

SEASONAL PREVALENCE OF INTESTINAL PARASITES IN THE UNITED STATES DURING 2000

OMAR M. AMIN

Parasitology Center, Inc., Tempe, Arizona; Laboratorio Analisis Clinicos, Nogales, Sonora, Mexico

Abstract. One-third of 5,792 fecal specimens from 2,896 patients in 48 states and the District of Columbia tested positive for intestinal parasites during the year 2000. Multiple infections with 2–4 parasitic species constituted 10% of 916 infected cases. *Blastocystis hominis* infected 662 patients (23% or 72% of the 916 cases). Its prevalence appears to be increasing in recent years. Eighteen other species of intestinal parasites were identified. *Cryptosporidium parvum* and *Entamoeba histolytica*/E. dispar ranked second and third in prevalence, respectively. Prevalence of infection was lowest (22–27%) in winter, gradually increased during the spring, reached peaks of 36–43% between July and October, and gradually decreased to 32% in December. A new superior method of parasite detection using the Proto-fix™-CONSED™ system for fixing, transport, and processing of fecal specimens is described. In single infections, pathogenic protozoa caused asymptomatic subclinical infections in 0–31% of the cases and non-pathogenic protozoa unexpectedly caused symptoms in 73–100% of the cases. The relationship between Charcot-Leyden crystals and infection with four species of intestinal parasites is examined and the list of provoking parasitic causes is expanded.

INTRODUCTION

Parasitologic investigations of large patient populations are rarely conducted in the United States, where the illusion of freedom from parasitic infections still predominates. Such investigations are considerably more common in third-world countries where endemic parasitoses are more readily documented.¹ In an attempt to address this problem we reported the results of routine examination of fecal specimens for parasites from 644 patients in the United States during the summer of 1996.¹ Prevalence, patient age and sex, and intestinal and extra-intestinal symptoms, as well as variables related to foreign travel, infected household contacts, and previous parasitic infections were reported. An expanded version of the summer 1996 report is herein presented, in which complete seasonal data of 12 species of parasites from a considerably larger population is analyzed with emphasis on prevalence, symptomology, and Charcot-Leyden crystals. Few studies of large patient populations in the United States^{2,3} or more geographically limited populations, e.g., California⁴ or Ontario, Canada,⁵ have been reported.

MATERIALS AND METHODS

A total of 5,792 fecal specimens from 2,896 patients (two specimens per patient) were collected and transported to Parasitology Center, Inc. (Tempe, AZ) in Proto-fix™ in plastic vials provided in mailable kits by UROKEEP (Chandler, AZ). Specimens were collected throughout the United States between January and December 2000 following physician's orders. Tests were ordered either as part of routine medical examinations or when patients experience changes in bowel habits, energy level, or normalcy after a foreign trip, bad meals, or other exposures. Specimens were processed and stained in CONSED™ according to manufacturer's (Alpha-Tec Systems, Inc., Vancouver, WA) directions. This procedure was used in 10,358 specimens by 1998, and was described, fully evaluated, and compared with other methods.⁶ The number of specimens found positive (number of individuals and of species of parasites) was significantly higher than in other methods compared, e.g., formalin-ethyl acetate or trichrome stain.⁶ These observations were supported by findings of other observers.^{7,8} The Proto-fix™-CONSED™

system involves filtering of fixed specimens, mixing with CONSED™ and ethyl acetate, vortexing, centrifugation, decanting all but the fecal plug, and mixing with CONSED™ diluting reagent. The plug is then transferred to and mounted on a slide for examination.⁶ All microscopic evaluations and identification were made by the same observer(s) blinded to patient information, e.g., symptoms, travel, etc. Positive results were quantified (number of organisms per high-power field on a scale of 1 to 4) from duplicate samples from the same patient.

