district,Kurdistan Region, Iraq , this study was conducted to investigate the prevalence of *Cryptosporidium* spp. and their associated risk factors among human population using a modified Ziehl-Neelsen staining technique.

2. MATERIALS AND METHODS

2.1. Study area

Zakho district, located 50 kilometers northwest of Duhok and a few kilometers from the Iraq-Turkey border, and it is the second largest city in Duhok Province. Annual rainfall exceeds 702 mm,

* Corresponding author

with very hot and dry summer temperatures exceeding 35°C and cool winters. Despite the fact that this region has few grazing areas for small ruminants, intensive farming is popular. Two of the region's most important water sources are underground aquifers and the urban water supply network.

2.2. Sample and data collection

A descriptive cross-sectional study was conducted on 700 fecal samples taken from outpatients in Zakho General Hospital and from the students of two primary schools located in the suburban area in Zakho District, between August 2021 to March 2022. Each fresh sample was collected in a clean, clearly labelled screw-topped samples tubs and kept frozen on a cold pack fully labeled with patients' information after taking a verbal consent from each participant, and from parents of infants and small ages. Furthermore, an approval was taken from the ethical committee of Zakho General Directorate of Health (680/3, 2021/8/3), for the use of data and samples in this study. Gender, age, residency, family size, level of education, and other information were collected via a questionnaire designed for the study. Daily, the collected specimens were transported to the Postgraduate Microbiology Laboratory, Biology Department, Zakho University for macroscopic and microscopic examination.

2.3. Sample Examination

2.3.1. Macroscopic Examination: Each fresh sample was examined with the naked eye for consistency, odor, and the presence of blood, mucus, and pus.

2.3.2. Microscopic Examination: Microscopic examination of stool specimens was done using the modified Ziehl Neelsen technique. On microscope slides, thin and thick fecal smears were prepared using a wooden applicator stick and dried. For 30 seconds, the slides were fixed in absolute methanol before being stained for approximately 1 minute using Kinyoun's carbol fuchsin. After that, the slide was carefully cleaned with tap water. Decolorization is reduced by agitating for 2 minutes in a trough of 1 % hydrochloric acid alcohol, rinsed with tap water. Counterstained for 2 minutes in 1% methylene blue, rinsed thoroughly, and air dried (John and Petri 2006). Each slide was examined at 40X and 100X (oil immersion magnification) to determine the presence and absence of oocysts. It was declared a positive result when an oocyst with the correct morphology (i.e., optical properties, internal structure, shape and size) was found in the sample. At least three slides were prepared and examined from each specimen's various parts.

2.4. Data management and analysis

All collected data were entered into an Excel document before being transferred to the Statistical Package for Social Sciences (SPSS) version 25 for data cleaning, which included running frequencies for each variable. A Chi-square *(X^2)* test was used to determine whether or not there is a statistically significant difference in the prevalence of *Cryptosporidium* with the independent variables, and the relationship between *Cryptosporidium* infection and risk factors was considered significant at $P< 0.05$.

3. RESULT

Using modified Ziehl Neelsen stain, the morphological characteristics of the oocysts were revealed which helped in micrometry of oocysts (Fig.1). Oocysts are slightly oval measured 6.9±0.72 µm in length and 5.8 ± 0.87 µm in breadth. The parasite was shown as thick pigmented pink to red bodies on a blue or purple background, with a distinct halo surrounding the oocyst (Fig.2).

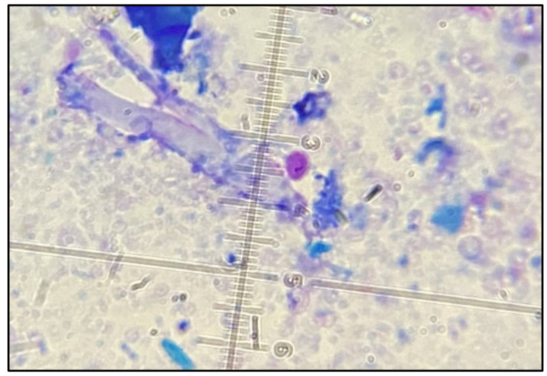

Figure (1): *Cryptosporidium* micrometry of oocyst 100 × (6.1× 5.6 µm in size) stained with Ziehl Neelsen stain

