Intestinal Parasites in Cambodians: Comparison Of Diagnostic Methods Used In Screening Refugees With Implications For Treatment Of Populations With High Rates Of Infestation

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Abstract: **We performed a retrospective review of screening parasitology examinations on a Cambodian refugee population served by an urban neighborhood health center. Five-hundred twenty of 1084 patients were examined for ova and parasites either by purged stool, which was examined immediately, or preserved stool, examined at a teaching hospital and proprietary laboratories. Overall, 335 (64 percent) of the tested patients had at least one parasite. The prevalence of infection varied by test technique (purged stool examined immediately, 86 percent; preserved stool examined at a hospital, 65 percent; preserved stool sent to a proprietary laboratory, 31 percent, $P < 0.01$).**

In this population where *Entamoeba histolytica* infection was 44 percent as measured by the purged warm stool technique, the cold preserved stool test had a measured relative sensitivity of 33 percent. Assuming a selectivity of 99 percent, it would take eight negative tests to reach a > 95 percent negative predictive value.

The high rate of intestinal carriage of pathogenic parasites in this population and the insensitivity of commonly available diagnostic tests make routine presumptive treatment of intestinal parasites an option when the purged stool examination is unavailable. (J Am Board Fam Pract 1991; 4:71-8.)

Since 1974, more than 884,474 Southeast Asian refugees have immigrated to the United States—75,130 arrived in 1987 and 1988.[1] By September 1988, 32,000 Indochinese refugees had been settled in New York State,[1] and more than 1000 are provided primary health care by the Family Health Center (FHC) of Montefiore Medical Center in the Bronx, N.Y.

Health problems affecting this population differ from those of the general U.S. population in significant ways. Earlier surveys have shown a high rate of tuberculosis, middle ear infections, anemia, hepatitis B antigenemia, positive VDRL tests, and psychological disorders, as well as intestinal parasites.[2-7]

In 1981, at the beginning of the settlement of Indochinese in the Bronx, protocols were developed for health maintenance in this population. During the summer of 1987, we reviewed our experience with Cambodians to study the prevalence of infestation with intestinal parasites and the accuracy of methods of diagnosis available to our clinic. Health care providers at the FHC had suspected that our established practice was inadequate because many patients were never examined for parasites in spite of protocols stating that this should be done. Moreover, many who were screened as negative later became symptomatic from intestinal parasites, which either should have been detected during initial screening or were contracted from another person who had not been screened.

While our review generally confirmed published data about the prevalence of infection with intestinal parasites in Indochinese refugees, it also showed serious problems with our diagnostic practice. This forced us to reassess our diagnostic and therapeutic strategies and to design a new protocol that would increase the number of refugees successfully treated for intestinal parasites. Though new immigration from Indochina has slowed, there remains a large population of infested Indochinese who have not been treated.

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They continue to be at risk for complications, and their community is at risk for further spread of some of these organisms.

Methods

During the summer of 1987 all the charts of Cambodian patients at the Montefiore Family Health Center were reviewed by the medical and nursing staff to evaluate our parasitology testing. The Family Health Center is a federally funded neighborhood health center serving approximately 18,000 residents of the Northwest Bronx. There were 1084 charts dating from 1980 to 1987. Because the charting system at the Family Health Center includes households, some charts included persons who had never been seen. Data recorded included age, gender, year of arrival to the United States, date tested for intestinal parasites, type of test, and laboratory performing the test.

As part of a screening protocol for asymptomatic patients, parasitology examinations were performed using either purged warm stools or preserved stools sent to the Montefiore Medical Center laboratory or to proprietary laboratories. Patients were not randomly allocated to any particular method; the choice of method rested on individual provider preference as well as the varying availability of tests. For the purged fresh stool technique, patients were sent to the Montefiore Medical Center Parasitology Laboratory where their stools were obtained following an oral purge with magnesium citrate and examined by the direct wet film technique and zinc sulfate concentration method.[8] For the preserved stool technique, patients were given kits consisting of three bottles, one containing polyvinylalcohol (PVA) preservative, another with formalin, and a third empty. These were returned with stool samples and sent to the Montefiore Hospital laboratory, where they were examined by the direct wet film technique, the formalin ether concentration method, and the trichrome stain technique.[9,10] Alternatively, collection kits containing bottles with PVA preservative and formalin, respectively, were sent to a proprietary laboratory where they were examined first by the formalin concentration method. If any suspicious signs were found, the trichrome staining technique was performed on the PVA preserved sample.

