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Parasitic Diseases in the Compromised Host

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1. Introduction

In the United States, Western Europe, and the mature health care delivery systems of Asia and Africa, two parasitic diseases should be carefully considered in the evaluation of fever and possible infection in the immunocompromised host: toxoplasmosis and pneumocystosis. In addition, three other diseases—strongyloidiasis, giardiasis, and babesiosis—occasionally occur in patients who are immunosuppressed or splenectomized. The ongoing epidemic of acquired immunodeficiency syndrome (AIDS) has called our attention to two coccidial protozoa—*Cryptosporidium* and *Isospora belli*—as causes of diarrhea. These entities should be suspected in any patient with compromised cell-mediated immunity and symptoms of gastroenteritis. It should, however, be acknowledged that in many of the developing countries of the world a number of other common parasitic entities can be expected to afflict both immunosuppressed and normal hosts. Thus, malaria and Chagas disease are occasional causes of transfusion-associated infection, and the risk of hematogenous trypanosomiasis is sufficiently great that in Brazil serologic screening for evidence of infection is analogous to the mandatory requirements for hepatitis serological testing in the United States.[1]

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Malaria has occasionally been reported in immunosuppressed patients in the United States who are presumably infected by the transfusion route[2]; fatal leishmaniasis (kala-azar) was observed in a renal transplant recipient who probably had reactivation of latent infection.[3] Clearly, a carefully taken history of travel or transfusion of blood products will be extremely important in ruling out unusual parasitic disorders. Not to be overlooked is the possibility of reactivation of disease initially acquired during travel to an endemic area but that remains quiescent for many years until host defenses are impaired.

2. *Pneumocystis carinii* Pneumonia

Few diseases of the immunocompromised host have proved as fascinating to the infectious disease clinician during the last decade than that associated with the presumed protozoan parasite, *Pneumocystis carinii*. During the more than 75 years that have elapsed since the description of the organism, numerous investigators have attempted in vitro cultivation of this pathogen. Successful cultivation in vitro has probably been achieved.[4] Nevertheless, the methods for propagation are quite complex and growth in vitro is not of the magnitude commonly obtained for commensal bacteria. Although Koch's postulates have not been fulfilled, and the disease remains a nosologic problem, effective chemotherapy and prophylaxis are now available. Reviews of recent developments in the biology and treatment of *Pneumocystis* infection have been published.[5,5a]

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2.1. Historic Perspective

The first description of pneumocystosis in animals and humans should properly be attributed to Chagas, who observed cyst forms in the lungs of guinea pigs that were also infected with *Trypanosoma cruzi*.[6] Chagas interpreted these forms as the sexual stage or the sporogonia of the trypanosome. The following year, Carini, also working in Brazil, observed similar cysts in lungs of rats experimentally infected with another trypanosome, *T. lewisi*. Carini's observation became known to the husband-and-wife team, the Delanoes, who saw similar cysts in the lungs of Parisian sewer rats and gave these cysts the name *Pneumocystis carinii*. In 1913, Chagas also described in an autopsy of an adult the first probable case of pneumocystosis in man. Some 30 years subsequently elapsed before the description of additional human cases of pneumonic disease associated with the presence of these cysts in lung tissue. During World War II, Vandermeer and Brug observed such cases of pneumonitis in malnourished orphans.[7] During the two decades following the end of World War II, outbreaks of pneumocystosis in premature infants and institutionalized orphans in Europe and the Middle East were described. These illnesses were associated with prematurity and malnutrition. In a study of autopsied infants that was published in 1952, Vanek and Jirovec provided the strongest histopathologic evidence for the etiologic relationship between *Pneumocystis* organisms and interstitial plasma cell pneumonia.[8] For this reason, Frenkel has argued that the human parasite should be called *Pneumocystis jirovecii* in honor of Dr. Otto Jirovec rather than *Pneumocystis carinii*.[9] The latter is responsible for pneumonia in animals, and proof that the organisms causing disease in humans and animals are identical is lacking.

In 1958, Ivady and Paldy, working in Hungary, reported the first successful use of pentamidine in the treatment of pneumocystosis.[10] This agent was previously used successfully in the treatment of African sleeping sickness. Following this successful therapeutic approach and the recognition of cases in immunodeficient children in the United States,[11] the National Centers for Disease Control assumed responsibility for distribution of pentamidine in the United States.

During the mid-1960s, it was recognized that a combination of pyrimethamine and sulfadiazine was also effective in the treatment of human pneumocystosis. Whether or not this combination yields treatment results equivalent to or perhaps better than pentamidine has never been resolved. Recent therapeutic advances have been based on carefully designed studies of experimental corticosteroid-induced infections in rats.[12] Using this model, Hughes and collaborators reported in 1974 the effectiveness of trimethoprim–sulfamethoxazole (another antifolate–sulfonamide combination) in therapy and prophylaxis of rat pneumocystosis.[13] Thereafter, the safety and therapeutic effectiveness of trimethoprim–sulfamethoxazole was demonstrated in *Pneumocystis*-infected children[14] and adults.[15] More recently, trimethoprim–sulfamethoxazole was successfully employed in the prophylaxis of *Pneumocystis carinii* infection in leukemic children[16] and in bone marrow transplant recipients.[17]

There is increasing recognition that any patient who is immunosuppressed and has dyspnea or lung infiltrates should be considered a possible case of pneumocystosis. As a result of the AIDS epidemic we have come to appreciate that *P. carinii* is the most common cause of life-threatening opportunistic infection and is usually the presenting infection that establishes the diagnosis of AIDS.[18,19] Any patient with pneumonia who does not belong to one of the high-risk groups predisposed to AIDS should still be considered for the diagnosis if he or she has received a blood transfusion since 1980. The prognosis with treatment of the first episode is still fairly good but recrudescent disease is common. Therapeutic failures, toxicity, or hypersensitivity to available medications has spurred the search for alternative agents for treatment.[20] At the same time the AIDS epidemic has triggered research into the basic biology of the disease, improved methods for diagnosis, and renewed investigation of prophylactic approaches.[20a]

2.2. The Organism

There have been several reports of limited (short-term) passage of *Pneumocystis* organisms in tissue culture from lungs of animals or humans with pneumonitis.[4] The belief that the forms identified as *Pneumocystis* are viable and pathogenic is based on their presence and uniform morphology in the pulmo-

nary alveolar spaces of patients and animals with pneumonitis, followed by their disappearance after treatment with folate antagonists—sulfonamides or pentamidine. Descriptions of the morphology of this microbe have been based on light and electron microscopy of pathologic material and study of organisms passed in tissue cultures. The organism appears to be a unicellular microbe with both cyst and trophozoite stages. The cystic unit contains up to eight oval bodies most often termed sporozoites. The cysts are best seen in silver-stained smears of infected fresh lung, whereas the sporozoites are better defined by Giemsa-stained imprint smears. Each sporozoite (or intracystic body) measures about 1–2 μm and has a deeply staining eccentric nucleus and a pale blue staining halo that is thought to be cytoplasm. However, the Giemsa stain is probably not the best technique for rapid recognition of pneumocysts, because it also stains background alveolar material and cell fragments rather than the characteristic cyst wall of the organism. Alternative methods for detecting *Pneumocystis* forms include the Gram–Weigert stain, which stains both cyst and sporozoite, and the toluidine blue stain, which delineates the cyst form but not the intracellular morphology.

The Gomori methenamine silver nitrate stain is, in our experience, the most reliable (but most time-consuming) procedure for identification of cyst forms in lung tissue. Pneumocysts identified by this silver stain have a thin, sometimes irregular gray/brown/black capsule and may appear almost round, disc-shaped, or crescentic in form (Fig. 1). These cysts are 4–6 μm in diameter and are almost as large as a red blood cell (RBC), often being mistaken for red cells if the silver stain is incorrectly carried out. Cysts often occur in clusters within the alveolar space. The internal structure of the cyst is variable. In lighter-stained round cysts, a pair of structures about 1 μm in length resembling opposed "commas" or parentheses is often seen, and these components are occasionally connected end to end by thin, delicate, strandlike structures. Other cysts contain only a marginal nodule. Some authorities suggest that the intracystic bodies may be thickened portions of the cyst wall.

In an urgent clinical situation such as following tracheal aspiration or lung biopsy, the Gram–Weigert, Giemsa, or toluidine blue stains may be used for rapid diagnosis.[21] Nonetheless, the silver stain should always be carried out for confirmation, preferably with a positive and negative control (such as a slide with yeast forms and/or erythrocytes). Staining procedures vary, but a modification that produces a "rapid silver stain" (reliable results in a total of 2 hr) is used routinely in many laboratories and detailed in Table 1.

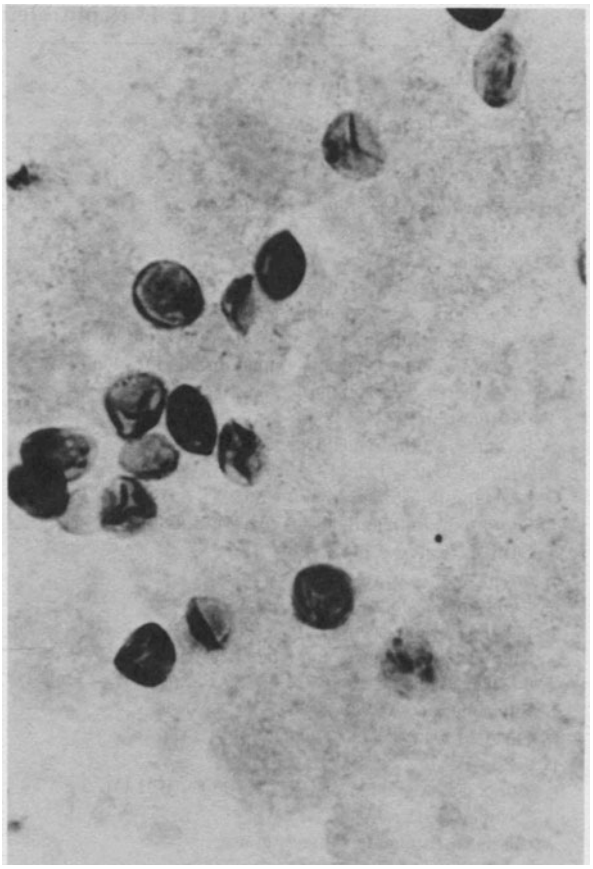


FIGURE 1. Gomori silver methenamine nitrate stain of impression smears of lung containing cysts of *Pneumocystis carinii*.

Electron microscopic evaluation reveals that the trophozoite is thin walled and has numerous evaginations of pseudopodlike projections called filopodia. These filopodia appear to anchor the organism to the alveolar septal wall and could explain the usual absence of pneumocysts in expectorated secretions. Although the smallest trophozoite form measures 1.5–2.0 μm in diameter, the cyst structure appears to be 3–5 μm in diameter. The mature cyst does not have filopodia but contains intracystic bodies measuring

TABLE 1. Rapid Methenamine Silver Stain

Solutions
- 5% Chromic acid
  - Chromic acid (CrO_3), 5 g
  - Distilled water, 100 ml
- 3% Methenamine
  - Hexamethylenetetramine, USP [$(CH_2)_6N_4$], 3 g
  - Distilled water, 100 ml
- Stock methenamine–silver nitrate
  - Silver nitrate, 5% solution, 5 ml
  - Methenamine, 3% solution, 1 dl
  - A white precipitate forms but immediately dissolves on shaking. Clear solutions remain usable for months at refrigerator temperature.
- 1% Sodium bisulfite
  - Sodium bisulfite ($NaHSO_3$), 1 g
  - Distilled water, 1 dl
- 2% Sodium thiosulfate (hypo)
  - Sodium thiosulfate ($NaS_2O_3 \cdot 5H_2O$), 2 g
  - Distilled water, 1 dl
- Working light green
  - Light green, stock solution, 10 ml
  - Distilled water, 40 ml
  - This solution is stable for 1 month.
- 5% Silver nitrate
  - Silver nitrate ($AgNO_3$), 5 g
  - Distilled water, 1 dl
- 5% Borax
  - Borax (photographic or USP, $Na_2B_4O_7 \cdot 10H_2O$), 5 g
  - Distilled water, 1 dl
- Working methenamine–silver nitrate
  - Borax, 5% solution, 2 ml
  - Distilled water, 25 ml
  - Mix, and add Methenamine–silver nitrate, stock solution, 25 ml
  - *Caution:* This preparation (working methenamine–silver nitrate solution) must be prepared fresh each time the stain is run. Do not try to reuse even for a stain run immediately following.
- 0 2% Gold chloride
  - Gold chloride, 1% solution ($AuCl_3 \cdot HCl \cdot 3H_2O$), 10 ml
  - Distilled water, 40 ml
  - This solution may be used repeatedly. However, when toning begins to fail and the organisms come out too black, it should be changed.
  - *Note:* 1% gold chloride solution is made from ampules and is diluted according to the directions accompanying it.
- Stock light green
  - Light green, S.F. (yellow), 0.2 g
  - Distilled water, 1 dl
  - Glacial acetic acid (CH_3COOH), 0 2 ml

Preparation of smear for methenamine–silver stain
- Material to be stained is smeared on a slide and allowed to air dry. Tissue may be ground and then smeared on a slide. Impression smears from cut tissue may be prepared.

Fixation
- The air-dried smears are placed in absolute methyl alcohol for 5 min Fix a control smear (fungus or *Pneumocystis*) at the same time.