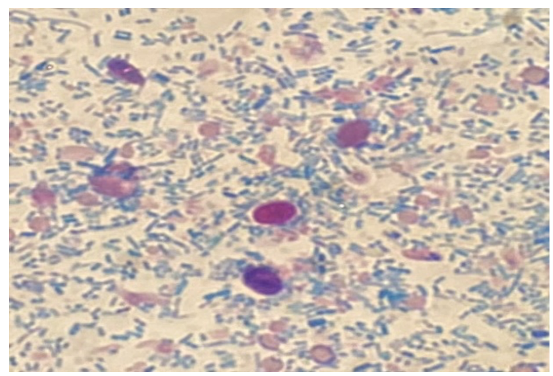

Figure (2): *Cryptosporidium* oocysts stained with modified Ziehl Neelsen staining × 100

The prevalence of *Cryptosporidium* oocysts among population and their association with some risk factors are summarized in Table 1. The overall rate of *Cryptosporidium* infection in the population was 87.57 % (613/700) in this study using the modified Ziehl Neelsen technique. The infection rate was slightly higher among males (88.4%) compared to females (85.5%) with non-significant difference (P >0.05) between both genders. As regard to age, the highest rates of the infections were reported among the ages 21-30, 31-40 and above 41 years which were 90.5%, 91.7% and 91.0%, respectively. Statistically the differences were significant between different ages (P < 0.05). Regarding to the nature of the stool, the formed stool specimens contained the highest rate of infection (89.2%), while the lowest rate (84.6%) was reported among liquid stool specimens, with non-significant difference (P>0.05)between both types of specimens.

Concerning family size, large families comprising of 6-10 members showed the highest rate (94.1%), while the lowest rate (72.2%) was found among families with 1-5 members. Statistically the differences between rate of infection and family size were significant (P<0.05). Regarding water sources, higher prevalence (94.8%) was seen in individuals who drink unfiltered municipal tap water, while people drinking commercial bottled water and those filtering municipal tap water showed a lower rate

(65.9%), with statistically significant differences ($P< 0.05$) between them. Suburban residents had a higher rate of infection (87.6%) as compared to urban one (75%) and significant difference was reported ($P< 0.05$) between both groups.

Regarding to occupation, students showed the highest rate of infection (90%), while the rates among other groups were lower as indicated in Table (1), but statistically the differences between all groups were non-significant ($P>0.05$). People in contact with animals showed the highest rate of infection (90.9%) in comparison with those not having animal contact (85.6%), there was a significant difference between both groups ($P< 0.05$). Illiterate people hadhigher rate of infection (94.7%) than the rate of tertiary educated people and those at university(82%), the difference between education and rate of infection was statistically significant($P<0.05$).

Table (1): Prevalence of *Cryptosporidium* spp. among the population in relation to some variables (No= 700).

Variables		Cryptosporidiosis		
		Number of infected	**%**	***P* value Chi-square**
Gender	Male	442/500	88.4	$P> 0.05$ $x^2 = 1.104$
	Female	171/200	85.5	
Age (years)	8 month -10	140/174	80.5	$P< 0.05$ x2 =13.524
	11 -20	71/84	84.5	
	21 – 30	192/212	90.5	
	31 – 40	110/120	91.7	
	> 41	100/110	91	
Consistency of Stool	Formed	404/453	89.2	$P> 0.05$ $x^2 =0.166$
	Liquid	209/247	84.6	
Number of Family size	1 – 5	135/187	72.2	$P< 0.05$ $x^2 = 57$
	6 – 10	383/407	94.1	
	More than 10	95/106	89.6	
Source of Drinking water	Municipal tap water	497/524	94.8	$P< 0.05$ $x^2 = 101.36$
	Commercial bottled water & filtered tap water	116/176	65.9	
Residency	Urban	240/320	75	$P< 0.05$ $x^2 = 149.19$
	Suburban	373/380	87.6	
Occupation	Worker	305/353	86.4	$P> 0.05$ $x^2 = 1.093$
	Other	218/247	88.3	
	Student	90/100	90	
Contact with animal (livestock):	Yes	240/264	90.9	$P< 0.05$
	No	373/436	85.6	$x^2 = 4.338$
Level of Education:	Illiterate	125/132	94.7	$P< 0.05$ $x^2 = 8.811$
	Primary education	254/289	87.9	
	Secondary education	159/188	84.6	
	Tertiary and university	75/91	82	

*Statistically significant at $p< 0.05$.