Test results were sorted by type of test (warm versus cold) and by laboratory. Further stratification was done by year of testing and age and sex of patient. Age groups were comparable with those in published reports. Analysis compared prevalence of all parasites with prevalence of pathogenic parasites. Organisms that were considered potentially pathogenic were *Entamoeba histolytica*, *Ascaris lumbricoides*, *Blastocystis hominis*, *Clonorchis sinensis* (and other flukes), *Giardia lamblia*, hookworm, *Hymenolepis nana*, *Strongyloides stercoralis*, and *Trichuris trichiura*.

The chi-square test was used to compare the distribution of parasites in the stratified cohorts and demographic characteristics of the groups by test methodologies. The level of significance was $P \leq 0.05$. Confidence limits for means values were calculated using the Student t-distribution ($P < 0.05$). Individual testing methods were compared for percent positive by the pooled *t*-test.

Results

Stool examinations were performed on 520 of the 1084 Cambodian patients who registered at the Family Health Center between 1981 and 1987. Three-hundred thirty-five patients (64 percent) harbored at least one intestinal parasite, and 302 patients (58 percent) had at least one known pathogen. One-hundred ninety-four patients (37 percent) harbored more than one parasite, and 165 patients (32 percent) had more than one

Table 1. Intestinal Parasites in Cambodian Refugees by Purged Stool Method (n = 271).

Intestinal Parasite	n	Percent Positive (±SEM)
Necator americanus	152	56 (±5.9)
Entamoeba histolytica	118	44 (±5.9)
Strongyloides stercoralis	78	29 (±5.4)
Hymenolepis nana	43	16 (±4.4)
Giardia lamblia	35	13 (±4.0)
Entamoeba coli	21	8 (±3.2)
Entamoeba nana	15	6 (±2.7)
Blastocystis hominis	13	5 (±2.5)
Ascaris lumbricoides	13	5 (±2.5)
Trichuris trichiura	10	4 (±2.2)
Dientamoeba fragilis	7	3 (±1.9)
Other	44	16 (±4.4)
Total	549*	

*233/271 patients positive for ≥ 1 parasites, 169 patients had ≥ 2 parasites.

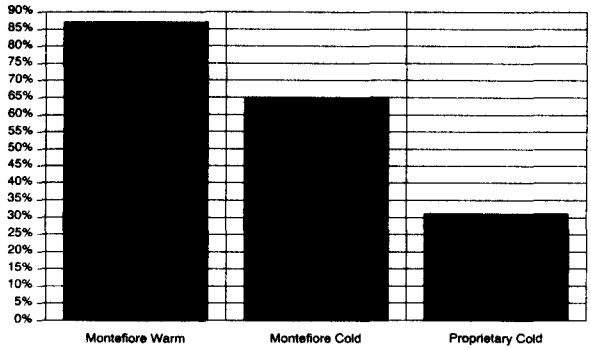

Figure 1. Percent of Cambodian patients who were positive for infection with intestinal parasites by different laboratory techniques.

pathogen. The most common parasites found are listed in Table 1.

There were 233 of 271 (86 percent) patients examined by the purged warm stool technique who were positive for parasites; 38 of 59 (65 percent) of those tested by preserved kit at the hospital laboratory were positive, and 59 of 190 (31 percent) who were tested by preserved kit at the proprietary laboratories were positive (Figure 1). These rates were not significantly changed when only pathogenic parasites were considered.

The superiority of the warm purged stool test also tended to hold up when analysis was done by individual parasite species, though it varied in magnitude depending upon the organism. The differences between the warm stool test and the preserved stool tests in respect to their relative abilities to detect infestation were most striking in regard to *Entamoeba histolytica*, where the purged test was 10 times more sensitive ($P < 0.01$); *Strongyloides stercoralis* (10 times more sensitive, $P < 0.01$); and hookworm (9 times more sensitive, $P < 0.01$). The purged stool method was more than twice as sensitive as the preserved kit method for *Ascaris*; however, the significance only reached $P < 0.10$. The differences between tests for detection of *Giardia lamblia* and *Tricuris trichiura* were not significant. *Blastocystis hominis* was more easily found with the hospital kit examination than the purged stool examination ($P < 0.01$) (Figure 2).