RESULTS

Prevalence. Nine hundred sixteen (32%) of 2,896 tested patients were infected with 18 species of intestinal parasites in the year 2000 (Table 1) in 48 states and the District of Columbia as follows: Alabama (2 infected of 3 tested, 67%), Alaska (6 of 14, 43%), Arizona (79 of 279, 28%), Arkansas (2 of 8, 25%), California (314 of 859, 36%), Colorado (17 of 88, 19%), Connecticut (4 of 24, 17%), Delaware (0 of 3, 0%), Florida (18 of 64, 28%), Georgia (28 of 72, 39%), Hawaii (5 of 9, 55%), Idaho (2 of 5, 40%), Illinois (30 of 92, 33%), Indiana (20 of 74, 27%), Iowa (16 of 44, 36%), Kansas (1 of 2, 50%), Kentucky (1 of 6, 17%), Louisiana (1 of 4, 25%), Maine (27 of 86, 31%), Maryland (15 of 64, 23%), Massachusetts (18 of 61, 29%), Michigan (4 of 22, 18%), Minnesota (10 of 28, 36%), Mississippi (1 of 2, 50%), Missouri (4 of 10, 40%), Montana (2 of 4, 50%), Nevada (7 of 28, 25%), New Hampshire (2 of 9, 22%), New Jersey (20 of 81, 25%), New Mexico (55 of 140, 39%), New York (75 of 230, 33%), North Carolina (3 of 16, 19%), Ohio (5 of 23, 22%), Oklahoma (1 of 2, 50%), Oregon (44 of 135, 33%), Pennsylvania (16 of 81, 20%), Rhode Island (2 of 9, 22%), South Dakota (0 of 2, 0%), Tennessee (1 of 3, 33%), Texas (21 of 90, 25%), Utah (2 of 7, 29%), Vermont (3 of 11, 27%), Virginia (5 of 20, 25%), Washington (12 of 36, 33%), Washington DC (3 of 8, 37%), West Virginia (2 of 4, 50%), Wisconsin (9 of 33, 27%), and Wyoming (1 of 5, 20%). *Blastocystis hominis* was the most frequently detected parasite in single and multiple infections, with *Cryptosporidium parvum* and *Entamoeba histolytica*/E. dispar ranking second and third, respectively. All parasites and their prevalences are listed in Table 1.

Symptoms. The term symptom in this study is defined as

TABLE 1
Seasonal prevalence of intestinal parasites in stool specimens singly or multiply infecting 916 of 2,896 patients in the United States examined in 2000

Month	Number of specimens examined	Number (%) of specimens infected			Number (%) of parasite species identified in single and multiple infections*															
		Single infections	Multiple infections	Total	AL	BH	CM	CC	CP	DF	EN	EC	EH/ED	EH	GL	IB				
January	155	38 (24)	4 (3)	42 (27)	—	27 (67)	1 (2)	1 (2)	4 (9)	—	6 (14)	1 (2)	1 (2)	—	2 (5)	1 (2)				
February	283	58 (20)	4 (1)	62 (22)	1 (2)	43 (69)	—	2 (2)	10 (16)	2 (3)	5 (8)	—	4 (6)	—	—	1 (2)				
March	254	69 (27)	10 (4)	79 (31)	1 (1)	53 (67)	—	2 (2)	18 (23)	2 (2)	3 (4)	7 (9)	5 (6)	1 (1)	2 (2)	1 (2)				
April	228	60 (26)	5 (2)	65 (28)	—	41 (63)	1 (1)	1 (1)	10 (15)	—	2 (3)	5 (8)	7 (11)	—	3 (5)	1 (1)				
May	326	71 (22)	10 (3)	81 (25)	—	43 (53)	1 (1)	3 (4)	18 (22)	3 (4)	4 (5)	7 (9)	6 (7)	1 (1)	5 (6)	2 (2)				
June	268	65 (24)	6 (2)	71 (26)	2 (3)	51 (72)	—	—	9 (13)	—	5 (7)	4 (6)	5 (7)	2 (3)	—	2 (3)				
July	198	58 (29)	6 (3)	64 (32)	—	50 (78)	—	1 (2)	8 (12)	—	4 (6)	2 (3)	7 (11)	1 (2)	1 (2)	2 (2)				
August	304	94 (31)	17 (5)	111 (36)	2 (2)	88 (79)	1 (1)	—	11 (10)	—	4 (4)	9 (8)	13 (12)	—	1 (1)	2 (2)				
September	227	82 (36)	13 (6)	95 (42)	3 (3)	81 (85)	—	3 (3)	9 (9)	—	5 (5)	5 (5)	5 (5)	—	—	—				
October	295	118 (40)	9 (3)	127 (43)	—	91 (72)	—	1 (1)	17 (13)	1 (1)	4 (3)	6 (5)	7 (5)	1 (1)	5 (4)	—				
November	181	56 (31)	6 (3)	62 (34)	2 (3)	51 (82)	—	1 (2)	5 (8)	1 (2)	—	2 (3)	3 (5)	1 (2)	—	3 (5)				
December	177	50 (28)	7 (4)	57 (32)	3 (5)	43 (75)	1 (2)	—	2 (3)	1 (2)	4 (7)	2 (3)	5 (9)	—	—	—				
Total	2,896	826 (29)	90 (3)	916 (32)	14 (2)	662 (72)	5 (1)	14 (2)	121 (13)	12 (1)	46 (5)	50 (5)	68 (7)	7 (1)	19 (2)	13 (1)				