4. DISCUSSION

Cryptosporidium spp. are found throughout the world, particularly in undeveloped and developing countries (Pumipuntu and Piratae, 2018). Transmission occurs primarily through the fecal-oral route via contaminated food and water, which may be directly and indirectly across contact with animal feces and cross contamination , which occur when hosts ingest oocysts of *Cryptosporidium*, (Pumipuntu and Piratae, 2018; Innes *et al.*, 2020)

The current study is a first one to address the occurrence of *Cryptosporidium* oocysts using modified Ziehl Neelsen stain and its relationship to some risk factors among humans in Zakho district, Iraq. The first case of cryptosporidiosis was reported in Iraq in 1996 (Mahdi *et al*., 1996). After that several studies were conducted in different areas of the country for estimating the prevalence of the Cryptosporidiosis among urban and rural population (Abdulsada, 2015;Ahmed *et al*., 2016; Azeez and Alsakee, 2017; Al-Saeed *et al*., 2020; Alkhanaq and Thamer, 2022). The high rate of infection recorded in the present study (87.57%) is in line with other studies conducted in various parts of Iraq, such as in Mid-Euphrates Area with an infection rate of 92.2% which was slightly higher than the present rate (Abdulsada, 2015). While lower rates of infection were reported in studies conducted in Al-Najaf City (58%) by Sayal (2019), Baghdad Province (47.33%) by Merdaw *et al*. (2018), Basra Province (23.8%) by Salim (2018) and Kirkuk City (22.68%) by Salman *et al*. (2015). The variation in the rate of infection may be due to the geographical and cultural differences, sample size differences, besides the fact that most studies focused on infants and children as well as different techniques used for diagnosis.

The present result showed a slightly higher rate of infection among males as compared to females, but the difference between both genders was non-significant($P>0.05$). This is in agreement with other studies in Iraq, in Erbil City,Basra Province and Karbala Provinces (Khoshnaw *et al.,* 2017; Salim, 2018 : AL-Yasary and Faraj, 2021) with infection rates (20.95% vs. 19.85), (24.2% vs. 23.5%) and (27.41% vs. 23.68%) respectively. The slightly different rate of infection among males may be due to the fact that males are more socially active than females, as well as females have a better immune system than males due to their genetic structural difference organized as microRNAs located on the female X chromosome (Klein and Flanagan, 2016). On the other hand, females pay more care to cleaning and hygienic habits. while, Sayal, (2019) and Salim and Al-Aboody (2019) in Al-Najaf City and Thi-Qar Province reported a higher rate in males as compared to females (55.2% vs 44.8%) and (58% vs 42%) respectively. Also, Al-Shamiri *et al*. (2010) in Yemen reported a higher rate in males as compared to females children (30.7% and 20%) respectively . This may be due to a close contact of male children with the contaminated play grounds outdoor area with their outdoor activities and getting contact with animals and soil, which can raise the risk of parasite transmission.

The present study showed a significant relationship between cryptosporidiosis and age. Age groups from 31 to over 41 years showed the higher rate of the infection, while the lowest rate of infection was among children from 8 months to 10 years. These results are in agreement with other studies conducted in southwest Uganda, southwestern China and Beni-suef, Egypt (Yang *et al*., 2017; Abdel Gawad *et al*., 2018; Nakibirango *et al.,* 2019), all of them reported the highest rate of cryptosporidiosis among age groups of 41-50 years and 31-40 years with rates of 17.3%, 25%, and 37.7%, respectively as compared to other ages. The precise cause of this higher level is unknown, but it could be due to immuno-physiological and ethological differences, as well as a low immune response, particularly among those over the age of 40.

Although, the rate of infection was less among children compared to other age group, but still the rate is high due to the fact that children were more exposed to the infective stage of the parasite by swimming in rivers and swallowing water especially in summer and less awareness. Besides that, most infected participates belong to age group (31-40 years) which had more close contact with domestic animals, which was thought to be a risk factor for infection, as the oocyte of the *Cryptosporidium* released form infected animals and transmitted to the human (Pumipuntu and Piratae, 2018). While the present results disagree with studies performed in Kirkuk City, Iraq, and Buner District, Pakistan, since they reported higher rates of infection among children less than 6 years which were 25.45% and 41.0% (Salman *et al.*, 2015; Khan *et al*., 2019), respectively.

