Prevalence of Cryptosporidiosis Among Population of Wasit province

By

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انتشار داء الابواغ الخبيئه بين السكان في محافظة واسط شيماء شاكر حميد السلامي جامعة واسط/كلية الطب

ألخلاصه:

تم إجراء الفحص ألمجهري ل 600 عينه خروج بعد صبغها بصبغة المقاومة للحامضيه المحوره لأشخاص راجعوا مستشفئ الكرامة والزهراء في محا فظة واسط من شهر تشرين الثاني 2010 ولغاية شباط 2011 للتحري عن طفيلي داء الابواغ الخبيئه بلغت عداد العينات المصابة (ألموجبه) 79 (13.1%). كانت نسبة انتشار الطفيلي اعلى بين الاعمار (50-41) كما أشارت الدراسة الى عدم وجود فروق معنوية بين الذكور والإناث بينت الدراسه بان نسبة الاصابه كانت اعلى بين الاشخاص الذين يعشون في المناطق الريفيه عما في المدينة وذلك لسوء الخدمات الصحيه فيها كما سجل شهر شباط اعلى الاصابات وشهر كانون الاول ادناه.

Abstract

A microscopic examination of faeces samples after staining by modified acid fast stain of 600 person was carried out in Al - karama hospital and Al-Zahra hospital , Wasit province from November 2010 to feberuary 2011 overall prevalence of cryptosporidiosis was 79 (13.1%) Prevalence increased rapidly in age group 5 (41-50)year and there were no significant variation between males and females .Prevalence was higher among families who live in rural area than in urban area .the highest infection rats was record in Feberuary and the lowest was record in December

Introduction

Cryptosporidiosis, also known as crypto,^[1] is a parasitic disease caused by *Cryptosporidium*, a protozoan parasite in the phylum Apicomplexa. It affects the intestines of mammals and is typically an acute short-term infection. It is spread

through the fecal-oral route, often through contaminated water;(1) the main symptom is self-limiting diarrhea in people with intact immune systems. In immunocompromised individuals, such as AIDS patients, the symptoms are particularly severe and often fatal.(4) *Cryptosporidium* is the organism most commonly isolated in HIV positive patients presenting with diarrhea.(6) Treatment is symptomatic, with fluid rehydration, electrolyte correction and management of any pain. Despite not being identified until 1976, it is one of the most common waterborne diseases and is found worldwide. The parasite is transmitted by environmentally hardy microbial cysts (oocysts) that, once ingested, exist in the small intestine and result in an infection of intestinal epithelial tissue. (2)

History

The organism was first described of Cryptosporidium in 1907 by Tyzzer, who recognised it was a <u>coccidian</u>.

General characteristics

Cryptosporidium is a protozoan pathogen of the Phylum Apicomplexa and causes a diarrheal illness called cryptosporidiosis. Other apicomplexan pathogens include the malaria parasite Plasmodium, and Toxoplasma, the causative agent of toxoplasmosis. Unlike Plasmodium, which transmits via a mosquito vector(12), Cryptosporidium does not require an insect vector and is capable of completing its life cycle within a single host, resulting in microbial cyst stages which are excreted in feces and are capable of transmission to a new host. However, studies show that synanthropic filth flies may be involved in the transmission of human and animal cryptosporidiosis. (10)

The pattern of *Cryptosporidium* life cycle fits well that of other intestinal homogeneous coccidian genera of the suborder *Eimeriina*: macro- and microgamonts develop independently; a microgamont gives rise to numerous male gametes; and oocysts serving for parasites' spreading in the environment.(11)

Electron microscopic studies made from the 1970s have shown the intracellular, although extracytoplasmic localization of *Cryptosporidium* species.

These species possess a number of unusual features:

- an endogenous phase of development in microvilli of epithelial surfaces.
- two morphofunctional types of oocysts.
- the smallest number of sporozoites per oocyst.
- a multi-membraneous "feeder" organelle.

DNA studies suggest a relationship with the gregarines rather than the coccidia.(14) The taxonomic position of this group has not yet been finally agreed upon.

The genome of *Cryptosporidium parvum* was sequenced in 2004 and was found to be unusual amongst Eukaryotes in that the mitochondria seem not to contain DNA.(9) A closely-related species, *C. hominis*, also has its genome sequence available.(5) CryptoDB.org is a NIH-funded database that provides access to the *Cryptosporidium* genomics data sets.

A number of *Cryptosporidium* infect mammals. In humans, the main causes of disease are *C. parvum* and *C. hominis* (previously *C. parvum* genotype 1). *C. canis*, *C. felis*, *C. meleagridis*, and *C. muris* can also cause disease in humans.(8)

Cryptosporidiosis is typically an acute short-term infection but can become severe and non-resolving in children and immunocompromised individuals. In humans, it remains in the lower intestine and may remain for up to five weeks. The parasite is transmitted by environmentally hardy microbial cysts (oocysts) that, once ingested, exist in the small intestine and result in an infection of intestinal epithelial tissue.(7)

Material & method

From November 2010 to February 2011, seventy nine positive case of cryptosporidiosis attending the parasitological unit of Al-Karama hospital of wasit province were investigated in this study .the were of both sexes and the ages ranged between 1day to 50 years .The diagnosis of these patients was established on the basis of through clinical examination .In all these case stool samples were found positive for cryptosporidiosis .normal healthy of similar age and sex were also include as controls For microscopy each stool sample should be examination by :

Modified Acid-Fast Staining Procedure(13)

This technique is useful for the identification of oocysts of the coccidian species (*Cryptosporidium*, *Cystoisospora*, and *Cyclospora*), which may be difficult to detect with routine stains such as trichrome. Unlike the Ziehl-Neelsen Modified Acid-Fast Stain, this stain does not require the heating of reagents for staining.