Staining procedure
- While slides are fixing, fill one Coplin jar (with a lid) with 5% chromic acid In a screw-capped Coplin jar, prepare the working methenamine silver.
- Place fixed slides into chromic acid, and put both Coplin jars into a 48°C water bath. After 2 min, transfer the jar of chromic acid to a 56°C water bath for 10 min
- Wash the slides briefly in running tap water.
- Place the slides into 1% sodium bisulfite for 30 sec.
- Wash in running tap water for 15 sec
- Rinse slides in four changes of distilled water.
- Place the slides into the jar of working methenamine silver in the 48°C water bath for 2 min
- Transfer the jar to the 56° water bath for 25 min or less.
- Check control slide at 20 min to see if organisms have stained. To check control slide, remove control and patients slides to a water-filled Coplin jar. Leave the Coplin jar with the silver solution in the 56°C water bath. Coverslip control slide and quickly look for proper staining of the organisms: cytoplasms should be light brown with dark brown parentheses in the center. If so, proceed. If not, return the slides to the silver until they are stained.
- Rinse slides with four changes of distilled water
- Tone in 0.2% gold chloride for 1 min
- Rinse in 2 changes of distilled water
- Place slides into 2% sodium thiosulfate for 1 min.
- Wash slides in running tap water for 15 sec.
- Counter stain in working light green for 30 sec.
- Rinse in one change of distilled water.
- Dehydrate and clear for 1-min intervals in two changes each of 95% ethanol, 100% ethanol, and xylene
- Mount slides in Permount.

Interpretation
- Cyst walls are delicately stained brown or gray. Intracystic structures resembling "commas" or parentheses are often present and stain black. The cytoplasm is usually clear. Fungal structures, in contrast, are sharply delineated in black.

1–1.7 μm across and is enclosed by a double membrane that is partly connected to the inner layer of the cyst wall. Collapsed cysts are crescent shaped and are presumably the same forms seen by light microscopy of silver stain specimens.

From electron microscopic observations, a life cycle for *Pneumocystis carinii* has been postulated by Campbell[22]: the mature round cyst undergoes dissolution or "cracking" that permits escape of intracystic bodies. At that point, the intracystic bodies

resemble a small trophozoite form. It appears that small trophozoites evolve to larger forms, their walls thicken, and a precyst develops which is devoid of intracystic bodies. The cycle is completed with the arrival of the mature cyst stage containing a quota of approximately eight daughter cysts. An alternative developmental cycle proposes that daughter cells form as well within thin-walled pneumocysts (i.e., trophozoites).[23]

2.3. Histopathology

Although the pathology of *Pneumocystis* pneumonia has been well studied, there still appears to be considerable confusion regarding the typical histologic pattern. The original descriptive term for pneumocystosis, interstitial plasma cell pneumonia, dates back to the pronounced plasma cell infiltration of the interalveolar septa documented almost exclusively in the newborns studied in nursery outbreaks occurring in Europe.[6] The alveolar walls are distended many times their normal thickness with resultant compression of alveolar spaces. In immunocompromised patients, thickened alveolar septa and cellular infiltration are less marked than in the plasma cell pneumonia of European description. Although some degree of interstitial mononuclear (lymphocyte or macrophage) infiltration has been observed, hyperplasia of alveolar lining cells appears most responsible for septal thickening. (Septal cell hyperplasia is a nonspecific reaction to infections of various etiologies, and it is quite common among immunosuppressed patients who have no evidence of *Pneumocystis* infection per se.) The predominant finding on hematoxylin and eosin staining is an intense eosinophilic foamy or honeycombed material rather than a plasma cell infiltrate that fills the alveolar spaces. This intraalveolar material is composed largely of inflammatory cells and degenerating bodies of pneumocysts, the latter staining bright red with the periodic acid–Schiff (PAS) stain because of their high carbohydrate content. Typical cysts or trophozoite forms of the parasite are seen only after application of a silver or Giemsa stain, respectively.

In a study of patients primarily with acute childhood lymphoblastic leukemia, three sequential stages of pneumocystosis were categorized by Hughes et al.[24] Stage 1 consists of isolated cysts in the alveolar septal wall with few cysts in the alveoli and little or no inflammation. Stage 2 is characterized by the presence of cystic organisms within macrophages fixed to the alveolar wall and desquamation of these alveolar cells; trophozoite forms are found associated with cysts and also lying free in the alveolar space. There is only minimal septal inflammation at this time. The third and final stage is characterized by a reactive desquamative alveolitis with cysts appearing in alveolar macrophages in various states of degeneration. This so-called foamy exudate of pneumocystosis is neither edema fluid nor exudative inflammation but largely a coalescence of inflammatory cells (mainly macrophages) and organisms. It has been conjectured that spread of pneumocysts through pulmonary tissue does not occur by direct invasion through the interstitium or through vascular spaces. Rather, coughing may expectorate cysts from alveoli into larger airways, and organisms are then swept back into previously uninvolved alveolar areas. This hypothesis of intraairway transfer is supported by the usual finding of heaviest parasitic concentration in dependent portions of the lung.

Whether or not *Pneumocystis* infection can spread beyond the lung is a matter of controversy. Several cases of so-called generalized pneumocystosis in infants have been reported in which parasitemia and organ dissemination were demonstrable.[6] Although in some outbreaks, particles consistent with the sporozoite form have been seen in blood smears, the accuracy of these observations has been challenged. However, there have been a few well-documented cases of disseminated disease.[25–25b] Silver stain-positive cysts have been found to invade the bone marrow, splenic capsule, colon, liver, pancreas, retroperitoneal tissue, skin, and retina. Nonetheless, such cases appear to be exceedingly rare, and one series of some 200 patients had no instance of generalized infection.[26] In another study, using immunofluorescent techniques, no fragments or antigenic material derived from the cysts appeared in various sections of lymph nodes, liver, spleen, and kidney.[27]

2.4. Conditions Associated with Pneumocystosis in Humans and Animals

Table 2 summarizes the conditions associated with this disease in man and animals. Some of the earliest outbreaks of infantile disease in humans were associated with malnutrition and crowding. It is remarkable that even in the 1970s malnourished infants

TABLE 2. Pneumocystosis in Humans and Animals

Pneumocystosis in humans
Malnutrition (epidemics)
Crowding (epidemics)
Immunodeficiency
Primary
Neoplasia plus therapy
Transplantation-associated
Acquired, secondary to retrovirus infection
Drug related (cortisone, cyclosporine, antithymocyte globulin)
Pneumocystosis in Animals
Natural
Rats, mice, guinea pigs, rabbits, dogs, monkey, sheep, goats, horses
Altered state
Cortisone treatment
Athymic (nude) mice

brought to the United States were found to have sporadic cases of *Pneumocystis* infection.[28] However, the major association is clearly with compromised host defenses of either a primary or acquired nature: congenital immunodeficiency, neoplasia, cancer chemotherapy, immunosuppression for collagen vascular disease, and corticosteroids or antithymocyte globulin given to prevent graft rejection. The nature of immunosuppressive therapy can be quite variable, but corticosteroids are the most consistent component of drug regimens associated with this infection. For instance, we have seen pneumocystosis complicating steroid treatment given for cutaneous inflammatory disorders such as pemphigus vulgaris or secondary to attempted reduction of brain edema with dexamethasone. On the other hand, patients with severe asthmatic disorders rarely develop this complication in spite of the tendency to use increasingly larger doses of cortisone to treat status asthmaticus. Perhaps the otherwise intact nature of host defenses in asthmatic states, the tendency to give alternate-day therapy in asthma, or the fact that relatively low doses of cortisone or its congeners are used may explain the lack of an association.

2.5. Predisposing Factors and Host-Defense Mechanisms

A precise appraisal of the relative importance of the roles of specific components of host defenses active against *Pneumocystis* infection cannot yet be made. Although the disease has been reported rarely in normal persons,[29] pneumocystosis usually emerges in settings of both impaired lymphocyte and humoral antibody function. That susceptibility to *P. carinii* is related to a disorder of T-lymphocyte-mediated immune mechanisms is supported by the findings that overt disease is inducible in previously normal rabbits or rodents after conditioning with corticosteroids or protein-calorie deprivation[12,13,30] and that infection is transmissible from both experimentally infected rats and human lung material to nude or athymic mice without corticosteroid conditioning.[31] The pattern of clinical disease in man suggests a T-lymphocyte abnormality inasmuch as the three major disorders associated with pneumocystosis are Hodgkin disease, lymphatic leukemia, and AIDS. Hodgkin disease is the lymphoma most often associated with pneumocystosis,[32] although it should be recognized that this association is well documented only in the modern chemotherapeutic era when corticosteroids are important components of chemotherapy for this underlying disease. Of the leukemias, pneumocystosis is most common in lymphatic neoplasms, particularly the acute childhood type (for which steroids are now invariably employed), but it is uncommon in myelogenous leukemia unless corticosteroids are used. Recently, the use of cyclosporine has been associated with increased risk of clinically significant *Pneumocystis* infection.

There is, however, some evidence that *P. carinii* infection is secondary to a humoral immune disorder as well as a cellular abnormality. From in vitro studies of pneumocyst–phagocytic cell interactions,[33] immune serum has been shown to enhance the interiorization of cyst forms by alveolar macrophages. Among the earliest reported cases of human disease are examples of patients with hypogammaglobulinemia or agammaglobulinemia. A review by the group at the Centers for Disease Control (CDC) of the association between pneumocystosis and primary immunodeficiency syndromes showed that infected patients had decreased serum immunoglobulins, impaired antibody synthesis, or both.[34]

2.6. Epidemiology and Transmission

Knowledge of the epidemiology and transmission of *Pneumocystis* infection is based on careful

studies of experimentally infected animals and limited clinical and serologic investigations in humans. Naturally occurring epizootics of *Pneumocystis* infection have been documented in a colony of nude mice,[35] a finding again emphasizing the importance of T-lymphocyte-mediated immunity. Other studies demonstrate transmission of disease in experimental animals previously treated with cortisone. Hendley and Weller employed a model using steroid-treated rats obtained by cesarean section that were originally barrier sustained.[36] When exposed to an air supply from standard infected rats where the disease was engendered by cortisone treatment, evidence for airborne transmission was obtained. Airborne spread to non-steroid-treated nude mice was documented by Walzer and colleagues.[31] The evidence in humans in favor of a contagious process derives from the initial observations of epidemic disease that occurred in nurseries and foundling homes in central Europe and Iran.[6] In addition, there have been scattered reports of *Pneumocystis* infection occurring in roommates[37] and in family members.[38] In the United States, there have been three well-studied episodes of institutional outbreaks that occurred in cancer treatment centers. The largest of these studies, carried out at St. Jude Children's Cancer Research Hospital by Perera et al.[39] failed to confirm patient-to-patient spread. It was the conclusion of these investigators that the disease in most cases arose independently from reactivation of latent infection triggered by intensive antineoplastic drug protocols. By contrast, however, Singer et al. reported, in 1975, a small cluster of 11 cases that occurred over a 3-month interval at Memorial Sloan-Kettering Cancer Center.[40] The epidemiologic investigation, which included serologic studies of patients and hospital personnel, suggested interpersonal spread at the institution. A third outbreak occurring among childhood leukemia patients treated at the Riley Hospital in Indianapolis, Indiana involved 11 cases over 10-month period.[41] Here too, serological studies suggested involvement of nursing and medical personnel either as cases of inapparent infection or as actual carriers of disease.

Our concept of the epidemiology of *Pneumocystis* infection therefore encompasses the original postulates of Meuwissen et al.[42] and Hughes [43] that the infection is a common occurrence among young children. If individuals later become intensely immunosuppressed, the infection may become reactivated. Some normal adults may not have had childhood infection, experience mild or inapparent infections when exposed, and "contain" the process because of intact host defenses. These individuals, however, might be healthy nursing or medical personnel who demonstrate evidence of asymptomatic seroconversion. Still, they could transmit the disease to high-risk patients within the hospital environment. On the other hand, such instances are probably much less common than reactivation of latent infection, i.e., person-to-person spread is usually unlikely.

2.7. Clinical Features (Non-AIDS Related)

Clinical and epidemiologic information concerning *Pneumocystis carinii* infection has been derived from two excellent sources in the United States: (1) analyses of cases whose treatment was evaluated by the CDC[32,44] and (2) comprehensive longitudinal studies of children with neoplasms treated at the St. Jude Children's Cancer Research Hospital in Memphis, Tennessee.[5,45] The information is largely complementary, but the findings of these two groups must be evaluated in the context of the sources of the data. With the CDC assessments, the data base consists of report forms returned to the Parasitic Diseases Drug Service during the period when pentamidine, solely distributed by CDC, was the principal mode of treatment. The epidemiologic data from the CDC are valuable in discerning major trends. By drawing on a large sample of cases, the pitfalls of relying on information from a few major institutions are avoided. However, in such a series there could have been a systematic bias toward inclusion of patients in whom appropriate diagnostic procedures were attempted, such as those treated in large university-type referral centers. The St. Jude's experience benefits from having a well-defined population of children with malignancy that has been observed by excellent clinicians over a long period of time. Such a study proves highly reliable information on specific risk factors for pneumocystosis.

