A CLINICO-EPIDEMIOLOGICAL STUDY OF GIARDIASIS IN CHILDREN IN RURAL PUNJAB, PAKISTAN

Khan I.A[1], Khan M.A[2], Raza F[3], Khan S.E[4]

Abstract:

Background:

Giardiasis is defined as symptomatic or asymptomatic infection by the ubiquitous enteric protozoan, Giardia lamblia that occurs from tropics to Arctic Greenland with global incidence of up to 30%. Although this cosmopolitan organism affects sexes and all ages, majority of cases occur in infants and children who are more vulnerable to its ill effects.

Objective:

This study was conducted, to investigate the prevalence of Giardia lamblia.

Methods:

This study was conducted on a randomly selected population of 232 children of both sexes

Result:

Giardia lamblia was the most commonly encountered parasite with a prevalence of 24.2%. All cases were subjected to a thorough clinical examination to differentiate symptomatic from asymptomatic, the figures being 88.5% and 11.5% respectively. In the symptomatic group, 21.7% were having atypical manifestations. Maximum cases were in the 5-9 years age group and males were more affected than females. Poor socio-economic status, overcrowding, inadequate sanitary practices and environmental contamination were among possible risk factors involved in etiopathogenesis. Parasitological and clinical cure were obtained in 96.1% and 91.3% of the cases respectively, with secnidazole. The cases were followed up for three months to look for relapse, which occurred in 16.6% of cases.

Keywords

Giardiasis, Giardia lamblia, Children, Punjab, Pakistan

INTRODUCTION:

Giardiasis is defined as symptomatic or asymptomatic infection by ubiquitous enteric pathogen Giardia lamblia that occurs from tropics to Arctic Greenland with global incidence of up to 30%. Although this cosmopolitan organism is included in "WHO's Neglected Disease Initiative" [1] it affects both sexes and all ages, majority of cases occur in infants and children who are more vulnerable to its ill effects. This fascinating parasite is one of the first to infect infants and young children in both developed and developing countries [2]. Infants may catch infection as early as three months [3], although neonates on their 4th postpartum day have been found to harbour this highly communicable protozoan [4]. Infection rates for infancy and childhood are variable throughout the world and may exceed 95% in areas like Peru where children are infected at least once by the time they reach their second birth day [5]. Whereas in the limited studies, conducted on its prevalence in Pakistan, this notorious parasite has been found to be the most commonly encountered one, with prevalence of up to 30.96% [6], no serious effort has been made to quantify its national or regional burden. Strangely enough, it has not even been enlisted in the intestinal parasites causing significant morbidity in Pakistan [7]. The present study is an attempt to highlight the magnitude of the problem.

1. ***Iqbal Akhtar Khan, Professor and Head,*** *Department of Community Medicine, Department of Medical Education & Research center, University of Lahore.*
2. ***Murad Ahmad Khan,*** *Department of Pharmacology, RAK Medical and Health Sciences University, Ras al Khaymah United Arab Emirate*
3. ***Fauzia Raza,*** *Department of Medicine, FMH College of Medicine & Dentistry, Lahore, Pakistan*
4. ***Sabiha Erum Khan,*** *Department of Pharmacology, FMH College of Medicine & Dentistry, Lahore, Pakistan*

.*=corresponding author:

E-mail: profiakhan@gmail.com

OBJECTIVES:

This cross-sectional, period prevalence, non-outbreak related, laboratory confirmed epidemiological and interventional study was conducted to:

i. Determine the prevalence of Giardia lamblia in the study area

ii. identify clinico-epidemiological characteristics of the cases

iii. Evaluate the effects of therapeutic intervention

MATERIALS AND METHODS:

This study was conducted on children of 0-14 years, in a rural area of Punjab (Pakistan) with population of approximately 6000 individuals. The sample size determination was done by WHO's formula [8]:

$$n = \frac{pq}{(E/1.96)^2}$$

Whereas:

n = maximum sample size required
p = maximum expected prevalence
q = (100 - p)
E = margin of sampling error tolerated

Substituting 'p', 'q', and 'E' by 20, 80 and 5 respectively the 'n' was calculated to be 246. From the sampling frame of all the available children, at the time of recruitment, 246 children were randomly picked up. The inclusion criterion was age 0-14 years of either sex. The exclusion criterion was apparently acutely ill or on medication for some chronic ailment.

