

INFECTION OF *CYCLOSPORA CAYETANENSIS* IN CHILDREN UNDER 15 YEARS OF AGE IN KATHMANDU VALLEY

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Abstract: *Cyclospora cayetanensis* is a rapidly emerging pathogen in developing countries like Nepal. It causes persistent diarrhea in children and in immunocompromised individuals. The study was carried out in children less than 15 years of age in Kathmandu Valley. A total of 302 stool samples were collected from different Sukumbasi (Slum children) areas of Kathmandu Valley. The samples were observed microscopically using normal saline and potassium dichromate preparation and the test was further confirmed by Sheather's sucrose flotation and Kinoyoun's modified acid fast staining technique. Out of the total sample processed *C. cayetanensis* oocysts were found in 24 (7.94 %) cases. The oocyst were distributed more in children of age group 3-6 years which constituted 5.49% of the total cases ($\chi^2=69.15$ p: 0.001) and distributed more in months of June (8) and July (8) ($\chi^2=9.042$ p: 0.250) whereas abdominal pain was the most common symptoms in infected children. These findings indicate that *C. cayetanensis* is rapidly emerging enteropathogen in children. So, active surveillance should be continued among diarrhea patients to look for different *C. cayetanensis* infection and to define the shifting patterns in the seasonal variations of this parasite.

Key words: *Cyclospora cayetanensis*; Diarrhea; Children; Faecal specimen.

INTRODUCTION:

C. cayetanensis is a coccidian parasite that causes prolonged diarrhea in human worldwide (Ortega, 1993). First reports of *C. cayetanensis* were made in Papua New Guinea (Ashford, 1973). More recent reports are from North, Central and South America, The Indian Subcontinent, South East Asia and Australia (Sterling *et al.*, 1997). The high endemic areas are Peru (Ortega, 1997), Haiti (Pape *et al.*, 1994) and Nepal (Sherchand and Cross 2002) Children acquire the infection by ingesting the sporulated oocyst. Water has been considered an important agent in outbreaks in Nepal (Rabold *et al.*, 1994; Sherchand and Cross, 1997). Leafy vegetables have also been reported as major source of infection (Ortega *et al.*, 1997) and Nepal (Sherchand *et al.*, 1997). The oocyst of *Cyclospora* measures about 8 μ m to 10 μ m in diameter. The life cycle completes in single host. The life cycle starts with the ingestion of sporulated oocyst which consists of 2 sporocysts and each enclosing 2 sporozoites. Inside the gut the sporozoites penetrate the epithelial cells and hence progresses to schizogony and gametogony. Sporogony and sporulation occur exogenously (DOB 2000) Individuals

infected with *Cyclospora* may experience prolonged watery diarrhea, abdominal cramping, Weight loss, anorexia, myalgia and occasionally vomiting. The incubation period may vary from 7-14 days on the basis of numbers of sporulated oocyst ingested. The parasite inflames the small intestine causing vilous atropy and crypt hyperplasia. Succesful treatment has been achieved by cotrimoxazole (160mg trimethoprim, 800 mg sulphamethaoxazole). A number of studies have been carried out in Nepal among expatriates and Tourists (Shim *et al.*,) but only few studies have been carried out in the local people. So this study was carried out to find out the prevalence of *C. cayetanensis* in children of Kathmandu Valley. The main aim of this study was to establish *C. cayetanensis* as a rapidly emerging pathogen in children. Since the biology and transmission issue of this parasite is not solved completely, further investigations on this parasite must be raised.

MATERIALS AND METHODS:

The study was conducted between April to November 2009 in Tribhuvan University Teaching Hospital, Public Health Research Laboratory, Maharajgunj and Dirgh Jeevan Health

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Care and Research Centre, Tripureswar, Kathmandu Nepal. A total of 302 samples were collected from different slum children of Kathmandu valley. Of them including Maharajgunj slum (113), Thapathali slum (66), Tripureswar slum (51), Koteshor slum (25) and Balkhu Slum (11). Data on predisposing factors were collected along with written consent from the respective children's parents. Stool sample was collected in clean wide mouth container. The 2.5 % potassium dichromate solution was used as a transportation as well as preservation of the specimen. The collected samples were observed in normal saline by direct microscopy. Sheather's sucrose flotation concentration technique was used further for detection of oocyst and then confirmed by Kinoyun's modified acid fast staining technique.

RESULTS:

Out of the total 302 stool specimen 24 (7.94 %) were found to

be positive for *Cyclospora cayetanensis* infection. Of the 24 positive cases 18 (75%) were male and 6 (25%) were female. Age wise prevalence of *Cyclospora cayetanensis* showed that the infection was more prevalent in children within age group 3-6 years of age. Regarding Monthwise infection (Table 2) significant infection was found in month of July (19.04 %) and in month of June (16.66 %). No infection by *Cyclospora cayetanensis* was detected in the month of September, October and November. As regards to sitewise prevalence of Cyclospora infection, there was no significant presence of *Cyclospora cayetanensis* on sitewise basis.

Co infection of Cyclospora with other intestinal parasites: Out of the 24 positive cases, 10 (41.66%) showed co infection with *Giardia lamblia*. 4 (16.66%) showed co infection with *Entamoeba histolytica* 4 (16.66%) showed co infection with *Blastocystis hominis* and 6 were detected without any co infection.