From the CDC data, the following observations may be made. The attack rate of pneumocystosis is highest in the age group less than 1 year (almost all affected patients in this group have a primary immunodeficiency) and progressively declines with advancing age.[46] Viewed in terms of numbers of patients, of some 194 patients with histologically

confirmed *Pneumocystis* pneumonia, leukemia was the most common underlying disease and was present in 91 of the 194 cases. Of these, approximately two-thirds or 58 out of 91 were of the acute lymphatic type, and 18 of the 91 were of the chronic lymphatic type. Hodgkin disease and other lymphomas were the second largest group of underlying disease, followed by primary immunologic deficiency diseases. Recipients of organ transplants were the fourth largest group of patients. It was possible to calculate the attack rate of pneumocystosis for the different leukemias: 1.1% per year for acute lymphatic disease, 0.05% per year for chronic lymphatic disease, and 0.2% per year for acute myelocytic disease.[46] However, if these rates are expressed in terms of a lifetime risk, it is likely that patients with lymphatic leukemia, who now have a considerably longer survival than those with myelocytic leukemia, are at even greater risk of contracting pneumocystosis. Indeed, it is rare in our experience to encounter a case of pneumocystosis complicating myelocytic leukemia unless the patient has been given corticosteroid therapy. Since corticosteroids are not routinely employed in many of the common chemotherapeutic induction regimens for acute myelocytic leukemia, those relatively few cases of pneumocystosis in the latter group of patients may be more related to steroid therapy rather than to the leukemia per se.

In the CDC analysis, the most common symptom of *Pneumocystis* pneumonia was dyspnea, which was observed in 91% of patients (Table 3). Fever was present in two-thirds of patients, and cough was present in one-half. It is important to recognize that only 7% of patients had productive cough, and only two patients had hemoptysis. In the great majority of children, tachypnea with respiratory rate exceeding 40 per minute was present as well as a weak dry cough. Furthermore, an absence of characteristic rales has been occasionally noted even when severe pneumonitis is present radiographically. One of the more remarkable observations in the St. Jude's pediatric population was the finding that 21% of documented *Pneumocystis*-infected patients had mild to moderate diarrhea at the onset or during the course of pneumonitis.[45] Whether this finding might have resulted from antimicrobial therapy is not known.

The duration of symptoms in 153 patients with confirmed pneumocystosis in the CDC analysis ranged from less than 7 to greater than 90 days, with a median of approximately 12 days. The widespread belief that pneumocystosis is an acute fulminant pneumonitis may be, in part, a reflection of delayed diagnosis or at least dilatory management until the patient has deteriorated to the point where diagnostic measures became a medical emergency.

TABLE 3. Clinical Features of 168 Patients with Histologically Confirmed *Pneumocystis* Pneumonia[a]

Clinical features	Number of patients	Percent
Symptoms		
Dyspnea	152	91
Fever	110	66
Cough	79	47
Productive cough	12	7
Hemoptysis	3	2
Chest pain	11	7
Night sweats	1	1
Signs (respiratory)		
Cyanosis	66	39
Rales	56	33
Breath sounds		
Decreased	22	13
Bronchial/tubular	14	8
Dullness	9	5
Rhonchi	7	4
Wheezing	2	1
Signs (other)		
Hepatomegaly	59	35
Splenomegaly	32	19
Radiographic findings		
Infiltrate		
Diffuse and bilateral	164	98
Unilateral	4	2
Effusion	8	5
Adventitious air[b]	6	4

[a]From Walzer et al [32]
[b]Pneumothorax, pneumomediastinum, etc

Illustrative Case 1

The patient was an 8-year-old white girl with a 1½-year history of acute lymphatic leukemia. After induction of remission with vincristine and prednisone, she relapsed and was successfully reinduced with vincristine, L-asparginase, prednisone, systemic methotrexate, and intrathecal methotrexate. She also received craniospinal radiation and 6-mercaptopurine. A month prior to this admission, she was admitted to the pediatric service of the hospital and given ampicillin 100 mg/kg IV q4h because of the 3-day

history of cough, fever, and vomiting. A chest radiograph showed a right middle lobe infiltrate Sputum grew *Streptococcus viridans, Neisseria* species, *Staphylococcus edpidermidis,* a few yeastlike organisms, and a few *Hemophilus* species. However, the nature of the respiratory pathogen was not identified, and after 4 days in the hospital the patient was sent home on a gram of ampicillin a day. For the 2 weeks prior to admission, she had a productive cough, a rapid respiratory rate, decreased appetite, lethargy, fever to 101°F (38.3°C), but no cyanosis or dyspnea. Her prednisone dose (40 mg/day) was tapered, but admission was prompted by a progressive right middle lobe and newly documented left lower lobe infiltrate (Fig. 2A). Physical examination revealed a tachypneic, irritable young white girl with a blood pressure of 120/50 mm Hg, respiratory rate 40, pulse 140, temperature 100.4°F (38°C). Physical examination of the lungs revealed diffuse inspiratory wheezes and rales greater on the left than on the right and greater at the bases than the apices. Complete blood count (CBC) revealed a hemoglobin (Hb) of 12.2 mg/dl hematocrit (Hct) 35%, white blood cell (WBC) 1600 with 68% segmented forms, 4% band forms, 18% lymphocytes, 10% mononuclear cells, and 140,000 platelets. A cold agglutinin titer was negative and a venous blood gas showed a pH 7.45 P_{CO_2} 40 mm Hg, P_{O_2} 33 mm Hg, with 66% saturation.

The patient was started on carbenicillin, gentamicin, oxacillin, trimethoprim–sulfamethoxazole, and erythromycin. The trimethoprim–sulfamethoxazole was initially given orally. Her respiratory status continued to deteriorate over the next several days, and she was given 35% O_2 by mask. She also developed a vesicular rash in the distribution of T 7–8 dermatone consistent with herpes zoster, and adenine arabinoside was started. At this point, she was taken to the operating room where under general anesthesia an open-wedge biopsy of the left lower lobe was performed. *Pneumocystis carinii* was detected in the lung biopsy, and the patient was started on intravenous trimethoprim–sulfamethoxazole. Blood levels of trimethoprim exceeded 5 μg/ml, 1 hr postinfusion. Varicella pneumonia was not excluded at the time, but subsequently cultures of lung tissue were negative for varicella. Thereafter, the patient showed gradual clinical improvement, although she demonstrated evidence of the inappropriate ADH syndrome. Her arterial blood P_{O_2} gradually rose from 55 mm Hg to 90 mm Hg on room air. A total of 14 days of intravenous trimethoprim–sulfamethoxazole was given, and the chest radiograph returned to normal over the next 4 weeks (Fig. 2B).

Comment. This case illustrates many of the typical findings of *Pneumocystis* infection complicating childhood leukemia: (1) the disease occurs during bone marrow remission, often when steroids are being tapered; (2) the duration of respiratory symptoms (particularly tachypnea) can be weeks, (3) there can be concurrent infections (in lung and other sites) like herpes zoster, (4) but appropriate therapy will result in complete radiologic resolution Furthermore, this case illustrates the problems with orally administered, empirical trimethoprim–sulfamethoxazole. Absorption may be incomplete, and the question of drug failure versus inadequate dosage remains. An aggressive diagnostic approach led to identification of the pulmonary pathogen (it was not varicella) and a more reliable therapeutic approach; intravenous trimethoprim–sulfamethoxazole was then used. Clearly, we do not condone the delay in undertaking a dignostic procedure as occurred in this case (see Section 2 9), nor do we advocate empirical therapy in patients able to undergo a biopsy procedure. It is fortunate that residual evidence of *Pneumocystis* infection was present in the biopsy specimen.

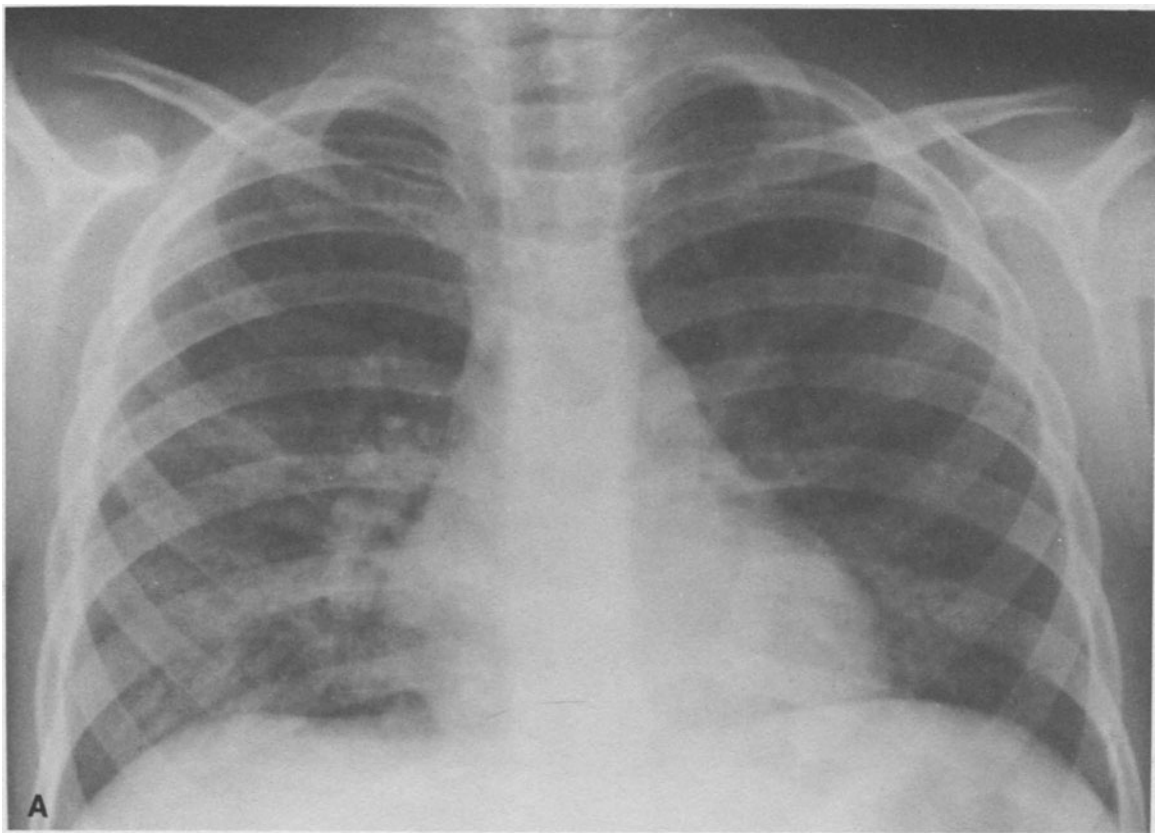
Relevant to our case is that one-fourth of the CDC-analyzed patients with confirmed pneumocystosis had another documented infection.[46] The simultaneous occurrence of cytomegalovirus infection with *Pneumocystis* pneumonia has been particularly common in recipients of organ transplants.[47]

An interesting comparison and contrast between the incidence of varicella–zoster and *Pneumocystis carinii* pneumonia in leukemic children has also been reported.[48] Whereas one-quarter of patients given the most intense chemotherapy regimen developed pneumocystosis, there was really no increased risk of varicella–zoster virus (VZV) infection. Twenty percent of patients who failed to achieve remission had VZV infections, and 30% in this category also developed pneumocystosis.

In related studies, Hughes and collaborators documented the risk of *Pneumocystis* pneumonia in relation to the phase and type of antileukemic therapy.[48] During a phase when treatment consisted of prednisone, vincristine, and L-asparginase over a 4-week cycle, no patient out of a total of 149 developed pneumocystosis During a succeeding phase that included intrathecal methotrexate and central nervous system (CNS) irradiation, this same group of 149 patients experienced a 4% incidence of pneumocystosis. Subsequent attempts at consolidation of remission over a 2–3-year period involved randomization into four treatment groups. Those patients maintained on methotrexate or methotrexate plus 6-mercaptopurine and cyclophosphamide experienced about a 5% incidence of pneumocystosis. By contrast, when cytosine arabinoside was added to methotrexate, 6-mercaptopurine, and cyclophosphamide, 10 of 41 children, or 22.4%, developed *Pneumocystis carinii* pneumonia The incidence of *Pneumocystis* in acute lymphocytic leukemia thus appeared to be a reflection of the intensity of chemotherapy. Extent of the neoplastic disease and mediastinal irradiation could have been additive factors.

