Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

Evaluation For Sociodemographic Risk Factors Associated With *Cryptosporidium Parvum* Infection In Children Under Five Years

Ali Ibrahim Ali AL-Ezzy* , Abeer Thair Khadim

Department of pathology, College of Veterinary Medicine, University of Diyala, Iraq.
Department of Medicine, College of Veterinary Medicine, University of Diyala, Iraq.
Corresponding author : alizziibrahim@gmail.com

Received: 1 -4-2021 Accepted: 20-5-2021 Published: 1-7-2021

Abstract :

Aims : Current study aims to investigate risk sociodemographic risk factors associated with cryptosporidium parvum infection in diarrheic children under five years in Baqubah-Diyala province

Methods : fecal samples were collected from (100)diarrheic children .Samples were stained by Lugol's iodine solution; Ziehl–Neelsen staining (ZN) ,cold &hot techniques for detection of C.parvum . Questions about Age ,sex ,family economic status, parents education level , residence were recorded and analyzed

Results :

The children who reside in villages have (1.160) possibility of C.parvum infection compared with those the city. The children who descended from families with low and middle economic status have the (1.016) &(1.022)chance as a possibility of C.parvum infection .The children who descended from families with intermediate and secondary education for fathers have (1.361) and (1.178) chance of *C.parvum* infection. The children who descended from families with primary ,intermediate and higher education for mothers have (2.143) ; (1.031) and (1.455) chance of *C.parvum* infection .

Conclusion :

Living in villages, economic status ,parents education increase the possibility of infection with *C.parvum* among children

Keywords :cryptosporidium parvum ,children, diarrhea ,Iraq

How to cite the article :
AL-Ezzy AIA, Khadim AT. Evaluation For sociodemographic Risk Factors associated with Cryptosporidium Parvum Infection In Children under Five years. Diyala Journal For Veterinary Sciences. 2021;1(2):100-14.

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

Introduction

Cryptosporidium is intracellular protozoan that live in the gastrointestinal tract of human and many vertebrates animals[1]. They are the widespread source of diarrheal disease among both immunocompetent and immunocompromised individuals throughout the world leading to considerable morbidity and mortality, especially in individuals less than five years[2] . The parasite is protected by an outer shell that allows it to survive outside the body for long periods of time and makes it very resistant to chlorine disinfectants[2]. Most cases of human cryptosporidiosis are due to infections with the human specific *C. hominis* or the zoonotic *C. parvum*[3]. Current evidence indicates that ruminants are a reservoir of zoonotic Cryptosporidium from where humans get infected by contaminated food and water or through direct contact with livestock, for example animal handlers [1, 2] .*C. hominis and C. parvum* are internationally the most commonly species infecting humans. Humans can acquire cryptosporidium infections through several transmission routes such as person to person transmission, zoonotic transmission, food borne transmission and waterborne transmission[4].A single oocyst is sufficient to cause infection and disease[5, 6]. When excreted, Oocysts are directly infectious and are able to survive for up to 6 months in a moist and cool environment. In water, oocysts remain viable for 140 days[7]. In immunocompetent persons, cryptosporidium infection usually asymptomatic. in children under the age of five and in immunosuppressed people, the infection leads to severe diarrhea. Nausea, vomiting, discomfort and low-grade fever are other clinical symptoms which may occur during an infection with Cryptosporidium [8] . Symptoms in immunocompromised patients can be very severe and even death has been described [9].In developing countries 45% of the children are experiencing an infection before the age of two [8, 9]

Current study aims to investigate sociodemographic risk factors associated with *C.parvum* infection in diarrheic children under five years in Baqubah-Diyala province

Material and Methods

Study area and study population

This study was conducted on newborn to less than 5 years old Iraqi children, living in the Baqubah city -Diyala province 33°45'34.71 N; 44°36'23.97 E[3, 10]

Ethical Approval :

This study was conducted according to the principles of Helsinki declaration. Before endoscopy, a full explanation about the purpose of this study to all patients was done. Dully-filled consent form obtained from all patients that agree to participate in the study[11, 12]. Approval of ethical review Committee of College of Veterinary medicine – Diyala University - Iraq, was taken prior to initiation of the work.