Different age groups showed different distributions of infecting organisms. *Giardia lamblia* and *Hymenolepis nana* were much more common in ages 0–19 years than in the older groups. Conversely, hookworm was much more common in groups older than 19 years. *Entamoeba histolytica* was found equally in all age groups (Figure 3).

The tested population was similar to all registered Cambodians in gender distribution (44 percent male, 51 percent female and 5 percent not noted) ($P < 0.05$). Patients whose stools were examined in the Montefiore Hospital Laboratory by the cold stool method were similar to the general Cambodian population in age distribution ($P < 0.05$). However, patients in the other two categories were not. There was a preponderance of children 0–18 years old in the tested group when compared with all registered Cambodians. The age distribution also was skewed in the group tested by the purged stool technique because it is restricted at our hospital to those > 10 years.

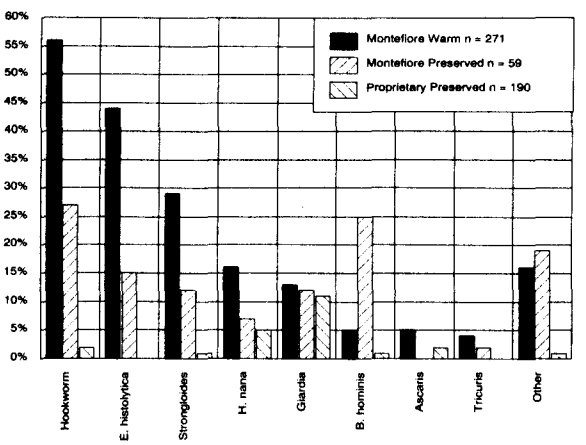

Figure 2. Percent of common pathogenic intestinal parasites by different laboratory techniques.

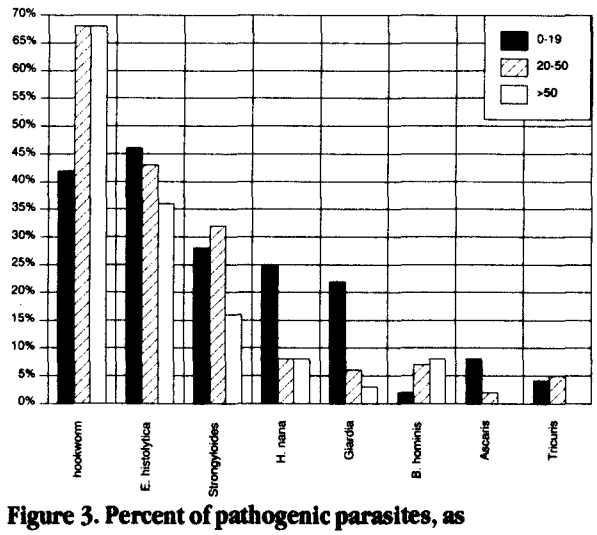

Figure 3. Percent of pathogenic parasites, as determined by the purged stool method, analyzed by age group.

Table 2. Percentage of parasite examinations positive for each of three methods as subdivided by year of patient's arrival in U.S. ± values are ±SEM calculated for a binomial distribution.

Year Arrived	Number Tested*	Percent Positive Montefiore Warm	Percent Positive Montefiore Preserved	Percent Positive Proprietary Preserved
Not known	30	81±2.0	0±24.1	8±3.9
1980	1	100±32.5	NA	NA
1981	89	88±0.4	50±24.1	100±46.0
1982	118	90±0.3	50±8.0	60±9.3
1980–82	207	89±0.2	50±6.0	67±7.8
1983	65	85±0.8	43±6.9	18±2.7
1984	96	83±2.7	70±2.4	36±0.7
1985	91	80±6.5	53±3.2	32±0.7
1986	28	100±8.1	100±6.9	24±2.7
1986–87	31	100±8.1	100±6.9	25±2.3

*Sum of all test types.
NA. Not applicable. No tests done using this method.