2.8. Radiologic Findings

The typical radiologic pattern in biopsy-proven pneumocystosis, observed in almost 100% of the CDC-reported cases, is a diffuse bilateral alveolar infiltrate.[32] Only 5% of patients in the CDC series were found to have a pleural effusion. Consistent with this experience, 78 out of the 80 patients studied at St. Jude had diffuse alveolar disease, and the two patients who had normal chest radiographs died within 48 hr, at which time pneumocysts were found in the lung at autopsy.[45] Several interesting reports suggest that the appearance of pulmonary infiltrates may be delayed and lag behind the abrupt onset of dyspnea.[49,50] Thus, dyspnea alone could be an important clinical clue in patients at high risk of developing pneumocystosis. Persons who suddenly experience tachypnea and are found to be hypoxic even in the absence of pulmonary infiltrates should be carefully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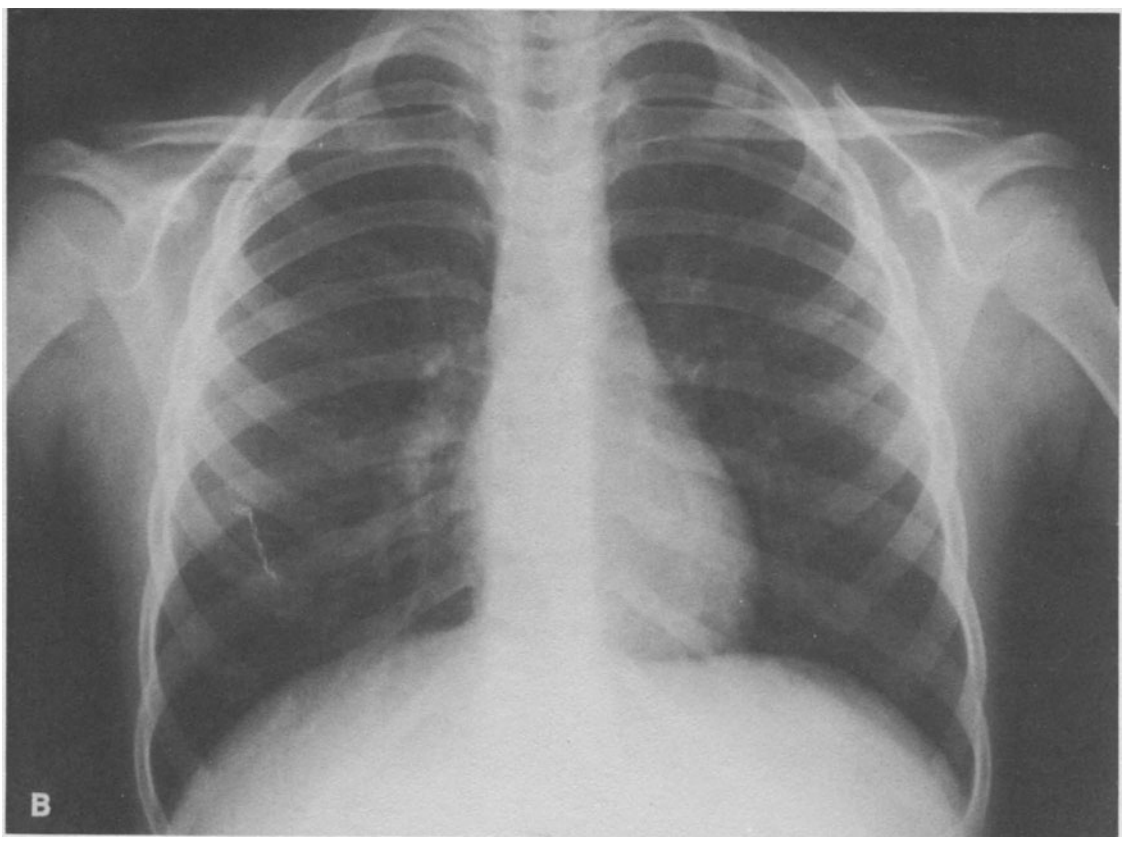
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evaluated and have daily chest radiographs, because the infiltrates may appear later. Furthermore, there are several reports that a lung scan, particularly with gallium,[50,51] may identify lung involvement prior to the radiograph (but is not specific for pneumocystosis). In several reported cases, the chest radiograph has remained normal despite severe dyspnea and hypoxemia.[52]

Another study of the radiologic features of *Pneumocystis carinii* pneumonia involved 30 patients with confirmed disease.[53] It was emphasized that the typical radiographic picture is an acute perihilar and basilar infiltrate progressing to diffuse alveolar consolidation within 3–5 days, usually unassociated with hilar adenopathy or pleural changes. However, in this study the incidence of atypical radiographic findings was appreciable, with 17 of 30 patients showing at least one atypical finding. Such findings included sparing of the apices of the lung, unilateral appearance of the infiltrate, lobar or segmental consolidation (in addition to diffuse involvement), and a pseudonodular pattern that could be confused with metastatic processes. Additional reports document that pneumocystosis may even cause "coin" lesions[54] or pneumatoceles.[55] Although pleural effusion is exceedingly unusual in pneumocystosis, this observation must be tempered by the recognition that pleural effusions can be a part of an underlying disease such as neoplasm or collagen vascular disorder that predisposes to *Pneumocystis* infection. Similarly, a finding of a mediastinal mass does not militate against the diagnosis of *Pneumocystis* infection of the lung. Although it is common to attribute a mediastinal mass to progression of underlying disease (lymphoma or leukemia), 43% of patients with mediastinal masses in the St. Jude Hospital series had *Pneumocystis* infection.[48] In summary, therefore, no radiologic finding per se completely excludes the diagnosis of *Pneumocystis*.

2.9. Diagnostic Approaches to Suspected *Pneumocystis carinii* Infection

The diagnosis of pneumocystosis still involves controversial issues: (1) What are the indications and value of attempting an invasive procedure to diagnose *Pneumocystis* infection? (2) Should patients with suspected infection initially receive an empirical trial of antimicrobial therapy that will "cover" this organism (as in the case illustrated)? (3) What diagnostic procedure is best or most expedient in different hosts? Much time is spent at the bedside or on ward rounds in debating these issues. Our working principles are that every reasonable attempt should be made to establish a specific diagnosis and that empirical therapy should be avoided if possible. Furthermore, we believe that open lung biopsy still represents the most reliable means of confirming diagnosis but is unquestionably the most invasive procedure. Patients with *Pneumocystis* infection are often critically ill and, in addition to being tachypneic and hypoxic, often are receiving antiinflammatory agents that impair wound healing. Some may have a marked bleeding diathesis. Obviously diagnostic procedures such as thoractomy or transbronchial biopsy may aggravate hypoxia and add to the risk of hemorrhage. Whether there can be compromises in the diagnostic approaches, i.e., less invasive procedures of equal sensitivity and specificity, is an issue that still remains unresolved.

The major argument in the past in favor of making a specific diagnosis was based on the observation that almost 50% of patients treated with pentamidine experienced a significant untoward reaction.[32] Thus, a proven diagnosis made pentamidine treatment a justifiable hazard, whereas a negative biopsy avoided toxic empirical therapy. With the availability of trimethoprim–sulfamethoxazole, an alternative argument has been raised: namely, that therapy is relatively nontoxic (at least in non-AIDS patients) and all clinically suspected cases should be given a therapeutic trial. If the patient improves, a possibly dangerous diagnostic procedure is averted. Walzer et al.[32] have no quarrel with this argument, nor do they object to the empirical use of trimethoprim–sulfamethoxazole in the patient in whom it is not possible, for good reasons, to attempt an invasive diagnostic procedure. On the other hand, trimethoprim–sulfamethoxazole is not specific for *Pneumocystis carinii*, and clinical improvement cannot be ascribed to the specific therapy directed against this protozoan parasite. That would be of little practical concern to

FIGURE 2. (A) Chest radiograph taken immediately prior to lung biopsy of Illustrative Case 1, subsequently proven to have pneumocystosis (B) Chest radiograph following successful completion of course of intravenous trimethoprim–sulfamethoxazole.

the improving patient, but the dilemma begins if the patient does not improve.

It has been amply demonstrated in a number of series, perhaps the best of which is from the National Cancer Institute, that *Pneumocystis* accounts for probably no more than one-third of the etiologies of diffuse pulmonary infiltrates.[56] To treat empirically with trimethoprim–sulfamethoxazole alone would delay the chance of identifying other treatable diseases (tuberculosis, aspergillosis, cryptococcosis, or interstitial pneumonitis secondary to anticancer drugs) where different therapy would obviously be indicated. The recent recognition of new, potentially treatable pulmonary infections is an important reminder of the value of making a specific diagnosis. Moreover, if a diagnostic procedure is carried out after a course of empirical therapy has been initiated, it may not be possible to identify the original provocative agent. The clinician is then left to deliberate if the initial therapy were correct, whether the patient might harbor a strain morphologically altered by therapy but resistant to the drugs given, or whether cysts were missed simply because of sampling error. Thus, early empirical therapy with trimethoprim–sulfamethoxazole may obfuscate the nature of underlying infection (whether caused by *Pneumocystis* or by other microbes); if a biopsy is obtained at a later point, the quality of the information derived is likely to be less reliable.

Perhaps a more trenchant question is whether benefit always accrues to the patient in whom a specific diagnosis of infection is made by lung biopsy. From an intellectual and protocol-oriented view, there is no argument about the desirability of determining the etiology of a patient's pulmonary infection. Unfortunately, some of the better retrospective studies of open lung biopsy have failed to discern a difference in mortality between patients who had a specific diagnosis made and those who did not or between those whose treatment was altered because of diagnosis and those in whom the results of biopsy did not affect treatment.[57] Yet, it is not possible to conclude from such a study that open lung biopsy is completely without value, since patients with specific diagnoses may constitute a higher risk group whose mortality rate might even be higher if they were undiagnosed or inappropriately treated. In fact, other studies[58,59] have reported lower mortality rates for patients who had a specific diagnosis established than for those who did not. Thus, a definite conclusion about the value of establishing a diagnosis can be made only in patients with a specific diagnosis prospectively randomized into treatment and nontreatment groups, and it is doubtful that such a study will ever be carried out.

Since survival statistics for immunosuppressed patients who develop pulmonary infiltrates are depressingly poor and mortality rates of many of the underlying diseases are also substantial, it is only reasonable to adopt an approach to the diagnosis of pulmonary pathology that is tempered by clinical circumstances. In patients who have failed to respond to conventional therapy and in whom the chances of experimental therapy working are bleak (refractory neoplasm), empirical anti-*Pneumocystis* therapy may be justified. We also favor empirical therapy if the patient has an uncorrectable bleeding diathesis that precludes invasive diagnostic measures. By contrast, aggressive diagnostic approaches are clearly indicated in dealing with new pulmonary infiltrates in young patients whose leukemias or lymphomas may be in remission.

An important question is whether nonspecific polypharmacy might lead to better patient survival and cost effectiveness than diagnostic open biopsy. It can be argued that by empirical use of a number of antimicrobial agents, most of the infectious problems found in immunosuppressed patients could be treated. The reservations about this approach include the following: (1) patients with undiagnosed neoplastic or drug-induced pulmonary infiltrates would not be correctly treated; (2) multiple drugs may interact in ways to potentiate organ toxicity (e.g., nephrotoxicity) or in ways we cannot anticipate; (3) dosage of empirical therapy may not be adequate or "pushed" with the confidence that exists when a diagnosis is established. In one study, two-thirds of patients having an underlying malignancy were found to have the same neoplastic process responsible for pulmonary infiltrates. Hospital mortality rate was only 8%,[60] emphasizing the importance of directed radiation/chemotherapy in these patients. Thus, a specific diagnosis is highly desirable whenever there is a strong suspicion of disseminated malignancy.

Fully 40% of the patients in Hughes's trial of trimethoprim–sulfamethoxazole versus pentamidine did not initially respond to the agent of first choice, and the total of 80% beneficial responses included

approximately 20% of the patients who were crossed over to the alternative regimen.[49] Whether these patients would have improved if they had been maintained on the initial agent selected is open to question. We have certainly seen patients who did not respond to pentamidine for up to 8 days when pentamidine was the only parenteral agent that could be used for treatment of *Pneumocystis* infection (see Section 2.10). An even more important question, however, is whether or not there may be differing susceptibility of *Pneumocystis* strains for the different antimicrobial agents that are available at this time. Without establishing a specific diagnosis, particularly of *Pneumocystis*, empirical therapy may thus miss those patients who could be failing on one regimen and could potentially benefit by a switch to the other or in whom the dose of one of the agents could be increased.

Two studies carried out in oncology centers[61,62] document the superiority of open lung biopsy over less invasive diagnostic procedures: simultaneous sampling was performed in each of these studies and, not surprisingly, the yield of specific diagnoses was greatest with open lung biopsy. Unfortunately, nonspecific or idiopathic pneumonitis was found in a disturbingly large proportion of cases in both series. In the final analysis, local experience and expertise should be the basis for selection of a diagnostic procedure. The yield from examination of expectorated sputum is low except in patients with AIDS, in whom the number of cyst forms appears to be high.[63]

Transtracheal aspiration in certain selected cases has readily yielded the diagnosis and is an approach that we would favor providing the patient's platelet count were in excess of 40,000 mm^3.[64] We do not advocate transtracheal aspiration solely as a means for making a diagnosis of pneumocystosis. It is recommended as a valuable means of diagnosing infectious pulmonary disease and has been particularly helpful in documenting nosocomial gram-negative bacillary and anaerobic lung infection.[65] In fact, in our hands, the successful identification of *Pneumocystis* by transtracheal aspiration has been a dividend of attempts to diagnose tuberculosis or other bacterial infection in the lung. Transtracheal aspiration may also be attempted as a prelude to fiberoptic bronchoscopy. Transbronchial biopsy or fiberoptic bronchoscopy with brushing are now readily available in a number of centers, but a major problem with endoscopic techniques is that material insufficient for all of the desired microbiologic as well as histopathologic studies is often obtained.

In some centers, particularly those oriented to pediatrics, percutaneous thoracic closed needle aspiration under fluoroscopic guidance has been carried out, usually without complication.[66] Perhaps in young children the lung seems to "seal" better after puncture, but our experience with needle aspiration or biopsy in the adult has not been so salutary. Indeed, puncture of the lung in an adult with a bleeding diathesis or thrombocytopenia may lead to both pneumothorax and uncontrolled bleeding.

Our recommendation is that transtracheal aspiration and either fiberoptic bronchoscopy with brushing or transbronchial biopsy be performed initially, but neither of the latter two techniques reliably diagnoses aerobic and anaerobic bacterial infection, hence the added value of transtracheal aspiration (TTA).

A variation of the bronchoscopic procedure is bronchoalveolar lavage, where the catheter is wedged into a bronchus and a fairly large amount of saline (200 ml) rapidly introduced and reaspirated.[67] The recovered fluid is concentrated for culture and examination, using such tools as monoclonal antibody staining for herpes viruses. While these procedures are being planned and executed, we see no harm in giving an IV dose of furosemide (100 mg) to those patients in whom congestive heart failure has not been excluded with a Swan Ganz type of pulmonary outflow catheter. This can lead to dramatic relief of dyspnea within hours and, coupled with radiologic improvement, might lead to postponement of more invasive procedures. If TTA and bronchoscopic procedures do not yield a diagnosis, we would unhesitatingly proceed to open lung biopsy. Thus, the basic approach must be a commitment to an escalating tempo of diagnostic evaluation. If one procedure fails to lead to a specific diagnosis, the next step should be undertaken immediately without delay.

