SEASONAL PREVALENCE OF INTESTINAL PARASITES IN THE UNITED STATES DURING 2000

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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. 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. 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 States2,3 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, 23%), Utah (5 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

<table>
<thead>
<tr>
<th>Month</th>
<th>Number of specimens examined</th>
<th>Number (%) of specimens infected</th>
<th>Number (%) of parasite species identified in single and multiple infections*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Single infections</td>
<td>Multiple infections</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Total</td>
<td>AL</td>
</tr>
<tr>
<td>January</td>
<td>155</td>
<td>38 (24)</td>
<td>4 (3)</td>
</tr>
<tr>
<td>February</td>
<td>283</td>
<td>58 (20)</td>
<td>4 (1)</td>
</tr>
<tr>
<td>March</td>
<td>254</td>
<td>69 (27)</td>
<td>10 (4)</td>
</tr>
<tr>
<td>April</td>
<td>228</td>
<td>60 (26)</td>
<td>5 (2)</td>
</tr>
<tr>
<td>May</td>
<td>326</td>
<td>71 (22)</td>
<td>10 (3)</td>
</tr>
<tr>
<td>June</td>
<td>268</td>
<td>65 (24)</td>
<td>6 (2)</td>
</tr>
<tr>
<td>July</td>
<td>198</td>
<td>58 (29)</td>
<td>6 (3)</td>
</tr>
<tr>
<td>August</td>
<td>304</td>
<td>94 (31)</td>
<td>17 (5)</td>
</tr>
<tr>
<td>September</td>
<td>227</td>
<td>82 (36)</td>
<td>13 (6)</td>
</tr>
<tr>
<td>October</td>
<td>198</td>
<td>65 (24)</td>
<td>6 (2)</td>
</tr>
<tr>
<td>November</td>
<td>181</td>
<td>56 (31)</td>
<td>6 (3)</td>
</tr>
<tr>
<td>December</td>
<td>177</td>
<td>50 (28)</td>
<td>7 (4)</td>
</tr>
<tr>
<td>Total</td>
<td>2,896</td>
<td>826 (29)</td>
<td>90 (3)</td>
</tr>
</tbody>
</table>

*The total number of specimens infected may be more than the number in the preceding columns because of multiple infections.

AL = Ascaris lumbricoides; BH = Blastocystis hominis; CM = Chilomastix mesnili; CC = Cyclospora cayetensis; CP = Cryptosporidium parvum; DF = Dientamoeba fragilis; EN = Endolimax nana; EC = Entamoeba coli; EH/ED = Entamoeba histolytica/E. dispar; EH = Entamoeba hartmanni; GL = Giardia lamblia; IB = Iodamoeba butschlii. Other parasites not listed include: Ancylostoma/Necator (1 specimen); Diphyllobothrium latum (1); Enterobius vermicularis (1); Taenia sp. (1); and Trichuris trichiura (2).
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. 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 years, less than 10% of the patient population studied during the summer of 1996 lived with infected household contacts (110 of 916, 12%) than uninfected (198 of 1980, 10%) patients, 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. By 1998, 3,373 (32.6%) of 10,358 specimens examined at Parasitology Center, Inc. were infected with parasites. 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 diarrhea surveillance. The very low prevalence of E. histolytica/E. dispar reported in a large survey reported in 1987 in the United States, but lower than the estimated prevalence of 4% in the United States. In developing countries with poor sanitation, the prevalence may reach as high as 50%. Prevalences quoted for E. histolytica infections are clearly misleading since more than 90% of these infections are due to E. dispar. 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. The importance of developing a simple and inexpensive 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.

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. In Canada, B. hominis is usually asymptomatic. 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. 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**

<table>
<thead>
<tr>
<th>Parasites species</th>
<th>Number (% of specimens)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blastocystis hominis</td>
<td>9 (26)</td>
</tr>
<tr>
<td>Cryptosporidium parvum</td>
<td>1 (3)</td>
</tr>
<tr>
<td>Entamoeba histolytica/E. dispar</td>
<td>8 (24)</td>
</tr>
<tr>
<td>Giardia lamblia</td>
<td>3 (9)</td>
</tr>
<tr>
<td>None</td>
<td>13 (38)</td>
</tr>
<tr>
<td>Total</td>
<td>34 (100)</td>
</tr>
</tbody>
</table>
(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. cayetensis are evident and may be attenuated with long-term suppressive therapy. During the asymptomatic periods, C. cayetensis is often absent (undetectable) 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 infected cases.

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Exposure, experiencing symptoms, seeking medical help, and testing may explain this time lag. 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 Orleans29 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 trichura.22 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.

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