Stool Samples collection and processing

The stool samples were collected from 100 children less than 5 years of age suffering from gastrointestinal illness[13, 14]. Sample collection took place from November 2016 to June 2017. The inclusion criterion was diarrhoea, defined as passage of three or more loose or liquid stools per day, or more frequently than is normal for the individual [7, 15]

The samples were obtained from Albatul teaching hospital-Baqubah . An ethical consideration and consent by the parents or guardians of the children was signed before getting the samples. The samples were collected in a special tightly capped leak proof containers. Each sample was labeled with the child's name, Sex and age[12, 15]. Additional information about each sample was obtained

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

from the hospital (place of residency and patient's hospital number)[9, 12].

Each sample was divided into two portions, one used for immediate examination ,other one preserved and stored in 10% formalin[16]. One volume of the fecal sample was mixed thoroughly using wooden applicator stick, with 3 volumes of 10% formalin[7, 8]. The sample was mixed again, and the specimen containers were sealed well. All samples were reinforced with parafilm, the container was inserted in a plastic bag, and samples were stored at 4ºC in the clinical pathology laboratory at college of veterinary medicine ,Diyala university .

Methods:

Direct Microscopy

I. Lugol's iodine solution

All stool samples were examined directly by emulsifying a small portion (approximately 0.1 ml) in a drop of Lugol's iodine on a separate slide according to [3, 7]

Staining Techniques

I. Ziehl–Neelsen staining (ZN)

a) Cold method of ZN Staining Of fecal smears (Modified Kinyoun's Acid-Fast Stain) according to [8, 17].
b) Hot method of ZN staining of fecal smears according to [8, 16, 17].

Questioners :

A list of Questions for each participant about age ,sex ,family economic status, parents education level , residence were recorded and analyzed[4, 18]

Statistical analysis:

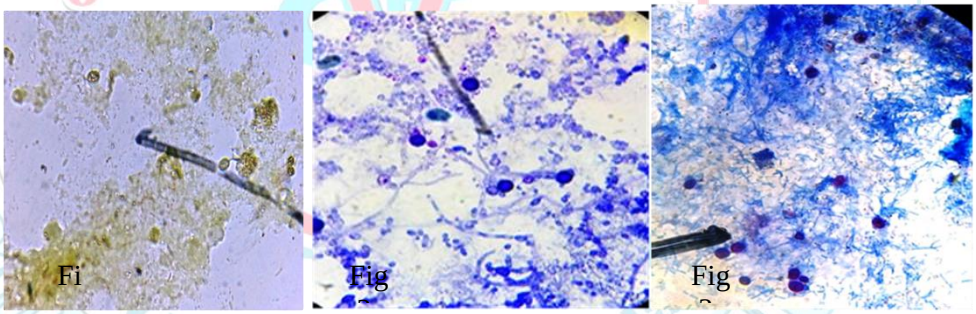
Person chi square test for categorical data used for correlation[19, 20]. The level of significance was 0.05(two-tail) in all statistical testing[9, 16, 21]; significant of correlations include also 0.01 (two-tail). The level of confidence limits was 0.095.Statistical analysis performed using SPSS for windows TM version 17.0[11, 13]

Results :

As shown in table (1) and figures (1,2&3),the minimum age of children presented with diarrhea was (1 month),while the maximum age was (48 month).The main affected age group was (1-6) month, in which (50%) was infected with *C.parvum*, followed by (7-12) month, in which (13%) was infected with *C.parvum.* The third age group was (31-36), in which (6%) was infected. Neither significant difference(p value=**0.442)** ,nor significant correlation was reported between positivity of *C.parvum* and age group of children (p value=**0.367)** .As shown in table (2),Male presented with diarrhea was (62%),in which (45%) was infected with *C.parvum* .Females represent (38%) of children presented with diarrhea ,in which (29%) was infected with *C.parvum.* . Neither significant difference(p value=0.679) ,nor significant correlation was reported between positivity of *C.parvum* and Sex of children (p value=0.683) .As shown in table (3), (38%) of children were reside in villages compared with (62%) in the city . *C.parvum* infection was reported in (27%) of children whose residence at villages compared with 47%) for those in the city . Neither significant difference(p value=**0.599**) ,nor significant correlation was reported between positivity of *C.parvum* and residence (p value=0.603).On the other hand the children who reside in villages have the greater possibility of infection with *C.parvum* in (1.160) time compared with those who reside at the city . As shown in table (4) ,(32%) of children with *C.parvum* infection descend from families with low economic status compared with (39%) from middle economy and (7%) from families with high economic status . Neither significant difference(p value=**0.964**) ,nor significant correlation was reported between positivity of *C.parvum* and economic Status (p value=**0.867**). The chil-