The diagnostic evaluation does not end with the surgical procedure. It does no good to carry out an open lung biopsy on an emergency basis if the proper services within the hospital are not mobilized to process such specimens rapidly. Bacteriologic, mycologic, and acid-fast studies are mandatory, but culture of the specimen for anaerobes and viruses is also clearly indicated. We believe that rapid methods

for staining cysts or trophozoites—Giemsa, toluidine blue, and Gram–Weigert stains—are valuable methods for making the diagnosis, but a definitive diagnosis rests on the identification of material by silver stain. Permanently fixed tissue sections take longer to process than impression smears of biopsy specimens, but silver stains of both materials should be performed using procedures such as those outlined in Table 1.

Some success has been reported in skilled and experienced hands by using an immunofluorescent technique for the detection of *Pneumocystis* in sputum or tracheal aspirates.[68] Monoclonal antibodies specific for *Pneumocystis* have been developed and these may be of diagnostic value.[69]

Serologic tests have been evaluated for more than a decade both in Europe and in the United States.[42,70] The value of a complement fixation test reported by some European workers has not been borne out when subject to critical scrutiny. An indirect immunofluorescence test for detecting circulating antibody using cysts prepared from either human lung or from animals has been reported to be positive in perhaps one-third of cases.[71] We believe, in fact, that the sensitivity and specificity of the test can be increased, but in our hands no more than two-thirds of patients are positive at time of presentation. If serologic tests are positive, they are suggestive of *Pneumocystis* infection. This may be an interesting way to recoup the diagnosis in patients who were treated empirically and whose serial antibody titers might be followed along the course of recovery.

Preliminary efforts at cultivating the parasite have been reported by several groups, but these techniques cannot be applied for diagnostic purposes.[4] In one report, cyst antigen harvested from in vitro passage has been used to produce antiserum which has been used in countercurrent immunoelectrophoretic tests to detect circulating *Pneumocystis* antigen. However, a positive test result seems more suggestive of pneumonitis rather than *Pneumocystis* infection.[72] Considerable debate has arisen regarding such antibody tests[73,74] and at present they cannot be considered of any diagnostic value.

2.10. Treatment

There are three regimens that have proven effective for therapy of *Pneumocystis* infection: pentamidine isethionate, the fixed combination of trimethoprim–sulfamethoxazole, and the combination of pyrimethamine and a sulfonamide. A variation of the latter two regimens is to substitute the sulfonamide with a sulfone such as diaminodiphenyl sulfone (Dapsone). Clinical experience has been greatest with pentamidine and trimethoprim–sulfamethoxazole. The numbers of cases treated with pyrimethamine and sulfadiazine is small, and no studies have been carried out to compare the latter regimen with trimethoprim–sulfamethoxazole. Pentamidine and trimethoprim–sulfamethoxazole have been evaluated in a comparative manner in at least one study in leukemics and found to be equivalent.[49] Pentamidine has been compared with pyrimethamine-sulfadiazine in a very small series where the preliminary results were similar. However, pyrimethamine–sulfadiazine was difficult to administer in severely ill patients.[76] No information is available on the relative effectiveness of the individual components of the two-drug combinations, that is, whether trimethoprim or pyrimethamine alone might be effective in the treatment of pneumocystosis. The latter question could be clinically important in the patient who has well-documented sulfonamide allergy. Many other agents have been tried over the years, including *p*-aminosalicylic acid, isoniazid, and amphtericin B, but no convincing evidence for their effectiveness has been presented. Another folate sulfonamide combination, pyrimethamine–sulfadoxine (Fansidar), is a long-acting combination that was effective prophylactically in studies carried out in Iran.[77]

Despite the availability of clinically effective agents, important questions still remain about the use of any of the three acceptable therapeutic approaches. One issue is that of pharmacokinetics and of the adequacy of some of the presently recommended dosage regimens. The second consists of potential cumulative toxicity when agents such as pentamidine are used with other nephrotoxic compounds such as aminoglycosides and amphotericin. Because of inability to achieve long-term cultivation of the causative agent, questions about drug resistance remain as yet unanswered. Since the folate antagonists bind dihydrofolate reductase, the site of action for some anticancer drugs such as methotrexate, treatment or prophylaxis with folate antagonists could potentiate chemotherapy aimed at an underlying neoplasm. An unanswered issue in man is

whether or not two or more of the effective regimens, e.g., pentamidine and trimethoprim–sulfamethoxazole, might be better than one alone. (There is no support for this in experimental animal studies.[78]) Finally, important bedside questions are duration of therapy and the clinical guidelines for adhering to one form of treatment before switching to alternative treatment.

2.10.1. Pentamidine

Pentamidine isethionate (Lomidine), 4-4′-diamidinodiphenoxypentane di-β-hydroxyethanesulfonate, is a diamidine compound with antiprotozoal and antifungal activity. Originally, this agent was synthesized as a hypoglycemic agent, and the problem of hypoglycemia persists as a potential complication of pentamidine therapy. Prior to the availability of pentamidine, the mortality from *Pneumocystis* pneumonitis was approximately 50% in the pediatric cases observed during European epidemics and close to 100% in immunodeficient children and adults.[76,77] In 1958, Ivady and Paldy first reported that pentamidine therapy of infantile *Pneumocystis* infection lowered mortality from 50% to 3%.[10] Several hundred cases of the epidemic disease in nurseries in Hungary were treated successfully with dose of 4 mg/day of pentamidine IM for 12–14 days. Clinical responses were usually apparent 4–6 days after the initiation of treatment, but radiographic improvement was often delayed for several weeks. Other investigators in Europe have reported comparable results in smaller series of children.

Pharmacologic effects of pentamidine vary depending on the route of administration.[75] In animals, a precipitous transitory fall in blood pressure has been noted immediately after injection, and renal toxicity has followed repeated administration. To avoid immediate toxic reactions associated with intravenous administration, IM injection is preferred, but in markedly thrombocytopenic patients, consideration must still be given to the intravenous route. Some evidence exists that pentamidine inhibits dihydrofolate reductase in tissues. In addition, pentamidine interacts and forms water-insoluble products with specific nucleotides and nucleic acids. Alternatively, it has been proposed that pentamidine acts through interference with aerobic glycolysis. There is no question that megaloblastosis of the bone marrow has been reported in patients treated with pentamidine, and this is paralleled by lower serum folate levels. Nonetheless, it has not been shown that pretreatment with folinic acid influences the therapeutic efficacy of pentamidine in rats infected with *Pneumocystis*.

Administration and Dosage. Each suspension of pentamidine isethionate must be freshly prepared with sterile distilled water. Under no circumstances should normal saline be used as a diluent because of the insolubility of the drug in such material. Pentamidine must be given parenterally because it is poorly absorbed via the oral route.

The dose for *Pneumocystis carinii* peneumonia is 4 mg/kg given IM once a day for 12–14 days, but in some circumstances (patients with AIDS) therapy may have to be continued for more than 2 weeks. For large adults (>80 kg), doses in excess of 200 mg may be considered.

The calculated daily dose should be dissolved in no more than 3 ml sterile distilled water in order to reduce the volume of injection. Solutions showing turbidity or slight crystalline remnants are still suitable for intramuscular but not intravenous injections. Such solutions seem to possess no more toxicity or loss of activity. However, it should be borne in mind that sterile abscesses can result from too large a volume of injection and from irritation from the drug itself. In situations in which IM injections are considered hazardous, administration via the intravenous route can be attempted providing that the patient is under constant observation, preferably in an intensive care setting. The total dose to be administered is dissolved in 25–50 ml of sterile distilled water and infused by drip infusion over a 30–60 mm period. A V- or Y-type intravenous line is employed with intravenous glucose solution running and the pentamidine dripped into the IV line. Blood pressure determinations should be taken at 10-min intervals during the administration of the drug. If a significant fall in blood pressure is observed, the infusion should be terminated and the blood pressure allowed to return to pretreatment levels. An attempt at reinfusion at one-half the prior rate may be undertaken if circumstances permit, i.e., the patient is stable enough and will also remain under constant observation.

A variety of side effects has been reported following intravenous administration of pentamidine.

These include hypotension, rapid pulse, flushing, dizziness, salivation, sweating, headache, nausea, vomiting, dyspnea, syncope, incontinence, epileptiform activity, and facial edema. Even following intramuscular injection, hypotension, tachycardia, nausea, and vomiting are commonly encountered.

In the experience accumulated by the CDC, 42% of all patients treated with pentamidine and 63% of those treated for 9 or more days recovered.[32] Adverse reactions were noted in 189 (47%) of 404 patients who received the drug for either confirmed or suspected infection. Fully 24% of patients developed impaired renal function; liver dysfunction was observed in 10%, hypoglycemia in 6%, hematologic disturbances in 4%, injection-site reactions (usually sterile abscesses) in 18%, hypotension in 2%, skin rashes in 2%, and hypocalcemia in 1%.

In other centers where there has not been a large preponderance of older and more debilitated patients, i.e., pediatric leukemia therapy centers, the therapeutic recovery rates have approached 80%, but the adverse effects have been similar to those reported by the CDC.[49]

2.10.2. Pyrimethamine and a Sulfonamide

For almost two decades, evidence has existed that the combination of pyrimethamine and a sulfonamide is effective in the treatment of *Pneumocystis* pneumonia. The first evidence came from animal studies of cortisone-treated rats carried out by Frenkel and colleagues.[9] Scattered case reports attest to the efficacy of this approach, but as noted previously, limited comparisons have been made to other forms of therapy such as pentamidine or trimethoprim–sulfamethoxazole.[76,77,79] In addition, Post and colleagues reported effective prophylaxis of the epidemic infantile infections with sulfadoxine plus pyrimethamine.[80] The adult dose of pyrimethamine and sulfadiazine, the latter being available in the United States, is 25–50mg/day pyrimethamine with 4 g/day sulfadiazine.

2.10.3. Trimethoprim–Sulfamethoxazole

Because of the success in experimental therapy obtained with pyrimethamine and a sulfonamide, it was only logical that somewhat similar drugs such as trimethoprim and sulfamethoxazole would be selected for study. Using the cortisone-induced rat model of pneumocystosis, Hughes and collaborators demonstrated that this fixed combination was as effective as pentamidine in the treatment of *Pneumocystis* pneumonitis.[13] In addition, when administered prophylactically, the combination successfully prevented the infection. Following the initial studies in animals, many trials have been reported, including pediatric[14,49] and adult patients.[15,81,82]

Some comments about the dosage of trimethoprim–sulfamethoxazole used in the human clinical studies of pneumocystosis are warranted. Pediatric trials carried out by Hughes used two doses, a low-dosage form (10 mg trimethoprim and 50 mg sulfamethoxazole/kg) and a higher-dosage form (20 mg trimethoprim and 100 mg sulfamethoxazole/kg) administered daily. The latter was found to be more effective. It is important to recognize that this dosage is three times that recommended for the treatment of bacterial infections in adults. Hughes and colleagues definitively compared trimethoprim–sulfamethoxazole and pentamidine in a randomized controlled study of 37 children with *Pneumocystis carinii* infections.[49] The basic study design involved a crossover from the initial drug regimen to the alternative regimen if patients failed to improve. Of 18 patients treated with pentamidine, 11 recovered, one died after receiving the drug alone, and six others required crossover to trimethoprim–sulfamethoxazole; three of these recovered. Of 19 patients treated with trimethoprim–sulfamethoxazole, 13 recovered after initial therapy, whereas six required crossover, of whom two recovered. The overall recovery rates (assuming that the initial agent was most effective and expressed in terms of the initial agent) were 78% with pentamidine and 79% with the combination of trimethoprim and sulfamethoxazole. This conclusion indicates that pentamidine and trimethoprim–sulfamethoxazole are equivalent in effect when used in appropriate dosage. On the other hand, our preference is for trimethoprim and sulfamethoxazole because of the lower incidence of serious side effects. No significant side effects were encountered in either the adult or pediatric studies that would have led to alteration of drug therapy or to termination of treatment.

Several issues of major importance are still unresolved. Can we be sure, for instance, that a failure to respond within 3 days represents a clinical failure

and is an indication for crossover? The first case illustrated in this review did not show clinical improvement until after the seventh day of treatment. We have observed that clinical improvement on either pentamidine or trimethoprim–sulfamethoxazole may be slow and take up to 5–11 days. If there are concurrent pathogens such as the mixture of CMV with a *Pneumocystis* infection, following radiologic changes per se might be a misleading way to evaluate treatment of pneumocystosis. Our present policy is to recommend that patients be given at least 72–96 hr of therapy with trimethoprim–sulfamethoxazole. If blood gases, respiratory rate, and chest radiography remain stable, we would still maintain them on this therapy for an additional 3 days. If, during this initial 72–96-hr period of observation, the patient has worsened in terms of blood gases and chest radiography, we would cross over to pentamidine. The most important clinical principle at this juncture is to consider other possible (simultaneous) infectious processes.

Considerations of dosage are one of the unresolved issues in trimethoprim–sulfamethoxazole treatment.[82] Some preliminary results of early passage of putative cysts in tissue culture have suggested that a concentration of trimethoprim of 5 μg/ml is necessary to inhibit the parasite. Indeed, the preliminary goal of our clinical studies has been to exceed this blood level at least in the postinfusion or posttreatment dose, i.e., 2 hr after ingestion of oral medication or ½ hr to 45 min after the completion of an infusion given over 1 hr. A more serious clinical problem is the question of adequacy of absorption of orally administered drug in critically ill patients. Patients who are intubated, tracheostomized, or in a state of altered consciousness may not be able to take either the tablet or liquid form of trimethoprim–sulfamethoxazole. Those who have undergone open lung biopsy may develop a postsurgical ileus in which the absorption of either component of the fixed combination will be variable.