*The total number of specimens infected may be more than the total in the preceding column because of multiple infections. AL = *Ascaris lumbricoides*; BH = *Blasitocystis hominis*; CM = *Cryptosporidium parvum*; DF = *Dientamoeba fragilis*; EN = *Endolimax nana*; EC = *Entamoeba coli*; EH/ED = *Entamoeba histolytica*/*Entamoeba dispar*; EH = *Entamoeba hartmanni*; GL = *Giardia lamblia*; IB = *Iodamoeba butschlii*; Other parasites not listed include *Ancylostoma*/*Necator* (in 1 specimen); *Diphyllobothrium latum* (1); *Eimeria vermicularis* (1); *Euteromona hominis* (3); *Retortomonas intestinalis* (2); *Taenia* sp. (1); and *Trichuris trichiura* (2).

any change in normal body function induced by direct or indirect action of parasites. Direct action includes invasiveness and tissue damage due to parasite feeding or migration. Indirect action results from parasite metabolic byproducts and toxic secretions. Symptoms are in two categories: 1) gastrointestinal, including (in order of observed frequency) flatulence, diarrhea, bloating, abdominal cramping, constipation, malabsorption/maldigestion, bloody or odorous stool, irritable bowel, mucus, and leaky gut, and 2) extra-intestinal (systemic), including (in order of observed frequency) fatigue, nervous/sensory disorders, pain, skin disorders, allergies, nausea, muscle weakness/pain, immune deficiencies, headache, fever/night sweats, insomnia, and weight changes. Most infected patients with parasitic symptoms experienced 1–4 gastrointestinal and/or extra-intestinal symptoms; they are simply called symptoms.

Of the 826 patients with single infections, 584 (70%) experienced overt symptoms and 242 (30%) had none (Table 2). Infections with pathogenic protozoa included 0–31% asymptomatic infections. Protozoans regarded as non-pathogenic were associated with symptoms in 69–100% of the cases. *Ascaris lumbricoides* produced no symptoms in one of 12 cases.

Multiple infections. Ninety patients (10% of cases) were concurrently infected with 2–4 species of parasites. Among these, 21 patients experienced no symptoms. These 21 cases involved 19 infections with *B. hominis*, 7 with *C. parvum*, 6 with *Endolimax nana*, 5 with *E. histolytica*/*E. dispar*, 4 with *Giardia lamblia*, 3 with *Entamoeba coli*, 2 with *Chilomastix mesnili*, *Cyclospora cayetensis*, and *Iodamoeba butschlii*, and 1 with *Retortomonas intestinalis*. The remaining 69 patients with concurrent infections were symptomatic. These 69 cases involved 58 infections with *B. hominis*, 22 with *C. parvum*, 19 with *E. coli*, 18 with *E. histolytica*/*E. dispar*, 16 with *E. nana*, 5 with *C. cayetensis* and *G. lamblia*, 2 with *Ascaris lumbricoides*, and 1 with *C. mesnili*, *Dientamoeba fragilis*, *Entamoeba hartmanni*, *I. butschlii*, *R. intestinalis*, and *Taenia* sp.

Seasonality. Monthly seasonal prevalence of single and multiple infections gradually increased from a minimum in February to a maximum between August and October, then decreased in December. Prevalence of infection with *Blasitocystis hominis* was lowest in May and highest in September

TABLE 2

Relationship between symptoms and species of intestinal parasites singly infecting 826 patients in the United States in 2000

Species of parasite	Number (%) of singly infected patients		
	Total	With symptoms	Without symptoms
Pathogenic protozoa			
<i>Blasitocystis hominis</i>	581	400 (69)	181 (31)
<i>Cyclospora cayetensis</i>	7	5 (71)	2 (29)
<i>Cryptosporidium parvum</i>	90	63 (70)	27 (30)
<i>Entamoeba histolytica</i> / <i>E. dispar</i>	40	30 (75)	10 (25)
<i>Giardia lamblia</i>	14	14 (100)	0
Non-pathogenic protozoa			
<i>Chilomastix mesnili</i>	2	2 (100)	0
<i>Dientamoeba fragilis</i>	11	8 (73)	3 (27)
<i>Endolimax nana</i>	26	18 (69)	8 (31)
<i>Entamoeba coli</i>	27	20 (74)	7 (26)
<i>Entamoeba hartmanni</i>	6	5 (93)	1 (7)
<i>Iodamoeba butschlii</i>	10	8 (80)	2 (20)
Helminths			
<i>Ascaris lumbricoides</i>	12	11 (92)	1 (8)

and November. Peak prevalences were observed in *C. parvum* in the spring (March). Oscillations in seasonal frequencies were not dramatic in other parasite species and most were represented in all seasons (Table 1).