Specimen:

Concentrated sediment of fresh or formalin-preserved stool may be used.

Reagents:

There are four steps to this procedure requiring the following solutions:

- 1. Absolute Methanol
- 2. Acid Alcohol: 10 ml Sulfuric Acid + 90 ml Absolute ethanol. Store at room temperature.
- 3. Kinyoun's Carbol fuchsin: may be purchased commercially.
- 4. 3% Malachite green: dissolve 3 g of malachite green in 100 ml of distilled water. Store at room temperature.

Procedure:

- 1. Prepare a smear with 1 to 2 drops of specimen on the slide and dry on a slide warmer at 60°C until dry. Do not make the smears too thick!
- 2. Fix with absolute methanol for 30 seconds.
- 3. Stain with Kinyoun's carbol fuchsin for one minute. Rinse briefly with distilled water and drain.
- 4. Destain with acid alcohol for 2 minutes. Rinse with distilled water and drain.
- 5. Counterstain with Malachite green for 2 minutes. Rinse briefly with distilled water and drain.
- 6. Dry on a slide warmer at 60°C for about 5 minutes. Mount with a coverslip using desired mounting media.
- 7. Examine 200 to 300 fields using $40\times$ or higher objectives. To confirm internal morphology, use $100\times$ oil immersion objective.(13)

Result & Discussion

A study on the prevalence of *Cryptosporidium parvium* in wasit province ,Iraq was made ,from November 2010 to Feberuary 2011.the patients were divided into five group (1,2,3,4,5) according to age (1day-10),(11-20),(21-30),(31-40) and (41-50) years respectively .

Table(1) show the distribution of positive cases in according to the age and gender group .These result showed the highest infection were 4%in group 5(41-50) years and lowest 0.8 % in group 3 (21-30) .the infection with cryptosporidiosis depended to immunity of host &accoure in children & adult suffer from diabetic and other disease causes immune depression (12)

Table (1) Prevalence of cryptosporidiosis in according to Age and Gender Groups

Age /Years	Male +Ve		Female +Ve			Total
	9/	6	%			%
Group 1(day-10)	10	1.8	8	1.3	18	3
Group 2 (10-20)	3	0.5	5	0.8	8	1.3
Group 3(21-30)	3	0.5	2	0.3	5	0.8
Group4 (31-40)	7	1.6	7	1.1	14	2.3
Group 5 (41-50)	16	2.6	18	3	24	4
Total	39	6.5	40	6.6	79	13.1

Table (2) represent the monthly distribution of infection .These result showed the highest infection in February (4.1%) and the lowest in December (2.5%)

Months	Case +VE	%	Case –Ve %			%
					Total	
November	20	3.3	160	26.6	180	30
December	15	2.5	135	22.5	150	25
January	19	3.1	141	23.5	160	26.6
Feberuary	25	4.1	85	14.2	110	18.3
Total	79	13.1	521	86.8	600	100

Table (3) shows the distribution of positive case according to the age and district groups. These result showed the higher infection in rural area 44(7.3%) than urban 35 (5.3 %), in group 5 (4%) more than other Probably , it can be ascribed: reduce parental personal ,eating habit and activities linked with soil contaminated with infected fecal matters. Previous studies had attributed the high endemidicity to poor environmental and personal hygiene, shortage of good water supply ,and toilet habits (3)

Table (3) Distribution of positive cases According to age &district Groups

Age /Years			Urban			Rural		Total
		Ve	%		+Ve	%		%
Group 1(day-10)			1.1	11		1.8	18	3
			7 7					
Group 2 (10-20)	4		0.6	4		0.6	8	1.3
Group 3(21-30)	2		0.3	3		0.5	5	0.8
Group4 (31-40)	6		1	8		1.3	14	2.3
Group 5 (41-50)	16		2.6	18		3	24	4
Total	35		5.3	44		7.3		79
								13.1

Conclusion

All cryptosporidiosis infections are caused by either ingestion or inhalation of the oocysts, therefore the main method of prevention should be to avoid or limit contact with the organism.

The best ways to prevent getting sick with cryptosporidiosis are to practice good hygiene, avoid water and food that might be contaminated and avoid fecal exposure during sex. There are also many prevention methods that focus on the • Maintain strict standards for water purification and filtration by using filters with a pore size of one to two micrometers.

(This will block the four to five micrometer *C. parvum* spores from entering the water supply).

• Boil water that is intended for consumption for at least one minute. (Heating to 72.4°C (162.3°F) or higher for one minute makes *C. parvum* oocysts non-infectious).

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