For these reasons, we recommend the parenteral form of trimethoprim–sulfamethoxazole, for any patient in whom there is a question about the adequacy of absorption of the drug from the gastrointestinal (GI) tract. One problem with this preparation is the relatively large volume of diluent recommended for the infusion of each ampule of trimethoprim–sulfamethoxazole. Originally, the manufacturer recommended that for each infusion some 200 ml of 5% dextrose in water (D_5W) also given, but it has now been shown that the total volume required for the infusion can be as little as 70 ml. The recommended oral dose for an adult is 20 mg/kg, in 3 divided dosages; the daily IV dose may be lower (12–15 mg/kg).

A valid question is whether physicians should really be concerned about a maximum dosage of the parenteral form of trimethoprim–sulfamethoxazole, since the primary aim of therapy in the initial stages of the disease is arrest of the growth of the parasite in the lung. We have obtained blood levels of trimethoprim as high as 15 μg/ml, three times the target goal, with no evidence of toxicity. It would seem prudent, therefore, that in the early stages of the disease, blood levels could be monitored to ensure adequacy of therapy but that there appears little danger of an initial therapeutic overshoot. After 7–10 days of trimethoprim–sulfamethoxazole therapy, the question of antagonism of folate synthesis becomes important but manageable. In *Toxoplasma gondii,* folate antagonists such as pyrimethamine fail to antagonize the mammalian dihydrofolate acid reductase. This is probably the case with *Pneumocystis.* Replacement therapy with folinic acid would be beneficial to mammalian cells but should not promote the growth of the parasite.

We feel that general reluctance to use large doses of trimethoprim–sulfamethoxazole in the early stages of the disease is actually a moot point. The effect of folate antagonism will not manifest itself for perhaps a week during which time the infection should come under control. At that point, there would apparently be little risk of giving the patient folinic acid to obviate an adverse effect on hematopoietic stem cells.

With regard to the issue of safety, trimethoprim–sulfamethoxazole has been associated with hypersensitivity reactions but rarely organ toxicity such as damage to liver or kidneys. Most (but perhaps not all) reported reactions appear linked to the sulfonamide: fever, diffuse erythematous or maculopopular rash, and rarely a picture of vasculitis or Stevens–Johnson syndrome (erythema multiforme). Such reactions have occurred in patients with hematologic malignancies (generally fewer than 10% of treated patients) but appear to be much more common in AIDS patients (up to 80% in patients with *Pneumocystis*). Most reactions can be managed by discon-

tinuation of drug. Mild reactions may require no treatment at all, allowing the patient to complete a course of therapy. However, a severe reaction may require corticosteroid therapy for a few days.

2.10.4. *Pneumocystis* in AIDS

While the clinical features, radiologic manifestations, and laboratory findings are similar in patients with or without AIDS in whom *P. carinii* infection develops, some important differences have been noted in some reviews,[83,84] as summarized in Table 4. The principal observations are that (1) the disease appears associated with a much heavier burden of organisms, (2) it can be more insidious in onset but more slow to respond, (3) clinical response may not correlate with eradication of organisms (as determined by repeat bronchoscopy), (4) clinical infection is associated with a high rate of recurrence, and (5) there is a high incidence of side effects to treatment, particularly with a sulfonamide.[85] Despite the high rate of reactions to trimethoprim–sulfamethoxazole the response rates are not significantly different from that obtained with pentamidine, and we still would recommend that an AIDS patient be started on trimethoprim–sulfamethoxazole unless a reaction to the latter compound has occurred previously. In those patients with a history of a sulfonamide reaction, initial therapy should consist of pentamidine.

TABLE 4. Some Differences between *Pneumocystis* Infection in AIDS Patients versus Those without AIDS

1. AIDS patients have longer clinical prodromes
2. Recovery of pneumcysts from sputum is easier, more invasive studies show a heavier cyst burden, and cysts often persist in lung secretions after treatment in AIDS patients
3. AIDS patients may take 5–11 days to respond to trimethoprim–sulfamethoxazole and require therapy longer than 14 days.
4. Incidence of hypersensitivity reactions to trimethoprim–sulfamethoxazole (e.g., maculopapular rashes) is much higher in AIDS patients—up to 80% in some series—and these characteristically occur after 7–10 days of treatment
5. Pentamidime appears to have less renal and hepatic toxicity in AIDS patients but is associated with occasional precipitous neutropenia and hypoglycemia.
6. Rate of recrudescence is high—in carefully followed AIDS patients not receiving prophylaxis. This rate exceeds 50%, as compared with much lower recurrence rates in leukemic patients.

Patients with AIDS being treated for an initial episode of pneumocystosis should be given a course of trimethoprim–sulfamethoxazole for 4–6 days. If they are not deteriorating the trimethoprim–sulfamethoxazole could be continued for up to a total of 9 days before crossover to pentamidine—but the crossover could be on day 4 if the patient's condition is deteriorating (rapid fall in oxygenation requiring intubation). Generally speaking, the mortality in patients who are crossed over[18] approaches 90% and may exceed this figure in those who are intubated. A first episode of *P. carinii* pneumonia has a good prognosis, with more than 75% of patients responding clinically. The response to successive episodes of the same infection is generally poorer, possibly because of other concomitant infections that cause fever and add to the picture of clinical deterioration. Alternative infectious etiologies should obviously be sought in the febrile deteriorating patient with persistent lung infiltrates, but whether the other possible co-pathogens, e.g., mycobacteria and cytomegalovirus (CMV) can really be effectively treated has not been established.

Pentamidine is not innocuous in AIDS patients. Newer insights into its pharmacology show it can persist and accumulate in the body[86] thus triggering the toxic reactions of neutropenia and hypoglycemia (and occasionally azotemia and liver function abnormalities). After the fourth day of treatment, clinicians should be alert to this agent's complications: alternate-day measurements of WBC, blood sugar, and creatinine are strongly recommended. Large amounts of intravenous glucose can reverse the hypoglycemia, but such infusions may have to be maintained for a few days. The neutropenia can be quite severe and may predispose the AIDS patient to acute bacterial septicemia such as caused by *Pseudomonas aeruginosa*. These toxic reactions are almost always reversible if the patient receives vigorous supportive care.

2.10.5. New Therapy for *Pneumocystis* Infections

The AIDS epidemic, in which *Pneumocystis* infection has figured prominently as the most important opportunistic infection, has triggered a search for

more effective, less toxic treatment. Some approaches include Dapsone plus trimethoprim[87] or the folate antagonist trimetrexate with leucovorin rescue.[88] Clinical results appear satisfactory with the former, but clear-cut superiority or reduction in side effects compared with trimethoprim–sulfamethoxazole is not striking. The ornithine decarboxylase inhibitor, α-difluoromethylornithine (DFMO), has been used on a compassionate clearance basis in the United States.[89] Evaluation of results with DFMO following pentamidine failure is difficult to interpret because of the long half-life of the latter. Results of therapy with DFMO are now being assessed in pilot studies. Used in large doses, DFMO can cause precipitous thrombocytopenia. Other experimental agents continue to be assessed in animal models of pneumocystosis and one class of compounds, sulfonylurea drugs, appears promising.[90]

2.10.6. Other Supportive Measures in Caring for the Patient during Active *Pneumocystis* Infection

Although we have detailed the pharmacologic therapy of pneumocystosis, the nature and quality of ancillary supportive measures in determining recovery cannot be overemphasized. Intubation and, if necessary, a tracheostomy may be required for adequate ventilation. Frequent monitoring of blood gases and adjustment of positive end-expiratory pressure (PEEP) mandates that these patients be managed in either centers for therapy of respiratory failure or intensive care units. Clinicians must be alert to bacterial superinfection and the dangers of oxygen toxicity.

One of the many unresolved issues about the management of *Pneumocystis* infection is whether high–dose corticosteroid therapy or therapeutic lung lavage has any role in the management of the refractory disease. From histopathologic sections of diseased lung, there is no question that the threat to the survival of the patient comes from the hypoxia secondary to the dense intraalveolar exudate. Anecdotal reports of the benefits of steroids or lung lavage in a manner analogous to the management of pulmonary alveolar proteinosis have been made available to the author, but these approaches need formal study. They might be considered in the desperately ill patient who is not improving in the face of apparently optimal pharmacologic therapy.

Another crucial factor affecting survival relates to the nature of the underlying disease. As has been repeatedly observed with the neoplastic disorders, recovery from infection is ultimately related to the ability to achieve a hematologic remission or some improvement in the status of the disease that initially predisposed to infection.

One situation where a major therapeutic decision must often be made occurs with proven *Pneumocystis* infection in the renal transplant recipient. There is no problem in supporting a patient who has received a renal homograft because of the ability to dialyze such a patient and maintain him in an acceptable state of renal function. The clinician therefore has the alternative of allowing the patient to reject his graft, which immediately results in a diminution if not a total withdrawal of immunosuppression, and thereby enhances the ability of the patient's own host defenses to combat this infection. This would appear to be an advisable approach if the renal transplant patient is not responding to an initial 3–5 days of therapy. In other transplant states such as cardiac or bone marrow transplantation, that "luxury" of allowing rejection to occur is not available.

2.11. Patient Isolation and Prophylaxis of *Pneumocystis* Infection

In general, we recommend that patients with pneumocystosis be placed in single-room isolation for the initial 3 days of therapy. Prudent measures such as handwashing before and after patient contact should obviously be employed, but mask and gown precautions seem unnecessary. Prophylaxis of medical personnel and contacts is not indicated, and prophylaxis of other patients in the hospital will depend on the clinical and epidemiologic circumstances.

Several combinations of folate antagonists with sulfonamides may serve as effective prophylactic agents, but the only medication studied extensively in the United States is trimethoprim–sulfamethoxazole. Widespread prophylactic use of this agent might well favor the emergence of resistant strains, something that, at this point, we would be unable to detect and could only suspect after clinical failures had occurred. On the other hand, in certain defined populations of leukemic children or organ transplant

recipients, in whom a high incidence of pneumocystosis has been observed, carefully administered prophylaxis appears justified. The threshold for instituting prophylaxis seems to be an annual incidence of 5% of cases of underlying disease per year. The evidence for the efficacy of prophylaxis comes from the peerless studies of Hughes and collaborators who, for 2 years, followed a group of 80 leukemic children, half of whom received placebo and half of whom received up to two tablets of trimethoprim–sulfamethoxazole (160 mg trimethoprim) on a twice-daily basis.[16] No *Pneumocystis* infections were documented in the prophylaxis group, and a significantly larger number were found in those individuals who were given placebo. After the code was broken in this double-blind study, all patients at that institution were maintained on trimethoprim–sulfamethoxazole, and a negligible incidence of *Pneumocystis* infection has been encountered.

Similarly, the risk of pneumocystosis in marrow transplant recipients has also been reduced to very low levels with prophylactic trimethoprim–sulfamethoxazole (see Chapter 20). Our preference as outlined in a study by Winston et al. of marrow transplant patients is to give intermittent prophylaxis.[17] The specific regimen calls for an administration to adults of three tablets of trimethoprim–sulfamethoxazole (240 mg trimethoprim) twice a day on 2 consecutive days of the week, scheduled in such a way as not to interfere with methotrexate administration. Methotrexate is usually given in marrow transplant recipients to prevent graft-versus-host disease, and the cumulative effect of trimethoprim–sulfamethoxazole and methotrexate may be a sudden precipitous fall in white count. Intermittent prophylaxis has also been recently proven efficacious in children with acute lymphatic leukemia.[90a]

Because of all the other confounding variables, the lowest amount of drug that will produce a prophylactic effect, as appears to have occurred in our bone marrow transplant series, would appear to be the most advisable regimen at this time. As marrow transplant patients recover from their immune deficiency, the risk of *Pneumocystis* appears to decrease, and prophylaxis may not be indicated after half a year post-transplant. In leukemic children, duration of prophylaxis may well be related to the duration that maintenance therapy is given in remission. Evidence now points to the need for a minimum of 5 months of prophylaxis, particularly during the phase of consolidating chemotherapy and the tapering of steroid dose.[41,91]

Second attacks of histologically proven *Pneumocystis* infection have been well documented.[92] We believe it prudent to give continuous prophylaxis to leukemic patients undergoing chemotherapy who have a history of proven pneumocystosis. Indications for prophylaxis of the renal transplant recipient are summarized in Chapter 21. Those with urinary tract infection will probably be treated for that condition with trimethoprim–sulfamethoxazole for 4 months irrespective of risk of pneumocystosis. It also seems prudent to give prophylaxis to those patients with active CMV infection in view of the association of that opportunistic infection with pneumocystosis.

Recommendations for patients with AIDS and an initial episode of pneumocystosis are summarized in Section 2.13.

We have no hesitation about using pentamidine in a patient who develops a histologically proven *Pneumocystis* infection after trimethoprim–sulfamethoxazole prophylaxis. On the other hand, a really careful history is important, since patients may not take their medication, and, indeed, it would be crucial to verify the history before designating such an example as a failure of prophylaxis.

2.12. Postinfection Fibrosis

With increasing numbers of patients surviving documented pneumonitis, the question has been raised as to whether some patients may develop pulmonary fibrosis as a result of this infection. Since repair of lung tissue is mediated by type II pneumocytes, the proliferative and repair response of this cell might actually cause lung damage. The following case history suggests that some patients may develop this postinfection fibrosis. Pulmonary infiltrates may take many weeks to resolve. An increasing number of patients in our experience have some residual changes as seen radiographically. If they eventually come to autopsy, pulmonary fibrosis may be observed. In our experience, this is more likely if the patient is an adult, has received either radiation to the lung or other agents known to injure the lung such as alkylating agents or bleomycin, and if the patient

has an underlying disease that can lead to pulmonary fibrosis per se, e.g., a collagen vascular disease. There is no established method for averting these long-term sequelae.