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

dren who descended from families with low and middle economic status have the greater possibility of infection with *C.parvum* in (1.016) and (1.022) time subsequently compared with those of high economic status (0. 0.949) As shown in table (5), (11%) of children with *C.parvum* infection descend from families with primary education for fathers compared with (24%) from families with intermediate education for fathers and(28%) from families with secondary education for fathers and (11%) were have higher education . Neither significant difference(p value=0.311) ,nor significant correlation was reported between positivity of *C.parvum* and education level (p value=0.802).On the other hand the children who descended from families with intermediate and secondary education for fathers have the greater possibility of infection with *C.parvum* in (1.361) and (1.178) time subsequently compared with those of primary education (0.259) and higher education for fathers (0.517) .As shown in table (6), 25% of children with *C.parvum* infection descend from families with primary education for mothers compared with (29%) from families with intermediate education for mothers and(12%) from families with secondary education for mothers and (8%) were have higher education. Neither significant difference(p value=0.199) ,nor significant correlation was reported between positivity of *C.parvum* and education level (p value=0.220).On the other hand the children who descended from families with primary ,intermediate and higher education for mothers have the greater possibility of infection with *C.parvum* in (2.143) ; (1.031) and (1.455) time subsequently compared with those of secondary education (0.366) .



Fi Fig Fig

Detection of *C.parvum* infection in children by lugol's iodine wet preparation (100x), Figure 1;ZN cold staining technique (100x) ,Figure (2); ZN hot staining technique (100x) , Figure (3)

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

Table(1): Age As A Possible Risk Factor Associated With *C.parvum* Infection In Children

Description Of Age for Children Presented With Diarrhea		
Minimum (month)	1	
Maximum(month)	48	
Mean± Std. Deviation	10.2950±11.76025	
Age group (month)	Infection with *C.parvum*	
	Positive	Negative
	NO.(%)	NO.(%)
1-6	50(50%)	13(13%)
07-12	13(13%)	6(6%)
13-18	1(1%)	1(1%)
19-24	0(0%)	1(1%)
25-30	1(1%)	2(2%)
31-36	6(6%)	3(3%)
37-42	2(2%)	0(0%)
43-48	1(1%)	0(0%)
Total NO.(%)	74(74%)	26(26%)
$\chi 2$	19.238	
P value	0.44**2**	
R	-0.091	
P value	0.367	

Table(2): Sex As A Possible Risk Factor Associated With *C.parvum* Infection In Children

Sex		Infection with *C.parvum*	
		Positive	Negative
		NO.(%)	NO.(%)
Male	62(62%)	45(45%)	17(17%)
Female	38(38%)	29(29%)	9(9%)
total NO.(%)	100(100%)	74(74%)	26(26%)
$\chi 2$	0.171		
P value	0.679		
R	-.041		
P value	0.683		

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

Table(3): Residence As A Possible Risk Factor Associated With *C.parvum* Infection In Children

Infection with *C.parvum*	Residence		Total
	Village	City	
Negative	11(11%)	15(15%)	26(26%)
positive	27(27%)	47(47%)	74(74%)
Total	38(38%)	62(62%)	100(100%)
Pearson Chi-Square	0.277		
P value	0.599		
R	0.053		
P value	0.603		
Risk Estimate	95% Confidence Interval		
	Value	Lower	Upper
Odds Ratio for Residence (village)	1.160	0.676	1.990
Odds Ratio for Residence (city)	0.908	0.626	1.317

Table(4): Family Economic Status As A Possible Risk Factor Associated With *C.parvum* Infection In Children

Infection with *C.parvum*	Economic Status			Total
	Low	Middle	High	
Negative	10(10%)	14(14%)	2(2%)	26(26%)
Positive	28(28%)	39(39%)	7(7%)	74(74%)
Total	38(38%)	53(53%)	9(9%)	100(100%)
$\chi 2$	0.073			
P value	0.964			
R	0.017S			
P value	0.867			
Risk Estimate	95% Confidence Interval			
	Value	Lower	Upper	
Odds Ratio for low Economic status	1.016	0.576	1.792	
Odds Ratio for middle	1.022	0.674	1.549	

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

Economic status			
Odds Ratio for high Economic status	0.949	0.103	8.723

Table(5): Education Status for children Fathers as a Possible Risk Factors Associated With *C.parvum* Infection