Informed consent was obtained from the parents/ guardians of every recruit, after explaining to them the objectives of the study, procedures involved and anticipated benefits for the potential participants. Incentives were free check up, free medications, free follow up and free health education sessions.

Stool microscopy being the easiest, safest, cheapest and traditional method was chosen as study instrument (diagnostic tool) for parasite detection.

A single fresh stool specimen was collected from each individual in a clean-capped container (without preservative) and examined immediately in a mobile clinic set up, made temporarily in the study area. In children who were less productive, digital rectal technique [9] was used to obtain an immediate specimen on a gloved finger. A specimen was considered positive if either trophozoites or cysts or both were found in that. A case was defined as an individual with a positive stool specimen irrespective of nature and extent of symptoms, attributable to infection by Giardia lamblia. A blood film was prepared for each and every case to ascertain level and type of anemia.

Parasitological cure was defined as non-detection of cysts or trophozoites of Giardia lamblia in three consecutive stool samples, collected and examined on alternate days. Clinical cure was defined as sustained and significant improvement in the symptomatology, attributable to sole infection with Giardia lamblia. Complete cure was defined as combination of the two. Follow up period (in whom complete cure was obtained) was defined as the period of three months in which bi-weekly clinical examination (to look for recurrence of the relevant symptoms); parasitological examination (to look for reappearance of Giardia in the stool) and hemoglobin estimation (in those with iron deficiency anemia) was done. All cases were subjected to detailed history taking and clinical examination. Information was obtained about the previous investigations (if any) and treatment (if any).

Ethical approval:

Institutional Ethics Committee of Muhammad Medical College Mirpurkhas

RESULTS:

Having excluded 14 children (non-responders), those included in the study were 232, the response rate being 94.3%. The sample profile has been tabulated as follows:

Table I- Sample Profile

Age	Male	Female	Total
0-4 years	33	32	65 (28%)
5-9 years	48	41	89 (38.4%)
10-14 years	38	40	78 (33.6%)
Total	119 (51.3%)	113 (48.7%)	232

Giardia lamblia was detected in the stools of 56 children, giving prevalence of 24.2%. Four of them were excluded from the study because of simultaneous infection with other parasites, leaving 52 having sole infection with Giardia lamblia. Their epidemiological and clinical features are tabulated (Table II)

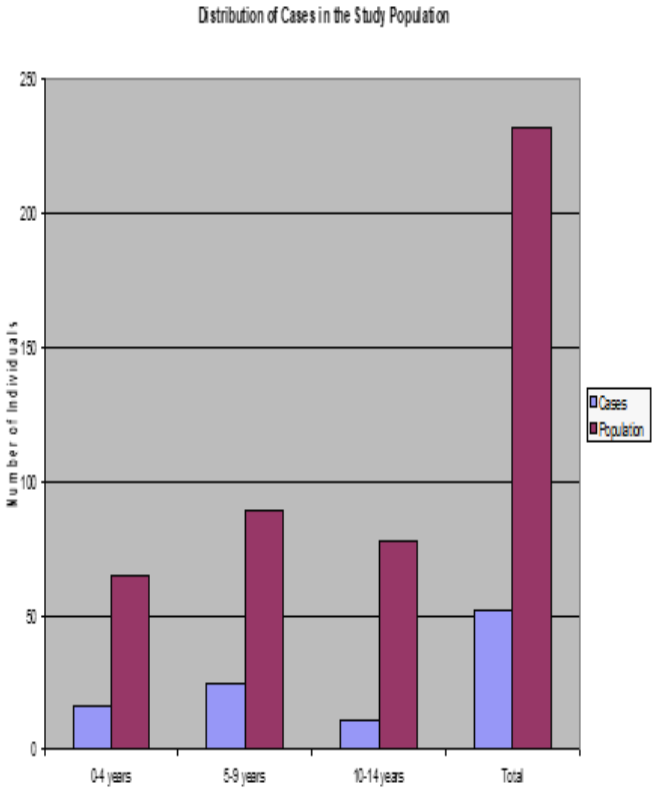
Table II – Sole Infection with Giardia lamblia

Age	Male	Female	Total
0-4 years	9	7	16 (30.7%)
5-9 years	14	11	25 (48.1%)
10-14 years	7	4	11 (21.2%)
Total	30 (57.7%)	22 (42.3%)	52

Three cases were <1 year (3 months, 4months, 4months). Maximum cases were in the 5-9 years age group. The distribution of cases in the study population is shown in Table III.