Illustrative Case 2

This patient was a 44-year-old white man with a long-standing history of ankylosing spondylitis, status postfusion of the spine, who had been receiving long-term corticosteroid therapy because of his rheumatologic disorder He had many manifestations of advanced disease including bilateral ulnar deviation, "swan neck" deformity of the fingers, and flexion contractures of his knees and toes. Outpatient therapy consisted of prednisone, 75 mg alternating with 120 mg every day. In addition, he had been treated with cyclophosphamide, 150 mg/day, PO, until 2 weeks prior to admission. During the addition of cyclophosphamide to his treatment, his prednisone dosage was gradually tapered to a level of 40 mg/day. Beginning 6 weeks prior to admission, the patient complained of symptoms of a mild cold accompanied by shortness of breath and low-grade fever. One month prior to admission he was seen in the emergency room and was treated with oral ampicillin for upper respiratory infection. A week later, however, he had more pronounced shortness of breath but was found to have a normal chest radiograph. Arterial blood gases (ABGs) at that time revealed a pH of 7.48, $Paco_2$ 31 mm Hg, Pao_2 65 mm Hg (3 weeks prior to admission) He was treated with intermittent positive-pressure respiration and bronchodilators and was discharged because the chest radiograph was within normal limits. However, in the ensuing 2 weeks, he had progressive shortness of breath and fever which intensified in the week prior to admission. He was admitted to the hospital with severe shortness of breath.

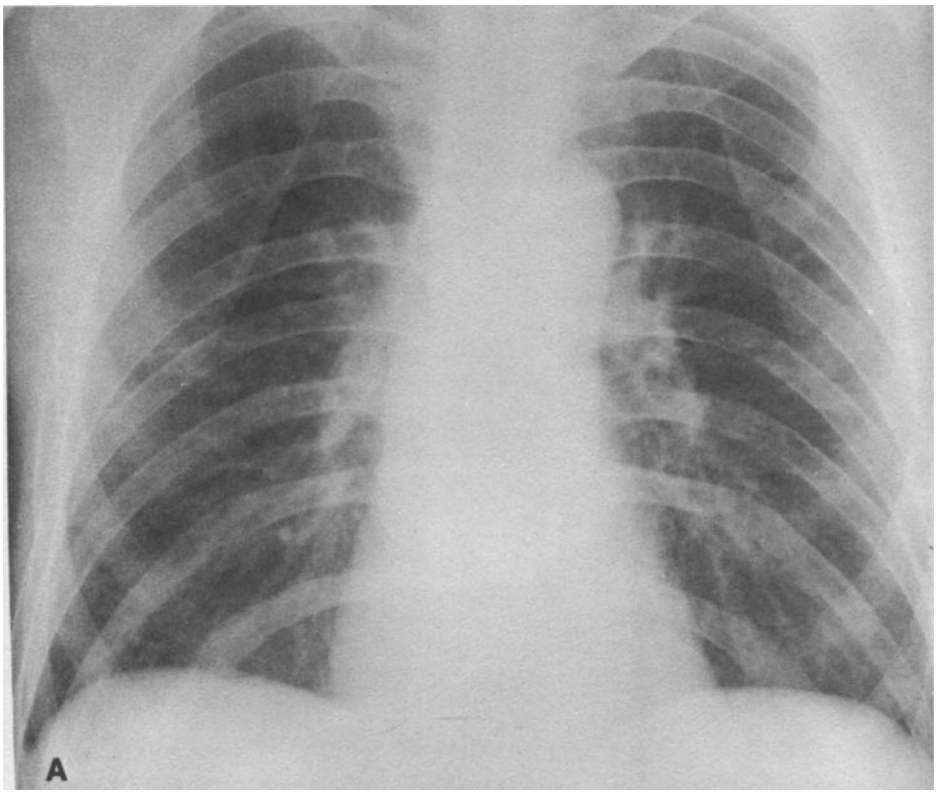
On admission, ABGs revealed pH 7.48, Pao_2 on room air of 30 mm Hg, $Paco_2$ 30 mm Hg, and a bicarbonate of 21 mg/dl. Chest examination revealed diffuse inspiratory rales, bronchial breath sounds, and tubular breath sounds. Chest radiography indicated bilateral interstitial and alveolar infiltrates (Fig. 3). The patient was subsequently intubated and eventually required tracheostomy Fiberoptic bronchoscopy performed through the tracheostomy tube yielded bronchial brushings that were diagnostic for *Pneumocystis carinii* on silver stain Intravenous trimethoprim–sulfamethoxazole was immediately begun at an initial dose of 14 mg trimethoprim/kg per day IV. The dose was divided into four doses given q6h intravenously. Initial peak trimethoprim levels were 4.8 μg/ml with a valley of 3.4 μg/ml. A week after therapy was initiated, peak trimethoprim levels stabilized in the range of 9 7–10.4 μg/ml, and valley levels were found to be in the range of 7 5–9.1 μg/ml

A host of medical complications were encountered during management in the respiratory intensive care unit (ICU) The patient became intermittently hypotensive, had persistent intermittent fevers, developed an upper GI hemorrhage, and experienced a cardiac arrest. Other medications such as aminoglycosides and cephalosporins were given for the possibility of a bacterial component to his pulmonary disease. His ABGs improved after treatment with a decrease in FIo_2 from 100% to 30% to maintain adequate oxygenation. One seizure was observed on the 14th day of trimethoprim–sulfamethoxazole therapy The patient became afebrile with a gradual improvement in blood gases, Pao_2 rising from 30 mm Hg on room air to 90 mm Hg on a 40% FIo_2. Repeat suctioning of tracheobronchial secretions from the tracheostomy tube failed to reveal forms consistent with *P. carinii*. Despite defervescence and increase in Pao_2, the patient's chest radiograph did not clear, and he required intensive care for a subsequent 3 months. His overall condition gradually deteriorated, he became hypoxic once again and he expired with recurrent fevers possibly caused by bacterial infections (although the etiology was never documented). His chest radiograph continued to show diffuse, persistent pulmonary infiltrates. A postmortem examination revealed findings consistent with 26 years of ankylosing spondylitis involving the entire spine. He was found to have diffuse amyloidosis, and the sections of lung revealed consolidation and extensive fibrosis but no *Pneumocystis carinii*

Comment. This case demonstrates many points. Although the patient did not have a neoplasm, he was treated with high doses of corticosteroids for a rheumatologic disorder, and cyclophosphamide had been recently discontinued before the intense flare of his symptoms. In addition, there was significant reduction of corticosteroid dosage prior to the exacerbation of his respiratory symptoms. Of considerable interest was the observation that the patient had approximately 6 weeks of symptoms with normal chest radiographs prior to the development of fulminant pneumonitis. The effect of corticosteroids in suppressing the inflammatory response coupled with exacerbation of disease when the dosage was tapered were considered important features of Case 2. This patient improved clinically, as has been our experience with the great majority of patients treated with intravenous trimethoprim–sulfamethoxazole, but pulmonary infiltrates failed to clear. An adequate dosage of trimethoprim–sulfamethoxazole was given, and the patient gradually became afebrile with increasing Po_2; therefore, no consideration was given to changing treatment to pentamidine.

From a diagnostic point of view, material adequate for establishing the diagnosis was obtained from fiberoptic bronchoscopy and brushing once an endotracheal tube had been inserted. This approach may be valuable in patients with marked hypoxia and demonstrates the ability to establish a diagnosis even if the patient requires intubation. On the other hand, the disturbing finding was that this patient gradually manifested a downhill course, dying in respiratory failure after approximately 3 months in the hospital. At autopsy, a diffuse fibrotic process but not *Pneumocystis carinii* was found in the lung. Many investigators believe that infection with *Pneumocystis* may cause extensive alveolar damage and pulmonary fibrosis. This case may be such an example. On the other hand, this patient had received cyclophosphamide and had amyloidosis secondary to this rheumatic disorder Oxygen toxicity may have contributed to postinfectious fibrosis. Death in this case cannot be attributed to pneumocystosis but perhaps to postinflammatory fibrosis that was an inexorable sequel to the infection. Postinfectious fibrosis has been rarely seen in children but may be more common in the adult form of the disease.

Clinically, patients have persistent pulmonary infiltrates, remain hypoxic, and may be suspected of having drug-resistant pneumocystosis. Our policy has been to maintain the initially se-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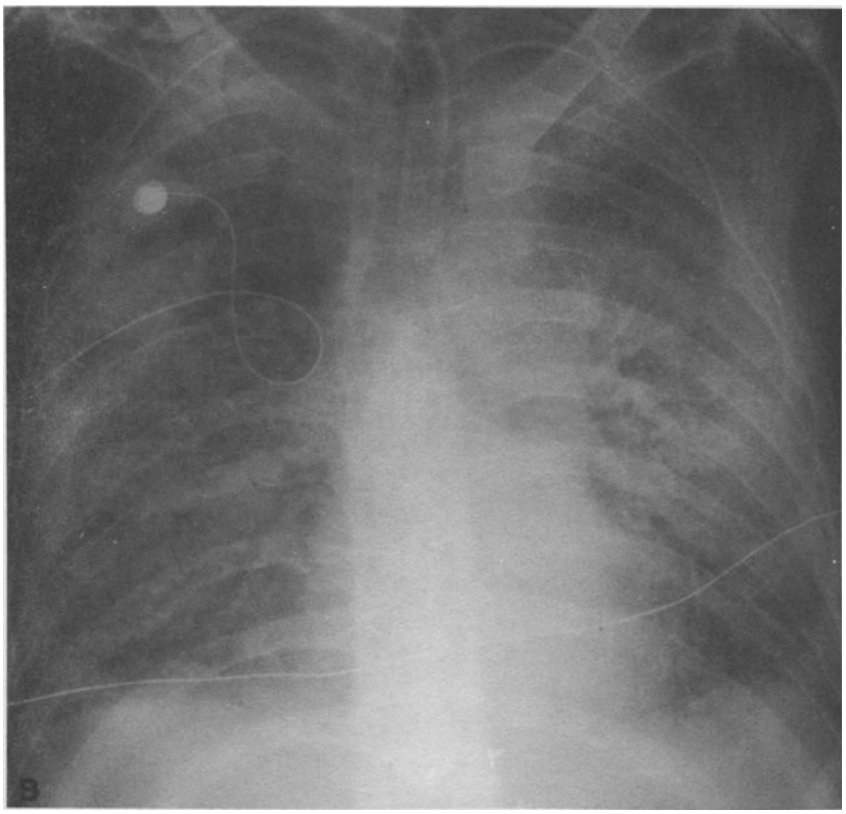
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lected medication if the patient is stable and experiences some defervescence.

2.13. Overview of Therapeutic and Prophylactic Approaches

Considerable progress in the management of *Pneumocystis* infection has been made during the past decade, but the experience during the current epidemic of AIDS indicates that newer, safer forms of therapy and prophylaxis are badly needed. The advent of trimethoprim–sulfamethoxazole has led to an oral and parenteral form of therapy which, although not convincingly superior to pentamidine, appears to be easier to give and is associated with fewer side effects except in AIDS patients. On the other hand, the seeds of a real clinical problem may be seen by the ready availability of an oral preparation, since we will not know if resistance develops unless we are confronted by a convincing set of clinical failures. Whereas trimethoprim–sulfamethoxazole is an excellent antibacterial agent for many gram-negative rods and most gram-positive cocci (with the exception of *Enterococcus* and *Pseudomonas*), its prophylactic use in the immunocompromised patient population should probably be restricted to patients treated in a program experiencing a high incidence of *Pneumocystis* infection.

What criteria should be applied in making the decision to institute routine prophylaxis? At the present time, prophylaxis would seem to be indicated in recipients of organ transplants and in leukemic children if their calculated annual incidence of pneumocystosis exceeds 5%. This recommendation is based on a review of the available literature and is not meant to be dogma: clearly, individual circumstances, magnitude of problem, ease of diagnosis of infection, and clinical success rate after the documentation of proven infection are all factors to be considered before prophylaxis is given.

In leukemic patients, prophylaxis can probably be stopped several months after the cessation of all chemotherapy (including corticosteroids) and the achievement of a solid remission.

For patients with AIDS, controlled data on prophylactic use have been difficult to marshall. The high documented recurrence rate mandates that some prophylactic measure be given, but perhaps most AIDS patients have had a hypersensitivity reaction to trimethoprim–sulfamethoxazole. Those who can tolerate this medication should continue to receive 160 mg trimethoprim (with a corresponding amount of sulfamethoxazole) twice a day, though less frequent dosing may suffice. One group has found that most patients reacting to trimethoprim–sulfamethoxazole can tolerate pyrimethamine–sulfadoxine (Fansidar), 25 mg : 500 mg, in a dose of one tablet of the fixed combination per week orally.[93] The reason that this preparation is generally well tolerated is unknown but might be related to the lower dose of the long-acting sulfonomide. Occasional severe reactions, not unlike Stevens–Johnson syndrome have been observed. Thus, this form of prophylaxis must be initiated with care: we advise it be started after all evidence of a previous drug reaction has subsided—either 2 weeks after trimethoprim–sulfamethoxazole or 4 weeks after pentamidime. The risk of a reaction must be judged against the risk of recurrent pneumocystosis and a severe reaction should be treated with a short course of corticosteroids.