The mix of tests used for screening and diagnosis varied from 1981 to 1987. Though the prevalence of infestation varied somewhat from year to year, the relative sensitivities of each method of testing did not vary significantly when analyzed by year of patient arrival to the U.S. (Table 2).

While steps were taken during the chart review to identify patients who were symptomatic from intestinal infestation, this information was dropped from the data when an extraordinarily low rate of symptoms was noted (under 1 percent). We could not discern whether providers were prevented from detecting symptoms because of language and cultural barriers and time pressures or whether the patients were truly asymptomatic. Therefore, our data should be taken as derived from a screening protocol for patients receiving general health maintenance.

Discussion

In this retrospective study of recent immigrants from Cambodia, we found an opportunity to survey the rate of infestation and to compare different diagnostic methodologies within a generally homogeneous population. Overall, 64 percent of those studied were infested with at least one parasite. When the results were separated by diagnostic method, 86 percent of those examined by the purged warm stool method were positive for at least one parasite compared with 65 percent positive when preserved stools were examined by the hospital parasitology laboratory and 31 percent when preserved stools were sent to the proprietary laboratories.

These findings are consistent with prior surveys of Indochinese refugees when the studies are stratified by laboratory methodology (Figure 4). Studies using stools preserved by PVA and formalin showed a 60–65 percent rate of infestation,[3,4,6,7] which correlates well with our hospital's rate of 65 percent using similar methodology. One study using unpreserved stool samples[2] recorded a rate of 35 percent, which nearly matches the rate found by the proprietary laboratories where we sent samples and where full examinations were done only on samples that appeared "suspicious." Another survey, which found a 32 percent rate, was done on patients who had been residing in the U.S. for at least 2 years, many of whom had been previously treated.[11] The only reported study that used purged warm stool samples[5] showed an 82 percent rate of infestation, which compares well to our laboratory's rate of 86 percent.

While the subpopulations examined by each method were comparable with the total Cambodian population served by our facility in regard to gender, they were not comparable for distribution among age groups. However, the differences in percent infested persisted when tests were compared within age groups.

Limitations of Our Study

The measured rate of infestation might have been biased by a tendency for ill persons to seek medical care and therefore be tested. The differences in rate of infestation, however, did not greatly vary between groups that were over-

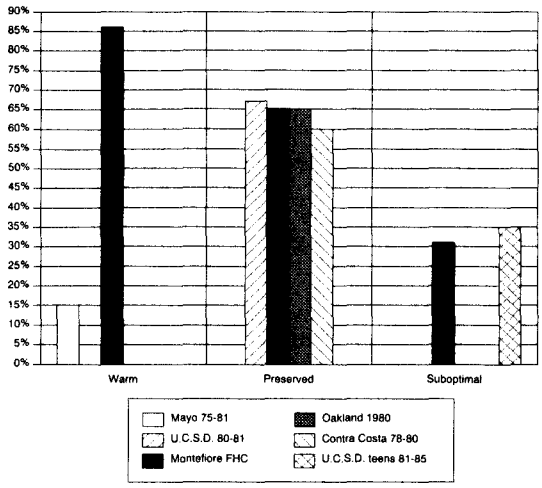

Figure 4. Percent of parasite infection in various published studies of Indochinese refugees sorted by testing methodology.

represented (0–20 years because of requirements for immunization and physical examinations for entry into public school) and those underrepresented (> 50 years).

While the false-positive rate for the warm stool examination is not known, we do not believe that overreading of samples at the Montefiore laboratory was a factor in the difference between the preserved and purged stool methods because the same technicians performed both tests. The purged warm stool method of examination has been cited by other sources as far superior to cold stool techniques.[8,12] We believe, therefore, that the rate of parasite infestation found by the purged warm stool method at Montefiore most closely represents the true rate of infestation in Cambodian refugees. Nonetheless, it has been our experience that after treatment, follow-up tests frequently reveal parasites that were not found on the original test. Whether this is a new infestation or a previously missed one is not clear. The latter seems most likely when the organism is one that cannot be transmitted directly from person to person. We believe that the true prevalence of intestinal parasites in this population is most likely higher than that found by one screening warm stool examination.