Infection with *C.parvum*		Father Education				Total
		primary	intermediate	secondary	higher education	
Negative		1(1%)	11(11%)	12(12%)	2 (2%)	26(26%)
Positive		11(11%)	24(24%)	28(28%)	11(11%)	74(74%)
Total		12(12%)	35(35%)	40(40%)	13(13%)	100(100%)
$\chi 2$		3.577				
P value		0.311				
R		-0.025				
P value		0.802				
		95% Confidence Interval				
Risk Estimate		Value	Lower	Upper		
Odds Ratio	Primary Education	0.259	0.035	1.908		
	Intermediate Education	1.361	0.775	2.389		
	Secondary Education	1.178	0.712	1.947		
	Higher Education	0.517	0.123	2.182		

Table(6): Education Status for children Mothers as a Possible Risk Factors Associated With *C.parvum* Infection

Infection with *C.parvum*	Mother Education				Total
	primary	intermediate	secondary	higher education	
Negative	5(5%)	10(10%)	9(9%)	2(2%)	26(26%)
Positive	25(25%)	29(29%)	12(12%)	8(8%)	74(74%)
Total	30(30%)	39(39%)	21(21%)	10(10%)	100(100%)
$\chi 2$	4.650				
P value	0.199				
R	-.124				

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

P value		0.220		
		95% Confidence Interval		
Risk Estimate		Value	Lower	Upper
Odds Ratio	Primary Education	2.143	.722	6.359
	Intermediate Education	1.031	.412	2.581
	Secondary Education	0.366	0.132	1.011
	Higher Education	1.455	.288	7.338

Discussion :

In current study, the main affected age group was (1-6) month, in which (50%) was infected with *C.parvum* diagnosed by wet preparation ,ZN hot an d ZN cold techniques , which come in accordance with that reported by [3, 4, 7, 8, 16, 22],they proved that the burden of Cryptosporidium increased in infants obviously between 3 and 6 months of age , a period that corresponds to changes in breast feeding practices. Followed by (7-12) month, in which (13%) was infected with *C.parvum* which come in accordance with [23]. The possible explanation for such higher detection rate , at age of (1-6) month and(7-12) month of children due to the possibility of exposure to the infections by Cryptosporidium spp. because they lack the knowledge about the good food and water. They or their parents may present food without washing their hands, play in soil and sewage water, exposed to more fecal oral contact or through contaminated food or water, or may be attributed to their weak immune responses [24, 25] .

In current study the *C.parvum* reported cases of less than 24 months represent 64% of all cases under investigation which come in line with [26] and lower than that reported by [27, 28] who reported that 77% of children less than two years was infected with cryptosporidium .This is simply because children at this stage try to explore the environment and identify any thing around via mouth ,that give permission to get entry of different pathogens including cryptosporidium spp. and this was proved by [29-31],reported positive correlation between *C.parvum* associated diarrhea and soil contamination with oocysts which reported in 32% to 85% of soil samples under investigation .

The current findings come in opposite with [24], who reported that 40.3% of preschool age group between 2 - 6 years old was infected with cryptosporidium spp. The third age group was (31-36) month, in which (6%) was infected. This come in discordance with [32], in Kuwait ,who found that the prevalence rate 73% represented children of more than two years compared to 27% of children less than two years of age and [24],who recorded that the lowest rate of cryptosporidium infection, (22.7%) was among infants of (1-24 months). In Kuwait [33] reported that (41.4 %) of the Cryptosporidium infected children were in the 4–8-year-old age group. No significant correlation was reported between positivity of *C.parvum* and age group of children (p value=0.367).This result come in accordance with [34-37] ,who reported that children of all age groups were susceptible to infection with Cryptosporidium spp. These results reflect that beside all factors that play a role in infection and all precautions to pre-

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

vent such infection the possibility of attack is still present and no age limitation for infection process .

Sex as a Possible Risk Factors Associated With *C.parvum* Infection In Children

Male presented with diarrhea was (62%),in which (45%) was infected with *C.parvum* .Females represent (38%) of children presented with diarrhea ,in which (29%) was infected with *C.parvum*. This finding come line with [34],reporting that 61% of males were infected parasites including *C.parvum* while the rest 39% of infected children were females. In Sweden [26],agree with finding of current study ,they reported that the rate of cryptosporidium infection was higher among males (45.1%) compared with (34.5%) for females . Current study reported no significant correlation was reported between positivity of *C.parvum* and Sex of children (p value=0.683). This finding come in agreement with that reported by [34, 35, 38-40] . The possible reasons for the absence of sex-related difference in the prevalence among the children might be due to the fact that there is no difference in sex behavior in children and risk factors of *C. parvum* infection such as food consumption, domestic animals contact and etc. Besides, the hygienic practices exercised by children of both sex are also essentially similar[41] .