According to the nature of the stool, the present results did not show any significant relationship between the consistency of stool and prevalence of *Cryptosporidium* spp. although, slightly higher prevalence was reported among formed stool specimens. This may be due to the fact that this parasite invades the jejunum and ileum of immunocompromised patients resulting in liquid diarrhea besides that it infects healthy individuals as well without producing any symptoms, or it is most likely due to the fact that formed stool specimens constituted the majority of the samples in the current study. These results are in contradiction with a study conducted by Koyee and Faraj (2015) in Erbil city, they reported that the rates of infection were higher among diarheal stool than formed stool (36.36% vs 6.66%). Also studies conducted in Asia, Buner District in Pakistan, and southern region of Cameron reported higher rates of infection in liquid stool than formed stool which were, 35% vs. 15.6%, 41.0% vs. 16.6%, and 13.40% vs. 2.2%, respectively (Dabas *et al.,* 2017; Khan *et al.,* 2019 ;Tombang *et al.,* 2019).

Regarding the number of family size, current results indicated the presence of a significant relationship between family size and rate of *Cryptosporidium* infection that showed higher infectivity rates among families with more than 6 members . This agrees with the study done in Bagdad City in Iraq by Al-warid (2012) who reported a higher rate of infection among families composed of more than 15 individuals with a rate of 28.32% . The high rates among large families/household, especially when there are one or more infected child in the family facilitates the transmission of the infection between the family members through participation in the use of home appliance, sharing food, changing diapers contaminated with the oocysts of the parasite as compared with small families (Medema and Schijven, 2001; Becker *et al.,* 2015).

In Zakho, the major source of drinking water is the tap water and this can indicate the importance of waterborne transmission in Zakho. The present result indicated that there was a significant relation between the sources of drinking water and the prevalence of *Cryptosporidium* infection, as the infectivity rate was more among people who drink municipal tap water than those drinking commercial bottled water and filtered municipal tap water at home . These are in agreement with other studies conducted in Sao Paulo City in Brazil, Beni-suef in Egypt , and Southern region in Cameron (Carvalho-Almeida *et al*., 2006; Abdel Gawad *et al.,* 2018; Tombang *et al*., 2019) who stated that tap water is associated with the higher rate of infection among people compared to mineral water gallon (55% vs. 12%), (22.5 % vs. 18.8%) and (7.14% vs. 0.89%), respectively. This could be because *Cryptosporidium* oocysts can survive chlorine treatment in water for months and can resist for 180 days in water and up to a year at 4 °C (Medema and Schijven, 2001). While, disagreed with the study conducted in Wasit Province, Iraq by Alkhanaq and Thamer (2022) who reported a higher rate among people drinking bottled water than those drinking tap water (54% vs 52%)

Regarding residency, the rate of cryptosporidiosis was higher among suburban residents than urban ones with a significant relation between them. This finding is consistent with the studies conducted in Beni- Suef, Egypt and Al-Diwaniyah and Wasit cities, Iraq (Abdel Gawad *et al.,* 2018; Mohammad, 2018; Alkhanaq and Thamer, 2022) who reported higher rates of infection in rural residents compared to urban ones (59.5% vs. 40.5%), (63.3% vs. 36.6%) and (64.7% vs. 40%) respectively. This may be due to the fact that rural environment is thought to be favorable for intestinal parasite transmission due to a lack of proper sanitary facilities, limited access to safe water, and frequent animal exposure (Mor and Tzipori, 2008; Daniels *et al*., 2015). However, urban areas are also at risk due to the possibility of water supply system contamination. While it disagrees with studies conducted in Erbil City, Iraq (Khoshnaw *et al.,* 2017), as they did not record any significant association between cryptosporidiosis and residency (rural 21.1% vs. urban 19.7%).

On the other hand, non-significant association was reported among the occupation and *Cryptosporidium* infection rate which disagrees with the study done by Nakibirango *et al*., (2019) who reported the rate of cryptosporidiosis in southwest Uganda with a higher rate of infection among peasants (50.7%) compared to students (2.2%).