Table III – Distribution of Cases in the Study Population

Age	0-4 years	5-9 years	10-14 years	Total
Cases	16	25	11	52
Population	65	89	78	232



The symptomatology of the cases was rich, male female distribution being 27 (58.7%) and 19 (41.3%). The detailed features are shown in Table IV.

Table IV - Symptomatology of the Cases

Symptomatic 46 (88.5 %)		Asymptomatic 6 (11.5 %)
Usual manifestations 36 (78.3%) **Break up** * Diarrhoea 32 (88.9%) Anorexia 23 (63.9%) Flatulence 11 (30.5%) Nausea 9 (25%) Malaise 9 (25%) Abdominal pain 7 (19.5%) Epigastric gnawing 5 (13.9%) Poor weight gain 5 (13.9%) Constipation 3 (08.3%) **often the stools were bulky, semi liquid, pale, white, foul and mucoid.* Almost all the cases were having more than one symptom **Duration of symptomatology** <1 month 7(15.2%) 1-6 months 25(54.3%) 7-12 months 10(21.7%) >1 year 4(8.7%)	Atypical manifestations 10(21.7%) **Break up** Chronic urticaria 5 (50%) Pruritis without urticaria 3 (30%) Arthralgia 2 (20%) All the cases presented with single independent symptom	None of them, although parasitologically positive for Giardia lamblia, exhibited any relevant clinical feature.

The information obtained from the cases regarding previous investigations (if any) and previous treatment (if any) has been gathered in Table V.

Table V - Information about Previous Investigations and Treatment.

	Symptomatic 46	Asymptomatic 6
Investigations	Break up (n = 46) Blood tests (unspecified) 5 (10.9%) - X Ray examination (abdomen?) 3 (6.5%) - Ultrasonograply (abdomen?) 6 (13%) - Stool analysis 0 (zero %) - None 32 (69.6%)	None
Treatment	Break up (n = 46) - Faith healers 9 (19.6%) - Allopathic medicine 13 (28.3%) - Alternative medicine 15 (32.6%) - None 9 (19.6%)	None

Domiciliary visits of randomly selected 25 houses (convenient sampling) were made to ascertain level of general cleanliness; and knowledge, attitude and practice regarding hand hygiene, fecal hygiene and kitchen hygiene, through a pilot tested well-structured checklist. Inadequate sanitary practices and sub-optimal living conditions were observed. There was abundance of flies and cockroaches. In some houses goats, sheep or cattle were found sharing occupancy with the human. Stray dogs had free access to yards. Cats, which have special place in human hearts since centuries, were quite common and had freedom of movements to every corner of the occupancy.

Review of the official documents to obtain background information about sources of drinking water and toilet facilities, in the rural Punjab, was conducted.

According to MICS (Multiple Indicators Cluster Survey of Pakistan), the proportion of households with safe/potable and convenient source of drinking water (defined as piped supply in the dwelling or hand pump in the dwelling) is 78%. Further breakdown is as following [10].

- Piped supply on the dwelling---------- 8
- Hand pump on the dwelling ---------- 70
- Hand pump outside the house -------- 5
- Tube well or motorized borehole outside the house ----- ————————=========————————— 10
- Others ---------- 7

In the recent PDHS (Pakistan Demographic and Health Survey), the corresponding figure for piped supply is 33.3 % [11] but this includes rural and urban Punjab and is not comparable with MICS data. Regrettably, the MICS definition of safe and potable water is far from scientific one,

which emphasizes on freedom from harmful chemical substances and pathogenic organisms.

According to the same MICS, the proportion of households with safe and convenient toilet facilities (defined as flush latrines connected with water-borne sewage system or flush latrines connected with septic tank within the dwelling) is 27%. Further breakdown is as following [10].

- Flush latrines connected with water-borne sewage——11%
- Flush latrines connected with septic tank----------16%
- No protected facilities go out somewhere or in fields ——69%
- Others ——————————————————4%

In the above referred PDHS, the corresponding figures for flush latrines connected with water-borne sewage and those connected with septic tanks are 26.5% and 18.1% respectively[11] but those include rural and urban Punjab and are not comparable with MICS data.