Residence as a Possible Risk Factors Associated With *C.parvum* Infection In Children

Current study reported that 38% of children were reside in villages compared with 62% in the city . *C.parvum* infection was reported in 27% of children whose residence at villages compared with 47% for those in the city .These results considered high that that reported by another studies in India by [42],reported that cryptosporidium infection in children descended from villages reach to 18% .No significant correlation was reported between positivity of *C.parvum* and residence (p value=0.603).This result come in line with that reported by [37, 38]. The reason for the difference in services provided between the city and the village in terms of sanitation and the high rate of injury in the children of the city is possible to predict the reason for the pollution of drinking water with sewage because of the damage to sewage networks and the possibility of transmission of injury easily. In addition to the high population density in the city compared to rural areas, which contribute in one way or another .

Children who reside in villages have the possibility of infection with *C.parvum* in (1.160) time compared with those who reside at the city which come in line with [27, 28]. This may attributed to several factors includes but not limited to the abundance of rivers and water bodies from the ponds in Diyala province . Besides, the usual presence of cows and sheep as well as wild birds and companion animals which have been implicated as potential reservoirs of infection [43] . Livestock are an important reservoir for Cryptosporidium, which contribute in one way or another in parasite transmission to children, especially if we take into account the playing outside the house in open spaces and on land which may be contaminated with animal fecal materials containing oocysts of *C.parvum* [44, 45] . In addition to that unhealthy habits of using human fecal materials and animal manure as a fertilizers to agricultural lands which causes open air pollution and the possibility of oocysts transfer due to low weight from one place to another through air currents, so it is possible that these children in the villages more vulnerable to infection than their peers in the cities .

Family Economic Status as a Possible Risk Factors Associated With *C.parvum* Infection In Children

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

In current study ,A total (32%) of children with *C.parvum* infection descend from families with low economic status compared with (39%) from middle economy and (7%) from families with high economic status. These results appear to be higher than that reported by [34], in which 10.8% of infected children were descended from families with low economic status, where as 9.8% of high economic status. In Bangladesh [27],reported that 50% of *C.parvum* positive children were descended from families with low economic status. The poverty and living in slum-dwelling areas in Diyala province reflect several points first one, living in poor neighborhoods and displacement camps reflects a painful reality about the absence of basic services and infrastructure, among them, sanitation services and the availability of health care clinics in the adequate number .In addition to add to the overcrowding in the population and the absence of health awareness. last but not least , the quality of water used in drinking in the event that the region is already equipped with .

No significant correlation was reported between positivity of *C.parvum* and economic status which come in agreement with [34]. The children who descended from families with low and middle economic status have the possibility of infection with *C.parvum* (1.016) and (1.022) time subsequently compared with those of high economic status (0.949).These findings come in agreement with [27, 34].This is simply due to whenever the economic status decrease this will accordingly leads to whether directly or indirectly to several effects includes but not limited to general healthy status, nutritional status, which play a role in infection with *C.parvum* and other diarrhea causing pathogens .

Parents Education Status as a Possible Risk Factors Associated With *C.parvum* Infection In Children

A total of (11%) of children with *C.parvum* infection descend from families with primary education for fathers compared with (24%) from families with intermediate education for fathers and (28%) from families with secondary education for fathers and (11%) were have higher education .No significant correlation between positivity of *C.parvum* and education level (p value=0.802) which come in agreement with [35, 46] .On the other hand the children who descended from families with intermediate and secondary education for fathers have the possibility of infection with *C.parvum* in (1.361) and (1.178) time .These results may attributed to the limitation of knowledge about feco-oral and another rout of transmission for the diarrhea causative agents .

As total of (25%) of children with *C.parvum* infection descend from families with primary education for mothers compared with (29%) from families with intermediate education for mothers and(12%) from families with secondary education for mothers and (8%) were have higher education. No significant correlation was reported between positivity of *C.parvum* and education level (p value=0.220) which come in agreement with [27, 35, 46].Children who descended from families with primary ,intermediate and higher education for mothers have the possibility of infection with *C.parvum* in (2.143) ; (1.031) and (1.455). This probably due to limited information of mothers about the diarrhea pathogens ,mode of transmission, prevention and basically the children usually under direct supervision of mothers ,which in turn play a critical role in their guidance and protect them from the bad habits including the oral exploration for any attractive substance which permit entry of diarrhea pathogens including cryptosporidium spp .