It is not clear why one test was chosen over another by a particular provider (physician or nurse practitioner). In later years, the warm stool test became less accessible because of limited ability to escort and translate for the Khmer-speaking patients. It was also difficult to order a "stat" warm stool test because of the long waiting list for Cambodian patients (translators needed to be present). Providers might have been inclined to order the cold stool examination in cases where there was some diagnostic urgency. The long wait for parasitology testing by the purged warm stool technique (occasionally up to 3 months) certainly discouraged some patients and may have contributed to the relatively low number of completed examinations in the population surveyed. It most likely contributed to the bias toward cold stool tests done in the years 1984–1987 (Table 2). Because of the charting system at the Family Health Center, where entire families are registered and charted together, many patients were never seen at the clinic, and this contributed to the low examination rate (50 percent).

Other Studies with Low Testing Rates

The low rate of completed tests is not confined to our clinic. Fourteen percent of examined refugees were unable to return a stool sample at the Center for Indochinese Health Education.[7] Ten percent of the teenagers examined at the University of California, San Diego, were unable to return stool samples.[2] A survey of Cambodian refugees 2 years after immigration reported 49 percent of patients were examined for ova and parasites.[11] In a study similar to ours, where all patient charts were reviewed (rather than only patients' seen for screening), physicians' compliance with published guidelines varied from 11 percent for mammography to 48 percent for stool blood testing and 75 percent for Papanicolaou smears.[13] This is consistent with other surveys where poor completion rates of recommended screening tests were reported.[13-17] The difficulty in arranging the warm stool testing and the need for complicated instruction and return visits for the diagnosis of intestinal parasites is a difficult task for some members of this population newly arrived in the U.S. Coincident with medical problems, there are social problems of an indigent immigrant population requiring investment in outreach, follow-up, social work, and translation. For busy health care providers, the task of explaining and obtaining a stool test is a task one might easily set aside.

Earlier surveys generally have sampled Indochinese refugees without subdividing by ethnicity, with the exception of some that focused on Vietnamese. One study that examined rate of

stool parasites by ethnic origin[3] did show significant differences in nationalities, though the total number of Cambodian samples was small (27 were examined). It should come as no surprise that Vietnamese residing in urban camps in Hong Kong experience different infectious diseases than Cambodians residing in rural camps in Thailand. By focusing exclusively on Cambodians, we assume that a more valid estimate of parasite rate has been obtained. It, therefore, can allow us to design appropriate protocols for presumptive treatment of this ethnic group.

Importance of Treatment

The very high rate of infestation with *Entamoeba histolytica* (36–47 percent), *Strongyloides stercoralis* (16–30 percent), and hookworm (43–68 percent) should be viewed with concern. *E. histolytica* is highly infectious and capable of causing colitis, dysentery, and hepatic abscess. Hookworm can cause chronic blood loss, sometimes resulting in profound anemia. *Strongyloides* is also directly transmissible and capable of causing serious complications. The potential ill effects of treating persons who are not infested must be weighed against the problems of not treating people who are infested. The treatment for *Ascaris*, *Trichuris*, and hookworm (mebendazole) is cheap and has few complications.[18] Similarly, *Hymenolepis nana* is easily treated with a single dose of praziquantel.[18] The treatment for *Strongyloidiasis* (thiabendazole) is slightly more toxic (frequently being complicated with occasional nausea and vertigo), but the short course makes these symptoms tolerable.[18] The treatments for *E. histolytica*, *Giardia*, and *B. hominis* are more problematic. Potential side effects are increased because of the necessary length of treatment, but all three could be treated by either metronidizole or iodoquinol plus quinicrine.[18]

We suspect that other practitioners have at times been tempted to abandon laboratory testing as redundant when the existence of a condition has increased. This urge is usually countered by the need for independent confirmation of clinical impression prior to treatment with costly or toxic drugs. However, laboratory testing itself can be costly, and when there is a high rate of disease, it can be potentially misleading. Interest in independent objective confirmation may create greater diagnostic uncertainty because of false-negative results, thereby missing the presence of potentially harmful infestation, which can lead to later morbidity and mortality.