In the current study people in contact with animals showed higher infectivity rate than those not getting contact with animals with a significant relationship between prevalence of *Cryptosporidium* and contact with animals. This is in agreement with the studies conducted in Baghdad City in Iraq, Iran and Sothern Egypt (Al-Warid, 2012; Kalantari *et al.,* 2018; Elshahawy and AbouElenien, 2019) who documented that the infectivity rates were higher among people getting contact with animals than those without contact (19.49% vs.13.89%), (24.70% vs. 15.20%) and (11.6% vs. 1.6%) with a significant relation between them. Similarly, Iqbal *et al.,* (2011) in Kuwait and Firoozi *et al.,* (2019) in Iran, in their studies confirmed that cryptosporidiosis is considered as one of the zoonotic diseases transmitted from animals to human especially from cattle, dogs and cats due to a close contact between human and these animals leading to the contamination of hands and food. Some studies reported that cattle release a large number of parasite oocytes compared to cats and dogs (Daniels *et al.,* 2015; Khan *et al.,* 2019). While Carvalho-Almeida *et al*.(2006) did not find any significant correlation between the rate of *Cryptosporidium* infection and animal exposure because they found a very slight difference between presence of animals in home with their absence (28.57% vs. 28.20%).

The current study indicated a significant relationship between the rate of *Cryptosporidium* infection and educational level that observed the highest rate among illiterates than literates. These results agreed with the study conducted by Elshahawy and AbouElenien (2019) in southern Egypt who documented that there was a significant relationship between the level of education and the rate of infection with higher infectivity rate among non-educated than university education (35.20% vs. 9.30%), respectively. This may be due to the fact that literate people practice better hygiene than illiterate people which leads to preventing and reducing infection transmission. While the current results disagree with the study conducted in Wasit City, Iraq by Alkhanaq and Thamer (2022) who observed a higher rate of infection among educated participants than non-eduacted ones (80% vs. 50.8%) respectively.

5. CONCLUSIONS

This study showed the importance of modified Ziehl Neelsen stain in the microscopic identification of cryptosporidiosis. Furthermore, the study reported a high rate (87.57%) of cryptosporidiosis among Zakho population.The age, family size, water source, residency, contact with animals and the level of education have significant risk effects on the prevalence of this disease. Therefore, improvements of family living conditions, environmental sanitation, safe water supply to the population, keeping animals out of human settings, and health education will have a great impact in reducing the rate of infection.

ACKNOWLEDGEMENTS

The authors express their sincere thanks to the Department of Biology, Faculty of Science, the University of Zakho for offering the laboratory facilities and financial support. Great thanks and appreciation to the ethical committee of Zakho General Directorate of Health and to the School staff for permitting us to take samples from enrolled patients and students.

CONFLICT OF INTEREST

There is no conflict of interest in the current study.

REFERENCES

Abdel Gawad, S. S., Ismail, M. A. M., Imam, N. F. A., Eassa, A. H. A., & Abu-Sarea, E. Y. (2018). Detection of *Cryptosporidium* spp. in Diarrheic Immunocompetent Patients in Beni-Suef, Egypt: Insight into Epidemiology and Diagnosis. *The Korean Journal of Parasitology*, *56*(2), 113–119.

Abdulsada, K. M.(2015). Molecular and Epidemiological Study of *Cryptosporidium* spp. in Mid-Euphrates Area. *Kufa Journal for Nursing Sciences*, 5(1), 179-189.

Ahmed, H. S., Abd, A.H., Mohammed, N.Q. (2016). Detection of *Cryptosporidium parvum* from feces samples of human and camels by using direct Polymerase Chain Reaction assay technique. *Al-Qadisiyah Journal of Veterinary Medicine Sciences*, *15*(2), 59–62.

Alkhanaq, M. N., & Thamer, G. (2022). Prevalence of *Cryptosporidium* spp . among Patients with Diarrhea at Wasit Province / Iraq.*Indian Journal of Forensic Medicine & Toxicology*,16(1), 771–780.

Al-Saeed, A. T., Abdo, J. M., & Gorgess, R. G. (2020). Cryptosporidiosis in Children in Duhok City / Kurdistan Region / Iraq. *JPMA. The Journal of the Pakistan Medical Association*, *70*(7), 1251–1255. https://doi.org/10.5455/JPMA.21273

Al-Shamiri, A. H., Al-Zubairy, A. H., & Al-Mamari, R. F. (2010). The prevalence of *Cryptosporidium* spp. in children, Taiz District, Yemen. Iranian journal of parasitology, 5(2), 26

Al-Warid, H. S., Al-Saqur, I. M., & Mahmood, S. H. (2012). Occurrence of *Cryptosporidium* spp. among people live in North of Baghdad. *European Journal of Scientific Research*, 78(4), 539-545.