Therapeutic Intervention:

Giardiasis was labeled "a self-limiting disease" in the past. However, with advancement in our understanding, there is serious concern about its potential harms if left untreated. There cannot be two opinions about treating a symptomatic infection. However, to treat or not to treat those with inapparent symptomatology is a debatable issue [12]. Occurrence of symptomatic disease in an extended family, in Kentucky, through an innocent appearing asymptomatic infant [13] and outbreak among several groups at an indoor swimming pool through a fecal accident by a handicapped child, harboring Giardia lamblia [14] are well documented. A food-borne outbreak amongst participants of a family function, consuming fruit salad, has been reported from New Jersey. The asymptomatic salad preparer had a diapered child and a pet rabbit (both found positive for Giardia lamblia) at home [15]. With this background information, to treat those with inapparent symptomatology, in the present study, the decision was based on following objectives:

- To prevent possible development of clinical disease in them
- To minimize risk of becoming potential source of infection, to others

From amongst the available therapeutic modalities secridazole was selected because of being more potent than metronidazole, very well tolerated, rapidly and completely absorbed after oral administration with longer terminal elimination half life of approximately 17-29 hours [16]. It has parasitological cure rate of 98% and clinical cure rate of 95% [17]. In a Venezuelan study, secridazole resulted in post-treatment reduction of levels of IgA antibody and IgG specific antibody by 76.5% and 52.9% respectively [18]. In the present study, secridazole suspension 30 mg/Kg body weight was administered to all cases, under supervision, as single oral dose with guaranteed compliance, in fact "One Minute Treatment"[19].

In three children (5.8%), who vomited after taking suspension, the dose was repeated. The others tolerated it well. The overall response was excellent. Parasitological cure was noted in 92.2% (50 out of 52) of the cases. Clinical cure was observed in 91.3% (42 out of 46) of the cases.

Relapse of symptoms occurred in 16.6% cases who were administered another course with satisfactory outcome. The cause of relapse was most possibly re-infection from household contacts, not included in the study.

Additional benefit of antigiardial therapy was noted in improvement of hemoglobin status, without iron supplementation. In the present study, 13 cases (28.3%) were found to have iron deficiency anemia (IDA). During follow up period, 9 of them (69.2%) improved their hemoglobin status, even though no iron preparation had been instituted to them.

DISCUSSION:

"Let us find out what parasites a man has and we will tell you under what standards of sanitation he lives." Anonymous Human parasitization with protozoan Giardia lamblia is a global problem, more so in developing countries because of substandard living conditions and unhygienic practices. Pakistan by no means is an exception where environmental contamination by unhealthy lifestyle, overcrowding, unsanitary sewage disposal, unsafe water and food provisions is mainstay in the continuity of endemnicity of this notorious pathogen. The observations made on domiciliary visits of the houses, in the study area, are not without interest since Giardiasis is known to have inverse correlation with the level of sanitary practices [9]. It has been documented that the viable cysts of Giardia may remain viable in fly's excreta for 24 hours after the insect has fed on contaminated faces[20,21]. Similarly, viable cysts have been found to survive in the intestine of cockroach for about 12 days after the insect has fed on infected material [21, 22]. It is tempting to postulate that these insects, serve as a mechanical vector of Giardia lamblia, in the study population. However, this issue warrants extensive investigation.

Most, if not all, cases of Giardiasis in infants and children are symptomatic. In endemic areas, majority of children become infected during early life and the first infection is almost always symptomatic [2]. Interestingly, in Israel, the first infection has been recorded to be asymptomatic [23]. In the present study, symptomatics account for 88.5% of the cases. These figures are comparable with those in United States [24], Germany [25], Chile [26] and Saudi Arabia [27] where the symptomatics were 81%, 74%, 75% and 72% respectively. This variation in symptomatology could be due to a variable host difference or a difference in the virulence of the parasite isolate or a combination of both [28].

Whereas, most of the cases exhibit clinical features related to digestive tract, a noteworthy percentage of cases have been documented in literature to present atypically. In the present study, 21.7% such cases were recorded, mainly with cutaneous manifestations. Since the first reported Giardia related urticaria [29], numerous cases of allergic manifestations including acute urticaria, chronic urticaria, pruritus without urticaria and erythema nodosum have been reported from various spots on the globe [30, 31, 32]. Complete resolution of symptoms in all the 8 cases in the present study, merely by antigiardial therapy, proved that the offending organism was Giardia lamblia and not any concomitant dermatological pathology.