A Rationale for Presumptive Treatment

Rather than expending limited resources for screening, it may be more cost effective among Cambodian refugees to provide presumptive treatment to a greater percentage of the population. A 1985 cost-benefit analysis applying the General Health Policy Model and its Quality of Well-being Scale (which includes factors of morbidity associated with illness, such as hospitalization, medication, and diagnostic costs, as well as the usual considerations of lost work time)[7] showed clear benefit in diagnosing and treating Indochinese refugees for intestinal parasites. When the rate of pathogenic parasites found in our study was substituted for those used in the above reference's cost-benefit analysis, the discounted cost-benefit utility went from $25,635 per well year produced to a net *saving* of $76,539. This saving would increase to $94,998 per well year produced if the costs of screening tests and examination were eliminated. These savings might be better actualized by assuring proper follow-up and treatment of all Indochinese refugees rather than by looking for parasites that are most likely present. To cancel this net benefit, treatment complications would have to cause hospitalizations in 1 percent of persons treated.

The rationale for presumptive treatment is even stronger in cases where fresh purged stool examinations are not available and false-negative tests are therefore more likely to occur. In our population, where the rate of *E. histolytica* infection was 44 percent as measured by the purged warm stool technique, the cold preserved stool test done at our laboratory had a measured relative sensitivity of 33 percent (because the purged stool test is imperfect, the true sensitivity would likely be less). Assuming a specificity of 99 percent, the predictive value of a negative test is only 60 percent. The predictive value of three consecutive negative tests is only 77 percent. It would take eight negative tests to reach a > 95 percent negative predictive value (Figure 5). Clearly, in our population, a single cold preserved stool test is unacceptable as a means for ruling out infestation with intestinal parasites.

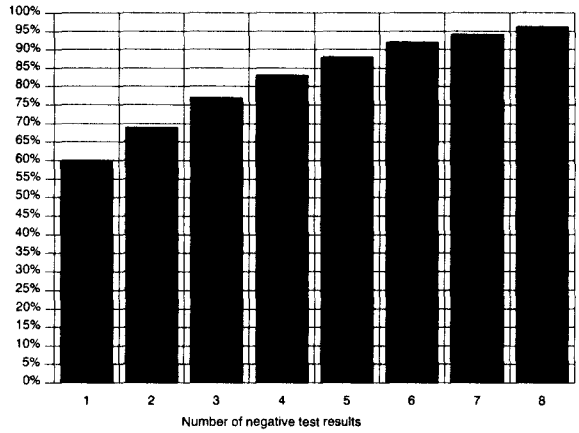

Figure 5. Negative predictive value of preserved stool test for *Entamoeba histolytica* for increasing number of negative test results where relative sensitivity = 33 percent, specificity = 99 percent, and prevalence = 50 percent.

This is in contrast to its use in a population with a low rate, when the negative predictive value would be high and the rationale for screening has to be questioned.

The ability of the cold preserved stool test to screen for intestinal parasites in our population is unacceptably low. A negative stool ova and parasite test (or even three negative tests) when done by the preserved stool technique can provide a false sense that intestinal parasites have been ruled out in this population. This can delay or even confound the correct diagnosis in persons who later become symptomatic, creating an even greater cost in potential morbidity and mortality.

While the purged stool technique does not suffer this flaw of insensitivity, its cost (both financial as well as difficulty of procurement) makes it hard to justify as a screening tool for a population where the rate of infestation is so high.

Proposal for Treatment

We suggest abandoning diagnostic screening in this group and treating presumptively. We recommend that newly arrived Cambodian refugees who are 19 years or older be treated with 3 days of mebendazole, which is effective against hookworm, *Trichuris*, and *Ascaris*. This should be followed by thiabendazole for 2 days to treat *Strongyloides*, then iodoquinol for 20 days to treat intraluminal *E. histolytica*. For children 6–19 years, a similar regimen could be instituted with the addition of one dose of praziquantel to treat *Hymenolepis nana*. In this age group, metronidizole might be substituted for lodoquinol in order to treat *Giardia lamblia*, or the lodoquinol should be followed by a course of diiodohydroxyquin.