Diyala Journal for Veterinary sciences
Open Access Journal Published by College of Veterinary Medicine
University of Diyala, Iraq
P-ISSN: 2410-8863
Vol. 1, NO. 2, June 2021
Proceedings of 2nd National & 1st International Scientific Conference
Of Veterinary Medicine & Science , (NISCVMS-2021)

In conclusion :

Living in villages, economic status ,parents education, Immune suppression, breeding or living in close contact with sheep and cows, hand washing practice ,previous history of diarrhea increase the possibility of infection with *C.parvum* among children

References

[1].Al-Ezzy AIA, Khadim AT, Hassun RH. Evaluation Of Cryptosporidium Parvum Infection In Calves Under One Year With Special Emphasis To Age And Gender In Baqubah-Diyala Province, Iraq. Diyala Journal of Agricultural Sciences. 2018;10(Special Issue).

[2].Al-Ezzy AIA, Khadim AT, Hassun RH. A comprehensive Evaluation of Transmission Methods for Cryptosporidium species with special emphasis to Cryptosporidium Parvum. Research Journal Of Pharmaceutical Biological And Chemical Sciences. 2017;8(5):555-70.

[3].AL-Ezzy AIA, Khadim AT. A Comprehensive Evaluation Of Diagnostic Techniques For Cryptosporidium Species With Special Emphasis To Cryptosporidium Parvum Diyala Journal For Veterinary Sciences. 2021;1(2):64-76.

[4].AL-Ezzy AIA, Khadim AT. Comprehensive Evaluation For The Life Style And Zoonotic Risk Factors Associated With Cryptosporidium Parvum Infection In Children Under Five Years. Diyala Journal For Veterinary Sciences 2021;1(2):77-92.

[5].Food Drug Administration Code F. Annex 3—public health reasons/administrative guidelines—chapter 2, management and personnel. 2009.

[6].Kothavade RJ. Potential molecular tools for assessing the public health risk associated with waterborne Cryptosporidium oocysts. Journal of medical microbiology. 2012;61(8):1039-51.

[7].AL-Ezzy AIA, Khadim AT. Clinical Evaluation for the wet mount preparations versus Ziehl–Neelsen staining modifications for Diagnosis and severity scoring of cryptosporidium parvum in children under 5 years. Diyala Journal For Veterinary Sciences 2021;1(2):126-38.

[8].AL-Ezzy AIA, Khadim AT. Accuracy of Ziehl Neelsen and Safranin Methylene Blue Staining modalities for Detection Of C.parvum Infection In Children under 5 years. Diyala Journal For Veterinary Sciences. 2021;1(2):188-202.

[9].AL-Ezzy; AIA, Khadim AT. Clinical Compatibility Between Negative Stains, Quick Gram Chromotrope, Gram And Giemsa Staining Techniques For Detection Of C.Parvum Infection In Children Under 5 Years Diyala Journal For Veterinary Sciences. 2021;1(2):173-87

[10].Hameed MS, Al-Ezzy AIA, Jalil WI, Ahmed A, Al Khalidi H. Impact of Stress Factors on Physiological Level of Interleukin 10 in Healthy Calves in Diyala Province–Iraq. International Journal of Pharmaceutical Research. 2020;12(2).

[11].Al-Ezzy AIA. Immunopathological and Modulatory Effects of Cag A+ Genotype on Gastric Mucosa, Inflammatory Response, Pepsinogens, and Gastrin-17 Secretion in Iraqi Patients infected with H. pylori. Open access Macedonian journal of medical sciences. 2018;6(5):794.

[12].Al-Ezzy; AIA, Al-Khalidi; AAH, Hameed; MS. Evaluation of C-Reactive Protein in Iraqi Children Presented with Acute Enteropathogenic Escherichia Coli Associated Diarrhea with Special Emphasis to Age and Gender. Gazi medical Journal. 2020;31(2).

[13].AL-Ezzy AIA. In Situ Nick End Labeling as a Molecular Immunopathological Indicator for the Severity of DNA Fragmentation and Gastroduodenal Tissue Damage among H. Pylori Cag APositive Patients. Indian Journal of Science and Technology. 2016;9(2).

[14].Al-Ezzy AIA. Evaluation of Endoscopy based H. pylori Diagnostic Techniques in Iraqi Patients with upper Gastrointestinal Disorders. Indian Journal of Science and Technology. 2016;9(22).