Table 1: Gender wise distribution of *C. cayetanensis*

Age group in years	Male	Female	Total parasite positive cases
0-3	98	65	4
3-6	45	25	11
6-9	18	15	5
9-12	14	6	2
12-15	9	7	2
Total	178	124	24

Age wise $\chi^2=69.15$ p: 0.001 (Significant)

Gender wise $\chi^2=1.144$ p: 0.285 (Not Significant)

Table 2: Month wise distribution of *C. cayetanensis*

Months	No of processed sample	Positive		Total
		Male	Female	
April	21	0	0	6(11.53%)
May	52	4	2	8(16.66%)
June	48	6	2	8(19.04%)
July	42	6	2	2(3.92%)
August	51	2	0	0
September	31	0	0	0
October	30	0	0	0
November	27	0	0	0
Total	302	18	6	24

Month wise $\chi^2=16.042$ p: 0.0250 (Significant)

Table 3: Site wise distribution of *C. cayetanensis* in different Sukumbasi areas.

Site of Collection	Total processed sample	Male	Female	Total
Maharajgunj	113	6	2	8
Thapathali	66	4	2	6
Tripureshor	51	4	2	6
Koteshor	25	4	0	4
Balkhu	11	0	0	0
Total	302	18	6	24

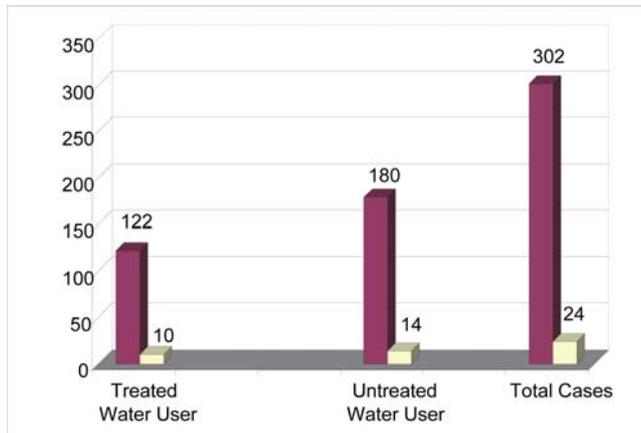
Site wise $\chi^2=3.561$ p: 0.469 (Not Significant)

Table 4: Frequency distribution of *C. cayetanensis* based on education of parent

Educational level	Total processed sample	Cyclospora Positive	Percentage
Illiterate	146	14	58.33%
Under S.L.C	109	8	33.33%
S.L.C and above	47	2	8.33%

Based on education of parent $\chi^2=0.17$ p: 0.68 (Not significant)

Figure 1: Frequency distribution of *C. cayetanensis* based on treatment of Water



Based on treatment of water $\chi^2=0.082$ $p=0.774$ (Not significant)

DISCUSSION:

Cyclospora cayetanensis is a rapidly emerging pathogen in developing country like Nepal. The study showed that the infection was higher in children less than 10 years of age. Similar results were obtained by Caryn et al., and Sherchand and Cross (2001). Sexwise prevalence of *C. cayetanensis* showed that there was no significant association between age and presence of infection. Similar report is shown by Amim et al., (1998). In the present study the infection was found to occur in children of slums. Though the sample size was small but the presence of parasite was significant. This may be attributed to low level of sanitation and maintenance of hygiene in children of slums. The present study also showed marked seasonal variation of *Cyclospora* infection with a high incidence of infection during warm rainy season i.e at July. This agrees with the findings by Hoge et al., (1993). But this finding does not agree with earlier findings by Ortega et al., (1997) in which peak infection occurred in warm dry seasons. *Cyclospora cayetanensis* was found in coinfection with other enteroparasites. Out of the 24 positive cases, 10 (41.66%) showed co infection with *Giardia lamblia*. 4 (16.66%) showed co infection with *Entamoeba histolytica* 4 (16.66%) showed co infection with *Blastocystis hominis* and 6 were detected without any co infection. Amin found the pathogens like *E. histolytica* and *G. lamblia* reduces the immunity which may result in new infection. Though actual biology of transmission of *C. cayetanensis* is not known. But it has been reported to be transmitted through water, soil (Ortega et al., 1997). Though oocysts of *Cyclospora cayetanensis* have been found in chicken, dogs and rats the possibility of human infection by zoonosis is not clear (Sherchand and Cross 2002; Garcia et al., 1996; Yai et al., 1997; Eberhard et al., 1999).

CONCLUSION:

This study concludes that *Cyclospora cayetanensis* is a rapidly emerging pathogen as a causative agent of diarrhea in children. Children under 10 years of age were found more susceptible for the infection. The parasite found more active

in dry rainy season. Contamination of water sources and usage of such water source leads for further making of suitable environment for this parasite. Further studies including all age groups might help to understand the actual transmission, pathogenicity and prevention of *Cyclospora* infection.

In addition, we need to develop quick, inexpensive, reliable molecular and diagnostic tools and to identify alternative efficacious agents for therapy as well as prophylaxis.

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