AL-Yasary, J. T. O., & Faraj, A. A. (2021). Comparison Study about Selected Human Infection of Zoonotic Cryptosporidiosis by Conventional Diagnostic Methods in Karbala Province, Iraq. *The Iraqi Journal of Veterinary Medicine*, *45*(1), 51–55. https://doi.org/10.30539/ijvm.v45i1.1042

Aniesona, A. T., & Bamaiyi, P. H. (2014). Retrospective study of cryptosporidiosis among diarrhoeic children in the arid region of north-eastern Nigeria. *Zoonoses and Public Health*, *61*(6), 420–426. https://doi.org/10.1111/zph.12088

Azeez, S. S., & Alsakee, H. M. (2017). *Cryptosporidium* spp . and rotavirus gastroenteritis and change of incidence after rotavirus vaccination among children in Raparin Pediatrics Hospital , Erbil , Iraq. *Medical Journal of Indonesia*, *26*(3), 190–197.

Becker, D. J., Oloya, J., & Ezeamama, A. E. (2015). Household socioeconomic and demographic correlates of *Cryptosporidium* seropositivity in the United States. PLoS neglected tropical diseases, 9(9), e0004080.

Bodager, J. R., Parsons, M. B., Wright, P. C., Rasambainarivo, F., Roellig, D., Xiao, L., & Gillespie, T. R. (2015). Complex epidemiology and zoonotic potential for *Cryptosporidium suis* in rural Madagascar. *Veterinary Parasitology*, *207*(1–2), 140–143. https://doi.org/10.1016/j.vetpar.2014.11.013

Carvalho-Almeida, T. T., Pinto, P. L. S., Quadros, C. M. S., Torres, D. M. A. G. V, Kanamura, H. Y., & Casimiro, A. M. (2006). Detection of *Cryptosporidium* sp. in non diarrheal faeces from children, in a day care center in the city of São Paulo, Brazil. *Revista Do Instituto de Medicina Tropical de Sao Paulo*, *48*(1), 27–32. https://doi.org/10.1590/s0036-46652006000100006

Clarke, J. J. (1895). Memoirs: A Study of Coccidia met with in Mice. *Journal of Cell Science,* 2(147), 277-283.

Dabas, A., Shah, D., Bhatnagar, S., & Lodha, R. (2017). Epidemiology of *Cryptosporidium* in pediatric diarrheal illnesses. *Indian pediatric*s, 54(4), 299-309.

Daniels, M. E., Shrivastava, A., Smith, W. A., Sahu, P., Odagiri, M., Misra, P. R., Panigrahi, P., Suar, M., Clasen, T., & Jenkins, M. W. (2015). *Cryptosporidium* and *Giardia* in Humans, Domestic Animals, and Village Water Sources in Rural India. *The American Journal of Tropical Medicine and Hygiene*, *93*(3), 596–600. https://doi.org/10.4269/ajtmh.15-0111

Elshahawy, I., & AbouElenien, F. (2019). Seroprevalence of *Cryptosporidium* and risks of cryptosporidiosis in residents of Sothern Egypt: A cross-sectional study. *Asian Pacific Journal of Tropical Medicine*, *12*(5), 232–238. https://doi.org/10.4103/1995-7645.259244

Fayer, R., Santín, M., & Macarisin, D. (2010). *Cryptosporidium ubiquitum* n. sp. in animals and humans. *Veterinary Parasitology*, *172*(1–2), 23–32. https://doi.org/10.1016/j.vetpar.2010.04.028

Feng, Y., Ryan, U. M., & Xiao, L. (2018). Genetic diversity and population structure of *Cryptosporidium*. *Trends in parasitology,* 34(11), 997-1011.

Firoozi, Z., Sazmand, A., Zahedi, A., Astani, A., Bafghi, A. F., Salmi, N. K., Ebrahimi, B., Tafti, A. D., & Ryan, U. (2019). Prevalence and genotyping identification of *Cryptosporidium* in adult ruminants in central Iran. *Parasites & Vectors*, 12(1), 1-6. https://doi.org/10.1186/s13071-019-3759-2

Gerace, E., Presti, V. D. M. L., & Biondo, C. (2019). *Cryptosporidium* infection: epidemiology, pathogenesis, and differential diagnosis. *European Journal of Microbiology and Immunology*, 9(4), 119-123.