[15].Al-Ezzy AIA. The Accuracy of Elisa Versus Latex Agglutination Tests in Diagnosis of Rotavirus Acute Gastroenteritis and the Clinical Usefulness of C-Reactive Protein in Iraqi Children. South East European Journal of Immunology. 2016;2016:1-5.

[16].AL-Ezzy; AIA, Khadim; AT, Humadi; AA. Clinical Agreements Between Ziehl Neelsen And Methylene Blue Staining Modifications For Detection Of C.Parvum Infection In Claves. Diyala Journal For Veterinary Sciences. 2021;1(2):145-58.

[17].Rekha KMH, Puttalakshmamma GC, D'Souza PE. Comparison of different diagnostic techniques for the detection of cryptosporidiosis in bovines. Veterinary world. 2016;9(2):211.

[18].AL-Ezzy AIA, Abdulameer SJ. Phenotypic Identification And Molecular Characterization Of Gliotoxin producing Aspergillus fumigatus Isolated From Hunters With Special Emphasis To Clinical Manifestations and Risk Factors In Diyala Province–Iraq. Diyala Journal for Veterinary Sciences. 2021;1(2):34-48.

[19].Jameel GH, Al-Ezzy AIA, Mohammed IH. Immunomodulatory, Apoptosis Induction and Antitumor Activities of Aqueous and Methanolic Extract of Calvatia Craniiformis in Mice Transfected with Murine Hepatocellular Carcinoma Cells. Open access Macedonian journal of medical sciences. 2018;6(7):1206.

[20].Al-Ezzy AIA. Immunohistopathological Role Of Bcl2 And P53 Gene Expression In Helicobacter Pylori Cytotoxin-Associated Gene A Positive Versus Cytotoxin-Associated Gene A Negative Antral Predominant non-atrophic gastritis in Iraqi patients. Asian Journal of Pharmaceutical and Clinical Research. 2017:142-8.

[21].Al-Ezzy AIA, Khazzal SA, Qasim AR. Isolation Of Proteus Mirabilis From Urinary Tract Infections Of Human And Ovine In Baqubah-Diyala Province. Diyala Journal of Agricultural Sciences. 2018;10(Special Issue).

[22].Pedersen SH, Wilkinson AL, Andreasen A, Warhurst DC, Kinung'hi SM, Urassa M, *et al.* Cryptosporidium prevalence and risk factors among mothers and infants 0 to 6 months in rural and semi-rural Northwest Tanzania: a prospective cohort study. PLoS neglected tropical diseases. 2014;8(10):e3072.

[23].Tellevik MG, Moyo SJ, Blomberg B, Hjøllo T, Maselle SY, Langeland N, *et al.* Prevalence of Cryptosporidium parvum/hominis, Entamoeba histolytica and Giardia lamblia among Young Children with and without Diarrhea in Dar es Salaam, Tanzania. PLoS neglected tropical diseases. 2015;9(10):e0004125.

[24].Al-Shamiri A, Al-Zubairy A, Al-Mamari R. The prevalence of Cryptosporidium spp. in children, Taiz District, Yemen. Iranian journal of parasitology. 2010;5(2):26.

[25].Mirzaei M. Prevalence of Cryptosporidium sp. infection in diarrheic and non-diarrheic humans in Iran. The Korean journal of parasitology. 2007;45(2):133.

[26].Adler S, Widerström M, Lindh J, Lilja M. Symptoms and risk factors of Cryptosporidium hominis infection in children: data from a large waterborne outbreak in Sweden. Parasitology research. 2017;116(10):2613-8.

[27].Korpe PS, Haque R, Gilchrist C, Valencia C, Niu F, Lu M, *et al.* Natural history of cryptosporidiosis in a longitudinal study of slum-dwelling Bangladeshi children: association with severe malnutrition. PLoS neglected tropical diseases. 2016;10(5):e0004564.

[28].Lemieux MW, Sonzogni-Desautels K, Ndao M. Lessons Learned from Protective Immune Responses to Optimize Vaccines against Cryptosporidiosis. Pathogens. 2017;7(1):2.

[29].Hong S, Kim K, Yoon S, Park W-Y, Sim S, Yu J-R. Detection of Cryptosporidium parvum in environmental soil and vegetables. Journal of Korean medical science. 2014;29(10):1367-71.