The 2 cases presenting with arthralgia involving knee and ankle had no improvement with the on-going antiarthritic treatment, by the local healthcare providers. Complete reversal of clinical picture merely with antigiardial therapy, instituted by the authors, proved that the etiology was Giardia related. Identical cases have been reported in Mexican children [33].

Although, the duration of symptomatology, based on perception of the parents/guardian ranged from <1 month to >1year, only 30.4 % opted for any investigation and those too irrelevant. Detailed questioning revealed that they had strong belief that the X-Ray examination and more recently known ultrasonography could give exact "picture" of the "internal disease". Some had similar optimism about (unspecified) blood tests. Strangely enough, no case was subjected to stool analysis. This is not surprising when we refer to the comments:

"Obtaining of a stool specimen from A normal Pakistani is not a simple affair"[34]

Since there is heavy urban bias in the distribution of health facilities, selection of health care provider is associated with ability to pay [7]. Alternative medicine practitioners and faith healers, being easily accessible, are in front line. Those practicing allopathic medicine in such set ups are mostly quacks.

Malabsorption of iron is a well-documented complication of Giardiasis. Successful antigiardial therapy has resulted in normalization of hemoglobin level even without supplemental iron[35, 36, 37]. In the present study, in 69.2% of anemics, the hemoglobin status improved without supplemental iron, simply by successful antigiardial therapy.

While discussing the epidemiology of Giardia lamblia in rural Lesotho, Esrey et al remarked, "available treatments are effective but the vast majority of infected people in developing countries remain undiagnosed and never receive treatment. Efforts at prevention should therefore, prove more effective in reducing morbidity due to Giardia lamblia since treatment is unlikely"[38]. With the background knowledge, that freedom from the offending parasite needs improvement in awareness of the magnitude of the problem; and change in attitude and practices, community therapy was directed towards prevention. Maximum concentration was given on health education at individual, family and group levels by exposure to adequate scientific knowledge. How to render the drinking water safe at domestic level, how to avoid contamination of stored water and leftover food and how to improve hand, kitchen and fecal hygiene with shoe-string budget were demonstrated to the participants in different sessions. "**No Survey without Service**" was adopted as a slogan in the present study.

Limitations of the study

The prevalence of Giardia lamblia, in the present study, is most probably an underestimate because only a single stool specimen was examined and direct stool microscopy was used for parasite detection. It is well known that fecal excretion of cysts is highly variable varying from high to low with periods of negativity ranging from 2 to 30 days[39]. It has been estimated that 15-50% of all Giardia infections may go undetected if only one specimen is examined[40]. Despite this background information existing facilities at the set up and cost restraints precluded the use of alternative diagnostic tools.

Family clustering of cases due to identical environmental conditions and lifestyle is well documented[41]. It is unfortunate, that, due to paucity of facilities, person to person transmission by inter-familial and intra-familial interactions could not be ratified.

Giardiasis in companion animals is common worldwide. Dogs and cats have been shown to harbor Giardia which may be transmitted to man by close contact [42]. Similarly, domestic ruminants have been implicated in transmission of Giardia[43]. The zoonotic potential in the endemnicity of Giardial infection, in the study area, needs to be evaluated.

Cysts of Giardia lamblia were recovered from all the different vegetables used for raw consumption, in Costa Rica[44]. In the present study, consumption of raw vegetables is a common practice. The potential source of infection in such cases needs to be investigated.

It was observed that, in study area, the farmers used untreated wastewater in agriculture. A comparative study conducted in Faisalabad (Punjab), on the use of untreated wastewater in agriculture, revealed significant increased risk of Giardial infection in wastewater farming households when compared with farming households using non-wastewater irrigation[45]. Such an investigation should be done, in the study area, to determine the potential role of wastewater in transmission of Giardia lamblia.

Authors' contribution

IAK designed the study protocol and primers. MAK, FR and SEK were responsible for patient recruitment, management and follow up. All authors participated in the analysis and interpretation of results. IAK and MAK drafted the manuscript. All authors read and approved the final version. IAK is guarantor of the paper.