Charcot-Leyden crystals. These crystals were found in 34 specimens of which 21 (62%) were infected with *B. hominis*, *C. parvum*, *E. histolytica*/*E. dispar*, and *G. lamblia*. No parasites were detected in the remaining 13 (38%) specimens (Table 3).

DISCUSSION

This study population was demographically similar to the 644 patient population studied during the summer of 1996 under the same circumstances by Parasitology Center, Inc.¹ In the present study, overall infection prevalence rates were comparable throughout the country and did not vary much between the southwest, the west coast, the midwest, and the east coast. Only the larger sample sizes are considered. Patient age was between 0 and 80 years, approximately twice as many females (1,945) than males (951) were tested, relatively more cases (550 of 916, 60%) had a history of foreign travel than non-cases (970 of 1980, 49%) within the last five ($P < 0.001$, by Fisher's exact test, relative risk [RR] = 1.36, confidence interval [CI] = 1.22–1.52. Relatively more infected (110 of 916, 12%) than uninfected (198 of 1980, 10%) patients lived with infected household contacts ($P = 0.117$, not significant, by Fisher's exact test).

Our new method of parasite detection, adopted since 1996, reflected prevalence rates considered closer to true prevalences compared with standard methods used.⁶ By 1998, 3,373 (32.6%) of 10,358 specimens examined at Parasitology Center, Inc. were infected with parasites.⁶ An almost identical prevalence of 32% is reported in this study (Table 1). This prevalence is markedly higher than reported prevalences in the United States of 20% (from 216,275 stool specimens) and 19.7% (from 178,786 stool specimens) reported by state diagnostic laboratories in 1987.² The markedly higher prevalence in our study ($P < 0.001$, by Fisher's exact test, RR = 1.9, CI = 1.84–1.99) suggests real increases in prevalence but does not exclude the possibility of differences in test populations. The results of the latter report also differ significantly from our finding in the composition of the component parasite species found, e.g., *B. hominis* was diagnosed in only 2.6% of the specimens examined compared with 23% (or 72% of all infected cases) in our study. In a 1984 study of 2,360 patients in the United States, prevalences of 20.6% for all parasitic species and 12.2% for cases of *B. hominis* infections were reported, with *B. hominis* constituting 59% of all

infections.³ The latter figure is much closer to our current finding of a *B. hominis* prevalence of 72% among all parasitic infections (Table 1). In 1995, overall *B. hominis* prevalences of 20–30% and greater than 15% were also reported from an unspecified number of patients.⁹

The prevalence of *B. hominis* reported herein (23%) is one of the highest ever reported in the United States and may be epidemiologically significant. Increasing prevalences are noted in more recent years. This prevalence is closest to that reported for Argentina (25%)¹⁰ and Switzerland (16.7–19.0%)¹¹ but considerably lower than those in other studies from Argentina (43%)¹² and Chile (61.8%).¹³

The second most prevalent parasite found was *C. parvum* (Table 1). Prevalences reported in surveys from North America (0.6–4.3%) and Europe (1–2%) are significantly lower than those reported for Asia, Australia, Africa, Central America, and South America (3–20%).¹⁴ *Cryptosporidium parvum* appears to be underdiagnosed in the western hemisphere; its seroprevalence in Europe and North America is usually between 25% and 35%.¹⁴ In a recent survey of 279 children from three clinics along the Texas-Mexico border, 196 children (70.2%) were found infected with *C. parvum*.¹⁵ Children living in a large non-border urban area were less frequently infected, drank more bottled water, and came from households with higher income.¹⁵ *Cryptosporidium* oocysts were observed in 27% of the drinking water samples taken from 66 surface water treatment plants in 14 states and one Canadian province.¹⁶

The *E. histolytica*/*E. dispar* prevalence (Table 1) is markedly higher than the prevalence of 0.9% reported in a large survey in 1987 in the United States, but lower than the estimated prevalence of 4% in the United States.¹⁷ In developing countries with poor sanitation, the prevalence may reach as high as 50%.¹⁷ Prevalences quoted for *E. histolytica* infections are clearly misleading since more than 90% of these infections are due to *E. dispar*.^{17,18} In the Philippines, a polymerase chain reaction survey of 1,872 patients detected 137 stools (7.3%) containing *E. dispar* and 18 stools (1.0%) containing *E. histolytica*.¹⁹ The importance of developing a simple and unexpensive way of distinguishing the two species to obtain information on true prevalence, pathogenicity, and treatment can not be overlooked.