Ghazy, A., Abdel-Shafy, S., & Shaapan, R. (2015). Cryptosporidiosis in Animals and Man: 1. Taxonomic Classification, Life Cycle, Epidemiology and Zoonotic Importance. *Asian Journal of Epidemiology*, 8(3), 48–63. https://doi.org/10.3923/aje.2015.48.63

Ghoshal, U., Jain, V., Dey, A., & Ranjan, P. (2018). Evaluation of enzyme linked immunosorbent assay for stool antigen detection for the diagnosis of cryptosporidiosis among HIV negative immunocompromised patients in a tertiary care hospital of northern India. *Journal of Infection and Public Health*, *11*(1), 115–119.

Innes, E. A., Chalmers, R. M., Wells, B., & Pawlowic, M. C. (2020). A One Health Approach to Tackle Cryptosporidiosis. *Trends in Parasitology*, *36*(3), 290–303. https://doi.org/10.1016/j.pt.2019.12.016

Iqbal, J., Khalid, N., & Hira, P. R. (2011). Cryptosporidiosis in Kuwaiti children: association of clinical characteristics with Cryptosporidium species and subtypes. *Journal of Medical Microbiology*, 60(5), 647–652. https://doi.org/10.1099/jmm.0.028001-0

John DT, Petri W. (2006). Medical parasitology. In: John DT, Petri W, eds. A Book. 9th ed.USA: Elsevier Inc.

Kalantari, N., Ghaffari, S., & Bayani, M. (2018). *Cryptosporidium* spp. infection in Iranian children and immunosuppressive patients: A systematic review and meta-analysis. *Caspian Journal of Internal Medicine*, *9*(2), 106–115. https://doi.org/10.22088/cjim.9.2.106

Khan, A., Shams, S., Khan, S., Khan, M. I., Khan, S., & Ali, A. (2019). Evaluation of prevalence and risk factors associated with *Cryptosporidium* infection in rural population of district Buner, Pakistan. *PloS One*, *14*(1), e0209188.https://doi.org/10.1371/journal.pone.0209188

Khoshnaw, K.H.S., Majeed, P.D., Hawezy, A.A. (2017). Prevalence of *Cryptosporidium* spp . among hospitalized children with diarrhea using ELISA and conventional microscopic techniques , in Erbil province. *Polytech. J*. 1–11.

Klein, S. L., & Flanagan, K. L. (2016). Sex differences in immune responses. *Nature Publishing Group*, 371(6527), 347-348. https://doi.org/10.1038/nri.2016.90

Kotloff, K. L., Nataro, J. P., Blackwelder, W. C., Nasrin, D., Farag, T. H., Panchalingam, S., Wu, Y., Sow, S. O., Sur, D., & Breiman, R. F. (2013). Burden and aetiology of diarrhoeal disease in infants and young children in developing countries (the Global Enteric Multicenter Study, GEMS): a prospective, case-control study. *The Lancet*, 382(9888), 209–222.

Koyee, Q. M., & Faraj, A. M. (2015). Prevalence of *Cryptosporidium* spp. with other intestinal microorganisms among regular visitors of Raparin Pediatric Hospital in Erbil City-Kurdistan region, Iraq. *Zanco Journal of Pure and Applied Sciences*, 27(4), 57-64.

Krumkamp, R., Aldrich, C., Maiga-Ascofare, O., Mbwana, J., Rakotozandrindrainy, N., Borrmann, S. & Eibach, D. (2021). Transmission of *Cryptosporidium* species among human and animal local contact networks in sub-saharan Africa: a multicountry study. *Clinical Infectious Diseases,* 72(8), 1358-1366.

Mahdi, N. K., Al-Sadoon, I. A., & Mohamed, A. T. (1996). First report of cryptosporidiosis among Iraqi children. In *Eastern Mediterranean Health Journal*, 2(1), 115–120. https://doi.org/10.26719/1996.2.1.115

Medema, G. J., & Schijven, J. F. (2001). Modelling the sewage discharge and dispersion of *Cryptosporidium* and *Giardia* in surface water. *Water Research*, *35*(18), 4307–4316. https://doi.org/10.1016/s0043-1354(01)00161-0

Merdaw, M. A., Al-Zubaidi, M. T. S., Hanna, D. B., Khalaf, I. A., Jassim, H. S. (2018). Genotyping of *Cryptosporidium* Spp . Isolated from Human and Cattle in Baghdad Province , Iraq. *Indian Journal of Natural Sciences*, *9*(51), 15925–15932.