Acknowledgements

We are grateful to Prof. Razi Muhammad, Prof. Zafar Abbas and Dr. Muhammad Ahmad Khan who provided technical support throughout and monitored the ongoing progress of the project. The parents/guardians of the participants owe appreciation for their extended cooperation. Rafiuddin Siddiqui and Hassam-ud-din deserve special thanks for their bibliographical assistance.

Funding

The authors personally contributed for the project.

Conflicts of interest statement

The authors have no conflicts of interest concerning the work reported in this paper.

REFERENCES:

1. Sovioli L, Smith H, Thompson A- Giardia and Cryptoridium join the 'Neglected Disease Initiative' - Trends Parasitol 2006; 22(5): 203-208

2. Islam A, Stoll BJ, Ljungstrom I, Biswas J, Nazrul H, Huldt G – Giardia lamblia infections in a cohort of Bangladeshi mothers and infants followed for one year – J Pediat 1983; 103: 996-1000

3. Pugh RJ, Newton RW- Giardiasis in infancy and childhood – Practitioner 1980; 224: 393-397

4. Farthing MJ. Mata L. Urrutia JJ, Kronmal RA - Natural history of Giardia infection of infants and children in rural Guatemala and its impact on physical growth-Am J Clin Nut 1986; 43:395-405
5. Gilman RH, Marquis GS, Miranda E, Vestegui M, Martinez H – Rapid re- infection by Giardia lamblia after treatment in a hyperendemic Third World Community – Lancet 1988; 1(8581): 343-345
6. Younas M, Shah S, Talaat A – Frequency of Giardia lamblia infection in children with recurrent abdominal pain – J Pak Med Assoc 2008; 58(4):171-174
7. National Health Survey of Pakistan - Medical Research Council. Health Profile of the People of Pakistan-Child Health -1998; 110
8. Vaughan JP, Morrow RH – Manual of Epidemiology for District Health Management – World Health Organization 1989; 175-176
9. Jones JE - Giardiasis- Primary Care 1991; 18(1): 43-52
10. Ministry of Health, Government of Pakistan- Multiple Indicators Cluster Survey 1995; 18,20,21
11. National Institute of Population Studies Pakistan, Macro International Inc United States – Pakistan Demographic and Health Survey (2006-2007) 2008; 190-191
12. Khan IA-Giardiasis of infancy and childhood: To treat or not to treat?- Medical Channel 2001; 7(1): 1-4
13. Blondell RD, Dedman EB – An extended family with Giardiasis – J Fam Pract 1984; 19(3): 388-392
14. Porter JD, Ragazzoni HP, Buchanon JD, Waskin HA, Juranek DD, Parkin WE – Giardia transmission in a swimming pool – Am J Public Health 1988; 78(6): 659-662
15. Porter JD, Gaffney C, Heymann D, Parkin W – Foodborne outbreak of Giardia lamblia – Am J Public Health 1990; 80(10): 1259-1260
16. Gillis JC, Wiseman LR-Secnidazole: A review of its antimicrobial activity, pharmacokinetic properties and therapeutic use in the management of protozoal infections and bacterial vaginosis. Drugs 1996; 51(4): 621-638
17. Di Prisco MC, Jimenez JC, Rodriguez N, Costa V, Villamizar J, Silvera A, Carrillo M, Lira C, Zerpa E, Lopez Y - Clinical trial with Secnidazole in single dose in Venezuelan children infected by Giardia intestinalis - Invest Clin 2000; 41(3): 179-188
18. Jimenez JC, Pinon A, Dive D, Capron M, Dei-Cas E, Convit J- Antibody response in children infected with Giardia intestinalis before and after treatment with Secnidazole – Am J Trop Med Hyg 2009; 80(1): 11-15
19. Khan IA, Khan MA – Giardiasis co-existing with pregnancy: A distressing and threatening condition – Medical Channel 1999; 5(3-5): 16-19
20. Root FM – Experiments on the carriage of intestinal protozoa of man by flies – Am J Hyg 1921; 1:131
21. Wang CC – [Giardiasis-in "Human Parasitology"] edited by W.X.Zhao – People Health Press of Peoples Republic of China 1994; 52-58