The remaining parasites recovered in this study were of minor importance and their overall prevalence was comparable or somewhat lower than those reported in other surveys. The very low prevalence of *C. cayetensis* agrees with other findings, suggesting underdiagnosis in indigenous populations in the United States.²⁰

Symptoms in 826 singly infected patients did not always agree with the purported pathogenicity of the parasites involved (Table 2). Approximately one-third of *B. hominis* infections were not associated with symptoms. Asymptomatic infections with *B. hominis* varied between 30% and 60% in various populations in the United States.^{1,3,9,21,22} In Canada, *B. hominis* is usually asymptomatic.⁵ It is not known if the degree of pathogenicity of *B. hominis* is related to the distinct immunologic, serologic, and genetic identity of the demes constituting that species.^{23,24} The epidemiologic significance of these findings and the *B. hominis* species complex question remain to be resolved. We regard *B. hominis* as a species complex usually showing pathogenicity based on our findings

TABLE 3

Relationship between charcot-leyden crystals in 34 patients and associated species of parasites in single infections

Parasites species	Number (%) of specimens
<i>Blastocystis hominis</i>	9 (26)
<i>Cryptosporidium parvum</i>	1 (3)
<i>Entamoeba histolytica</i> / <i>E. dispar</i>	8 (24)
<i>Giardia lamblia</i>	3 (9)
None	13 (38)
Total	34 (100)

- (1) table 2) and those reported by others; see the review by Garcia.²⁵ This parasite will cause asymptomatic subclinical infections similar to most pathogens.
- Asymptomatic periods in the intermittent and recurrent infectious cycle of *C. caryeensis* are evident and may be attenuated with long-term suppressive therapy. During the asymptomatic periods, *C. caryeensis* is often absent (undetected) in stool specimens. Asymptomatic infections with *C. parvum* are directly related to the immune status of the host²⁶ and have been previously reported in immunocompetent persons.¹⁴ The asymptomatic cases of *E. histolytica/E. dispar* are attributed to *E. dispar* infections. These observations also apply to the 21 asymptomatic patients of the 90 (23%) multiply infected cases.
- Six species of presumably non-pathogenic protozoa singly infecting 72 patients were associated with symptoms (Table 2). It is our experience that a host body, particularly if immune-compromised, will not be indifferent to the presence of foreign organisms irrespective of their purported non-pathogenic status. Until recently, *B. hominis* was considered to be a harmless yeast. The literature is beginning to show awareness of the pathogenic potential of such "harmless" organisms such as *Dientamoeba fragilis*,^{25,26} *E. coli*,^{1,27,28} and *E. hartmanni*.^{1,25,29} The non-pathogenic status of these organisms is questioned in light of our findings.
- Seasonal studies of human parasite infections are rarely conducted in the United States. An increase in the prevalence of *C. parvum* (comparable to ours) in the spring observed in patients from New Orleans³⁰ and elsewhere¹⁴ was attributed to the warm wet spring weather. However, our overall seasonal data and that of *B. hominis* show the highest prevalence between August and October. The latter dates do not essentially negate possible spring/early summer prevalence peaks. Exposure, experiencing symptoms, seeking medical help, and testing may explain this time lag.
- Charcot-Leyden crystals are breakdown products of eosinophils found usually in feces and occasionally in sputum and body tissues.³¹ The crystals have been traditionally associated with *E. histolytica* infections,³² but have been more recently found in patients infected with *Ancylostoma* spp., *A. lumbricoides*, *Isospora belli*, and *Trichuris trichiura*.²⁵ In the present study, it was also found in patients singly infected with *B. hominis*, *C. parvum*, and *G. lamblia*. The 13 patients with crystals but with no detectable infections were probably infected with a cyclic parasite such as *E. histolytica*.
- Acknowledgments: Appreciation is extended to Karim O. Amin for technical help and support.
- Author's address: Omar M. Amin, Parasitology Center, Inc., 903 South Rural Road, #101-318, Tempe, AZ 85281 and Laboratorio Analisis Clinicos, Av. Obregon 28-9, Nogales, Sonora, Mexico.
- Reprint requests: Omar M. Amin, PO Box 28372, Tempe, AZ 85285.
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