[30].Dado D, Izquierdo F, Vera O, Montoya A, Mateo M, Fenoy S, *et al.* Detection of zoonotic intestinal parasites in public parks of Spain. Potential epidemiological role of microsporidia. Zoonoses and public health. 2012;59(1):23-8.

[31].Yu J-R, Seo M. Infection status of pigs with Cryptosporidium parvum. The Korean journal of parasitology. 2004;42(1):45.

[32].Iqbal J, Hira P, Al-Ali F, Philip R. Cryptosporidiosis in Kuwaiti children: seasonality and endemicity. Clinical microbiology and infection. 2001;7(5):261-6.

[33].Iqbal J, Khalid N, Hira PR. Cryptosporidiosis in Kuwaiti children: association of clinical characteristics with Cryptosporidium species and subtypes. Journal of medical microbiology. 2011;60(5):647-52.

[34].Osman M, El Safadi D, Cian A, Benamrouz S, Nourrisson C, Poirier P, *et al.* Correction: Prevalence and Risk Factors for Intestinal Protozoan Infections with Cryptosporidium, Giardia, Blastocystis and Dientamoeba among Schoolchildren in Tripoli, Lebanon. PLoS neglected tropical diseases. 2016;10(4):e0004643.

[35].Yang Y, Zhou Y-B, Xiao P-L, Shi Y, Chen Y, Liang S, *et al.* Prevalence of and risk factors associated with Cryptosporidium infection in an underdeveloped rural community of southwest China. Infectious diseases of poverty. 2017;6(1):2.

[36].Abu Samra N, Jori, F., Cacciò, S.M., Frean, J., Poonsamy, B. & Thompson, P.N. Cryptosporidium genotypes in children and calves living at the wildlife or livestock interface of the Kruger National Park, South Africa. Onderstepoort Journal of Veterinary Research. 2016;83(1): 1024.

[37].Salyer SJ, Gillespie TR, Rwego IB, Chapman CA, Goldberg TL. Epidemiology and molecular relationships of Cryptosporidium spp. in people, primates, and livestock from Western Uganda. PLoS Neglected Tropical Diseases. 2012;6(4):e1597.

[38].Kiani H, Haghighi A, Seyyedtabaei SJ, Azargashsb E, Zebardast N, Taghipour N, *et al.* Prevalence, Clinical Manifestations and Genotyping of Cryptosporidium Spp. in Patients with Gastrointestinal Illnesses in Western Iran. Iranian journal of parasitology. 2017;12(2):169.

[39].Bera P, Das S, Saha R, Ramachandran V, Shah D. Cryptosporidium in children with diarrhea: a hospital-based Study. 2014.

[40].Sim S, Yu J-R, Lee Y-H, Lee J-S, Jeong H-G, Mohamed AAWS, *et al.* Prevalence of Cryptosporidium infection among inhabitants of 2 rural areas in White Nile State, Sudan. The Korean journal of parasitology. 2015;53(6):745.

[41].Ali MA, Khamesipour A, Valian HK, Rahi AA. Diarrhea caused by Cryptosporidium parvum in Kut, Iraq using different methods. Sch J App Med Sci. 2014;2(3D):1134-8.

[42].Parsons MB, Travis D, Lonsdorf EV, Lipende I, Roellig DMA, Kamenya S, *et al.* Epidemiology and molecular characterization of Cryptosporidium spp. in humans, wild primates, and domesticated

animals in the Greater Gombe Ecosystem, Tanzania. PLoS neglected tropical diseases. 2015;9(2):e0003529.

[43].Lal A, Fearnley E, Kirk M. The risk of reported cryptosporidiosis in children aged< 5 years in Australia is highest in very remote regions. International journal of environmental research and public health. 2015;12(9):11815-28.

[44].Sarkar R, Kattula D, Francis MR, Ajjampur SS, Prabakaran AD, Jayavelu N, *et al.* Risk factors for cryptosporidiosis among children in a semi urban slum in southern India: a nested case-control study. The American journal of tropical medicine and hygiene. 2014;91(6):1128-37.

[45].McKenna ML, McAtee S, Bryan PE, Jeun R, Ward T, Kraus J, *et al.* Human intestinal parasite burden and poor sanitation in rural alabama. The American journal of tropical medicine and hygiene. 2017;97(5):1623-8.

[46].Siwila J, Olsen A. Risk factors for infection with soil transmitted helminths, Cryptosporidium spp., and Giardia duodenalis in children enrolled in preschools in Kafue District, Zambia. Epidemiology Research International. 2015;2015.