22. Young MD -- Cockroaches as carriers of "Giardia" cysts – J Parasit -1937; 23: 102
23. Ish-Horowicz M, Korman SH, Shapiro M – Asymptomatic Giardiasis in children – Pediat Infect Dis J 1989; 8 (11): 773-779
24. Birkhead G, Vogt RL – Epidemologic surveillance for endemic Giardia lamblia infection in Vermont – Am J Epidemiol – 1989; 129(4): 762-768
25. Stolte M, Vogele Dirks H – Giardiasis: A simple diagnosis that is often delayed – Z. Gastroenterol – 1991; 29 (8):373-377
26. Goldin AJ, Apt W, Aguilera X, Zulantay I, Warhurst DC, Miles MA – Efficient Diagnosis of Giardiasis among nursery and primary school children in Santiago, Chile by the capture ELISA for the detection of fecal Giardia antigens – Am J Trop Med – 1990; 42(6): 538-545
27. Khan IA – Giardiasis: Public health significance and intervention programme – Medical Spectrum 1999; 20 (8,9): 3-7
28. Farthing MJG – Host parasite interactions in human Giardiasis – Quat J Med 1989; 70(3):191-204
29. Harris HR, Mitchell JH – Chronic urticaria due to Giardia lamblia – Arch Dermatal Sphilology 1949; 59: 587-589
30. Khan IA, Murtaza G – Pruritus without urticaria: An allergic manifestation of Giardiasis – J Baqai Med Univer 1998; 1(1): 22-26
31. Khan IA, Murtaza G – Role of Giardia lamblia infection in aetiopathogensis of chronic urticaria – Spectrum 1999; 20(1,2): 9-12
32. Khan IA – Allergic manifestations of common enteric protozoa-Surgery of 100 cases – J Pak Assoc Derm 2002; 12: 3-9
33. Meza-Ortiz F – Giardiasis associated arthralgia in children – Arch Med Res 2001; 32(3): 248-250
34. Ansari MAR, Sapru ZA – Intestinal parasite infections in children of a primary school of Lahore - A preliminary survey – Pak J Health 1964; 14: 87-105
35. Veghelyi PV, Lancos FJ- Avitaminosis A in Giardiasis – Am J Dis Child - 1949 ; 78: 257-259
36. De Vizia, B.Poggi V, Vajro P, Cucchiara S, Acampora A – Iron malabsorption in Giardiasis – J Paed 1985; 7: 75-78
37. Jimenez JC. Rodriguez N, Di Prisco MC, Lynch NR, Costa V – Haemoglobin concentrations and infection by Giardia intestinalis in children: effect of treatment with Secnidazole – Ann Trop Med Parasitol 1999; 93 (8): 823-827
38. Esrey SA, Collett J, Miliots MD, Koornhof HJ, Makhales P – The risk of infection from Giardia lamblia due to drinking water supply, use of water, and latrines among pre-school children in rural Lesotho – Int J Epidemiol 1989; 18(1): 248-253
39. Danciger M, Lopez M – Numbers of Giardia in the feces of infected children – Am J Trop Med Hyg 1975; 24 (2): 237-242
40. Goka AK, Rolston DD, Mathan VI, Farthing MJ -- The relative merits of faecal and duodenal juice microscopy in the diagnosis of giardiasis – Trans R Soc Trop

Med Hyg 1990; 84(1): 66-67
41. Galazka SS - Giardia lamblia: A clinical - epidemiological case report - J Fam Prac 1982; 15(6): 1165-1169
42. Meloni BP, Thampson RC, Hopkins RM, Reynoldson JA, Gracey M - The prevalence of Giardia and other intestinal parasites in children, dogs and cats from Aboriginal communities in the Kimberley - Med J Australia 1993; 158(3): 157-159
43. Buret A, den Hollander N, Wallis PM, Befus D, Olson ME – Zoonotic potential of Giardiasis in domestic ruminants - J Infect Dis 1990; 162(7): 231-237
44. Monge R. Chinchilla M. Reyes L - [Seasonality of parasites and intestinal bacteria in vegetables that are consumed raw in Costa Rica] - Rev Biol Trop 1996; 44(2 A): 369-375
45. Ensink JH, van der Hoek W, Amerasinghe FP - Giardia duodenalis infection and wastewater irrigation in Pakistan - Trans R Soc Trop Med Hyg 2006;100(6): 538-542