We recognize that these are somewhat complicated regimens that will require evaluation for compliance, acceptability, and effectiveness. Alternative regimens can be entertained for this as well as other immigrant groups. The strategy of testing for asymptomatic infection prior to treatment misses too much to justify its continued use.

Cambodians appear to tolerate and comply with treatment better than with our earlier strategy of asking for stool samples. Certainly, our clinicians' time appears to be better spent in administering and monitoring treatment than in ordering questionable diagnostic tests and risking the nontreatment of a large percentage of infested individuals because of test failures and failures to test.

In other immigrant groups that suffer from high burdens of intestinal parasites, we recommend an initial survey, either using the purged stool method or multiple samples examined by the preserved stool method, to determine the nature and estimate the prevalence of parasites. After that, protocols for presumptive treatment can easily be constructed.

Diagnostic testing should be reserved for symptomatic patients and possibly for follow-up tests of their cure. Diagnostic tests preferably should be of the fresh purged stool type. When this is not available, and ruling out parasite infestation is imperative, eight separate samples rather than the traditional three should be obtained.

References

1. U.S. Committee for Refugees. 1988 statistical issue. Refugee Reports 1988; 12:7.
2. Fitzpatrick S, Johnson J, Shragg P, Felice ME. Health care needs of Indochinese refugee teenagers. Pediatrics 1987; 79:118-24.
3. Tittle BS, Harris JA, Chase PA, Morrell RE, Jackson RJ, Espinosa SY. Health screening of Indochinese refugee children. Am J Dis Child 1982; 136:697-700.
4. Catanzaro A, Moser RJ. Health status of refugees from Vietnam, Laos, and Cambodia. JAMA 1982; 247:1303-8.
5. Sutherland JE, Avant RF, Franz WB 3d, Monzon CM, Stark NM. Indochinese refugee health assessment and treatment. J Fam Pract 1983; 16:61-7.
6. Ariaa F. Intestinal parasites among Indochinese refugees and Mexican immigrants resettled in Contra

Costa County, California. J Fam Pract 1981; 12: 223-6.
7. Anderson JP, Moser RJ. Parasite screening and treatment among Indochinese refugees. JAMA 1985; 253:2229-35.
8. Faust EC, D'Antoni JS, Odom V, et al. A critical study of clinical laboratory technology for the diagnosis of protozoan cysts and helminth eggs in feces. Am J Trop Med Hyg 1938; 18:169.
9. Young KH, Bullock SL, Melvin DM, Spruill CL. Ethyl acetate as a substitute for diethyl ether in formalin-ether sedimentation technique. J Clin Microbiol 1979; 10:852-3.
10. Melvin DM, Brooke MM. Laboratory procedures for the diagnosis of intestinal parasites. Atlanta: Centers for Disease Control, 1982. (DHEW publication no. 82-8282).
11. Molina CD, Molina MM, Molina JM. Intestinal parasites in southeast Asian refugees two years after immigration. West J Med 1988; 149:422-5.
12. Markell E, Marietta V, John D. Medical parasitology. 6th ed. Philadelphia: W.B. Saunders, 1986.
13. Woo B, Woo B, Cook EF, Weisberg M, Goldman L. Screening procedures in the asymptomatic adult. Comparison of physicians' recommendations, patients' desires, published guidelines, and actual practice. JAMA 1985; 254:1480-4.
14. Romm FJ, Fletcher SW, Hulka BS. The periodic health examination: comparison of recommendations and internists' performance. South Med J 1981; 74:265-71.
15. Cohen DI, Littenberg B, Wetzel C, Neuhauser D. Improving physician compliance with preventive medicine guidelines. Med Care 1982; 20:1040-5.
16. McDonald CJ, Hui SL, Smith DM, et al. Reminders to physicians from an introspective computer medical record. A two-year randomized trial. Ann Intern Med 1984; 100:130-8.
17. Davidson RA, Fletcher SW, Retchin S, Duh S. A nurse-initiated reminder system for the periodic health examination. Implementation and evaluation. Arch Intern Med 1984; 144:2167-70.
18. Drugs for parasitic infections. Med Lett Drugs Ther 1990; 32:23-32.