Mohammad, F. I. (2018). Detecting of virulence factors COWP gene and CP15 gene for *Cryptosporidium parvum* by polymerase chain reaction (PCR). *Al-Qadisiyah Journal Of Pure Science*, *23*(2), 39-47. https://doi.org/10.29350/jops.2018.23.2.736

Mor, S. M., & Tzipori, S. (2008). Cryptosporidiosis in children in Sub-Saharan Africa: a lingering challenge. *Clinical Infectious Diseases*, *47*(7), 915–921.

Nakibirango, J., Mugenyi, V., Nsaba, D., Nsimemukama, A., Rugera, S. P., & Okongo, B. (2019). Prevalence of cryptosporidiosis and hygiene practices among HIV/AIDS patients in southwest Uganda. *HIV/AIDS - Research and Palliative Care*, *11*, 141–145. https://doi.org/10.2147/HIV.S206195

Pumipuntu, N., & Piratae, S. (2018). Cryptosporidiosis: A zoonotic disease concern. *Veterinary World*, *11*(5), 681–686. https://doi.org/10.14202/vetworld.2018.681-686

Ryan, U., Fayer, R., & Xiao, L. (2014). *Cryptosporidium* species in humans and animals: current understanding and research needs. *Parasitology*, *141*(13), 1667–1685. https://doi.org/10.1017/S0031182014001085

Ryan, U., & Hijjawi, N. (2015). New developments in *Cryptosporidium* research. *International Journal for Parasitology*, *45*(6), 367–373. https://doi.org/10.1016/j.ijpara.2015.01.009

Ryan, U. M., Feng, Y., Fayer, R., & Xiao, L. (2021). Taxonomy and molecular epidemiology of *Cryptosporidium* and *Giardia*–a 50 year perspective (1971–2021). *International Journal for Parasitology*, 51(13-14), 1099-1119.

Salim, M. (2018). Epidemiological study on *Cryptosporidium* among children in Basra Province-Iraq. *Journal of Physics: Conference Series*, 1032(1), 012072. https://doi.org/10.1088/1742-6596/1032/1/012072

Salim, A. R., & Al-Aboody, B. A. (2019). Molecular Detection and Prevalence of *Cryptosporidium parvum*, Among Patients with Diarrhea at Al-Rifai City/Thi-Qar Province. *Iraqi Journal of Biotechnology*, 18(2).

Salman, Y. J., Sadek, W. S., & Rasheed, Z. K. (2015). Prevalence of *Cryptosporidium parvum* among Iraqi displaced people in Kirkuk city using direct microscopy, flotation technique and ELISA-copro antigen test. *Int. J. Curr. Microbiol. App. Sci*, *4*(11), 559–572.

Sayal, R. A. (2019). Epidemiological Study of *Cryptosporidium* Infection in Al-Najaf City. *Int J Pharm Qual Assur,* 10(1), 128–131. https://doi.org/10.25258/ijpqa.10.1.20

Smith, H. V, Cacciò, S. M., Cook, N., Nichols, R. A. B., & Tait, A. (2007). *Cryptosporidium* and *Giardia* as foodborne zoonoses. *Veterinary Parasitology*, *149*(1–2), 29–40. https://doi.org/10.1016/j.vetpar.2007.07.015

Thomson, S., Hamilton, C. A., Hope, J. C., Katzer, F., Mabbott, N. A., Morrison, L. J., & Innes, E. A. (2017). Bovine cryptosporidiosis: impact, host-parasite interaction and control strategies. *Veterinary Research*, *48*(1), 1-16. https://doi.org/10.1186/s13567-017-0447-0

Tombang, A. N., Ambe, N. F., Bobga, T. P., Nkfusai, C. N., Collins, N. M., Ngwa, S. B., Diengou, N. H., & Cumber, S. N. (2019). Prevalence and risk factors associated with cryptosporidiosis among children within the ages 0 – 5 years attending the Limbe regional hospital , southwest region , Cameroon. *BMC Public Health*, *19*(1), 1-10.

Tyzzer, E. E. (1907). A sporozoan found in the peptic glands of the common mouse. *Proceedings of the Society for Experimental Biology and Medicine*, 5(1), 12-13.

Yang, Y., Zhou, Y., Xiao, P., Shi, Y., Chen, Y., Liang, S., & Yihuo, W. (2017). Prevalence of and risk factors associated with *Cryptosporidium* infection in an underdeveloped rural community of southwest China. *Infectious Diseases of Poverty*, 6(1), 1–10. https://doi.org/10.1186/s40249-016-0223-9