For the AIDS patients who cannot tolerate a sulfonomide containing preparation, a single intramuscular dose of pentamidime can be given every 2–4 weeks on an outpatient basis. This appears to be effective; studies are now under way to evaluate aerosal administration of pentamidime as a way of avoiding the systemic toxicity of that agent.

3. Babesiosis

More than 70 species of protozoan parasites belonging to the genus *Babesia* have been isolated and identified throughout the world. Only within the last two decades has it become apparent that these species are pathogenic for man, and, remarkably, three of the first four human cases reported in the literature died

FIGURE 3. (A) Chest radiograph of Illustrative Case 2 for symptoms of cough and dyspnea several weeks before documentation of *Pneumocystis* infection. (B) Despite defervescence and clinical improvement, bilateral diffuse pulmonary infiltrates persist in Case 2 during trimethoprim–sulfamethoxazole treatment.

of acute disease.[94] All these first four patients had been splenectomized, a procedure that presumably rendered them susceptible to this infection. Although cases of human babesiosis are apparently uncommon, the worldwide distribution of these protozoan parasites and the results of epidemiologic studies indicating that mild and subclinical infections occur form a sufficient basis to recommend that disease caused by *Babesia* species should be considered among the diagnostic possibilities in immunocompromised febrile patients.

It is likely that epidemics of babesiosis date back to biblical times, when devastating epizootics occurred among cattle and domestic animals. The protozoan parasite was first described by Babes in 1888 who identified an intraerythrocytic agent which he mistakenly took for a bacterium.[94] Five years later, however, Theobald Smith and F. L. Kilborne investigated an epidemic of fever in cattle and established that the pathogen was both protozoan and transmitted by a blood-sucking tick.[95] *Babesia* species subsequently have been isolated from sheep, goats, horses, swine, dogs, cats, rodents, and many other warm-blooded animals. A number of species of ticks have been shown to transmit babesiosis including the common *Ixodes* and *Dermacentor* ticks. Thus, a history of tick bite can be a particularly important clue in establishing the diagnosis of babesiosis. Clearly, however, a tick bite may be an easily overlooked part of the history and an experience that the patient may readily forget or even be unaware of, so the apparent absence of tick contact does not exclude the diagnosis.

The occurrence of babesiosis in animals is worldwide, and thus, the potential for human disease must be viewed in this light. The first case was described in the Balkans, the second in California, and the third case in Ireland. Seroepidemiologic studies suggest that the disease occurs in Europe and Central and South America. More recently, a series of interesting reports of human disease has emanated from the Martha's Vineyard, Nantucket, and the eastern Long Island portions of the United States.[96–98] Careful epidemiologic studies have suggested that this infection is more common than the scattered appearance of cases with clinical disease. Furthermore, although many of these cases have been associated with bovine ticks, epidemiologic studies have failed to implicate tick contact in some patients. Noteworthy about the outbreaks in the Massachusetts area has been that the majority of cases have been in patients with intact spleens. It now appears that although splenectomy increases susceptibility to babesiosis, it is not the only factor predisposing to clinical disease.

3.1. Clinical Features

Although babesiosis is recognized as having a broad spectrum of manifestations from inapparent infection to a fulminating illness, perhaps the most important factor to remember is that it can present as a malarialike illness. Indeed, an examination of blood smears can result in the identification of morphologic forms that may be easy to confuse with *Plasmodium* species.

The severe clinical forms of babesiosis have a predilection for individuals without spleens, and four such cases have proven to be fatal with a clinical course terminating in about a week. One recent case report describes a patient splenectomized for Hodgkin disease who developed fatal infection caused by *B. divergens*.[99] High fever, chills, anemia, jaundice, hemoglobinemia, hemoglobinuria, hypotension, anuria, and coma have been serial clinical manifestations. The anemia clearly appears to be of a hemolytic type comparable to that observed with malaria. Serious illnesses have lasted about 1 month in those persons who have recovered, and even following recovery, patients have required prolonged convalescence because of postinfection asthenia.

It appears from careful documentation of the history of tick feeding that the incubation period ranges from 7 to 21 days, with a median of approximately 2 weeks.

Those individuals with moderate to severe illness usually have had intact spleens and have tended to be older than 40 years of age. There is some indication that although exposure may be the same for persons of all ages, clinical disease has a predilection for the elderly. The best-studied cases of this form of babesiosis have been summarized in the report of the Nantucket cases by Ruebush et al.[97] In decreasing order of frequency, symptoms have included fever, fatigue, myalgia, arthralgia, mental depression, drenching sweats, shaking chills, nausea and vomiting, hyperaesthesia, and splenomegaly. Lymph node

enlargement, rash, and abnormal neurologic signs were absent. Temperature elevations were as high as 106°F. Following clinical infection, some of these patients have experienced long periods of low-grade fever and lassitude with symptoms lasting for up to 2 years.

Subclinical and mild infection have been documented by retrospective serologic studies and/or microscopic examinations of blood films. An epidemiologic survey carried out by Ruebush and colleagues included the study of a total of 964 specimens, 577 from Nantucket, 154 from Martha's Vineyard, and 100 from Cape Cod.[97] Twenty-one specimens were considered positive with a titer by indirect immunofluorescence of 1:64 or greater. Eleven of the 577 samples were collected from patients at the Nantucket Hospital for routine diagnostic tests, and ten of 133 specimens were obtained from Nantucket residents and visitors who had a history of tick bite or fever. Of the 19 seropositive patients who could be contacted, six had a febrile illness in the previous 6 months and had been hospitalized, but all were well at the time of blood specimen collection, and none had a history of malaria. Of the hospitalized patients, one had had a short febrile illness with recovery followed by relapse. Of the three hospitalized patients, one was found to have a blood smear showing *Babesia,* whereas blood from the other two was inoculated in hamsters. *Babesia microti,* the most common species found in the Eastern United States, was isolated in both instances. Similar documentation of mild or innapparent infections have occurred in low gulf coast areas of Mexico, Nigeria, and Georgia.

The potential for transmission of babesiosis has been raised during the evaluation of putative cases of transfusion-associated malaria. Healy and colleagues identified a case of probable babesiosis while attempting to trace the source of a case that was diagnosed as *P. vivax* infection.[100] One of five blood donors had light parasitemia; the clinical disease in both the donor and the recipient of the transfusion could either have been malaria or babesiosis. The serologic evidence favored babesiosis. Thus, the potential for human infection caused by transmission of blood from donor to immunocompromised host remains as much a possibility with this organism as it does with malaria. Moreover, in the United States, the reservoir for *Babesia* infection would appear to be far larger than that for malaria.

3.2. Laboratory Diagnosis

Human infection has been documented with three relatively small *Babesia* species, *bovis, divergens,* and *microti.* A presumptive diagnosis of babesiosis can be made by identification of the parasite in host erythrocytes. Thin blood films are stained by the Giemsa technique as for the identification of malarial parasites (Fig. 4). In severe infections, up to 50% of patients' erythrocytes may be parasitized. The merozoites appear to be pear-shaped, round, or oval, varying in size from 1 to 5 μm in length. All species of *Babesia* invade and propagate in erythrocytes only. They are easily mistaken for *Plasmodium* species, either *falciparum* or *vivax.* However, unlike *falciparum* parasites, *Babesia* species do not leave residual hemozoin pigment after they ingest vacuole-stored hemoglobin. Therefore, the absence of pigment in the parasitized blood cell is a hallmark of babesial infection. *Babesia* multiply by forming two or four or more merozoites, and in heavy infections, these forms may be seen outside of the erythrocytes. Tetrad forms (''Maltese crosses'') may be produced by budding. The presence of these tetrad forms will reliably distinguish *Babesia microti* from the *Plasmodium* species.

Next to direct visualization of *Babesia* in blood smears, serologic techniques are the most important means for diagnosis and are crucial for epidemiologic studies. Serodiagnosis is the only practical means for identifying subclinical infection, since positive blood smears are usually obtained only from patients with clinical disease. An indirect immunofluorescent antibody test has been developed and evaluated by Chisholm and co-workers at the CDC.[101] The antigen is a strain of *Babesia microti* harvested from hamsters. After an initial incubation of this antigen with human serum, antihuman immunoglobulin labeled with fluorescein is added. A diagnostic titer is considered to be 1:64 or greater. It is not unusual for acute-phase specimens to have titers equal to or greater than 1:1024. Seropositive samples may cross-react with a number of species of *Babesia* and three species of *Plasmodium* including *vivax, falciparum,* and *brasilianum.* However, titers

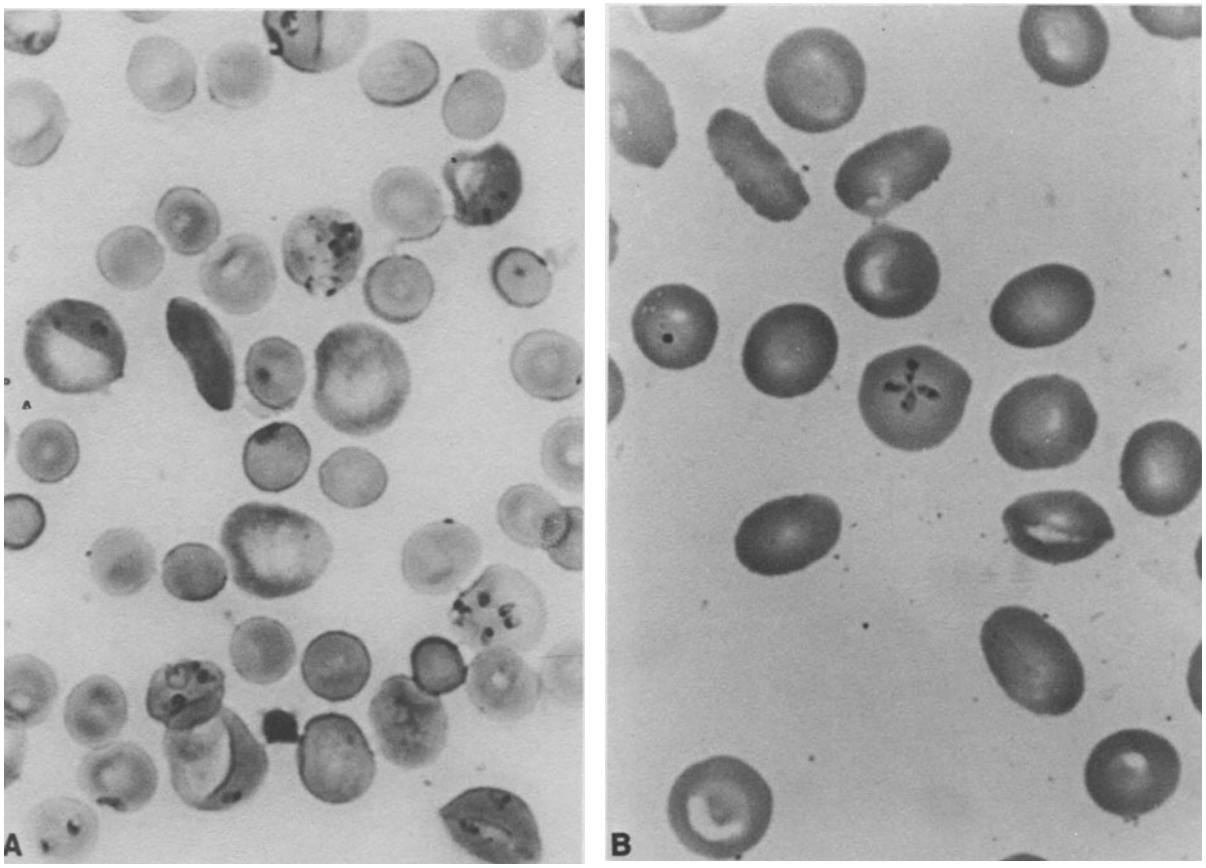

FIGURE 4. (A) Wright-stained smears of peripheral blood demonstrating numerous ring forms from case of human babesiosis. These forms are easily confused with *Plasmodium* species (B) Classic tetrad form (''Maltese cross'') that distinguishes *Babesia* infection from malaria.

against the infecting *Babesia* species are higher than to the other *Babesia* and plasmodial antigens tested.

Physicians should be aware of the identity between the ticks that are the carriers of Rocky Mountain spotted fever and *Babesia* species. Clinically, other diseases that must be considered in differential diagnosis are Weil disease, leptospirosis, and brucellosis. Dammin reviewed the histopathologic laboratory findings in the mild and severe cases.[94] Cases of fatal disease have been deeply jaundiced and showed marked congestion of abdominal viscera, lungs, and brain. This is the result of plugging of capillaries by parasitized erythrocytes in a manner that resembles malarial infections.

3.3. Treatment

It is logical that chloroquine would be one of the first agents to be used for the treatment of babesiosis because of the initial confusion of this entity with malaria. Studies reported by Ruebush and Spielman demonstrate that chloroquine phosphate, 1.5 g initially and 0.5 g/day for 2–12 weeks, results in symptomatic improvement and abatement of fever within 3–7 days.[96] However, parasitemia persisted for more than a month in three patients, and convalescence was prolonged. Indeed, there is no definite relationship between chloroquine therapy and reduction of parasitemia in humans and animals.[102] The blood

of patients receiving chloroquine will still infect experimental animals, and the major effect of this agent may be to suppress inflammation. Conversely, symptomatic improvement in the absence of chloroquine has occurred. It appears that most persons infected with *Babesia microti* can clear the infection without specific antiparasitic therapy if they have intact host-defense mechanisms.

Several alternatives to chloroquine treatment are available. A number of tetracyclines reduce parasitemia. Pentamidine and diminazene (as Berenil available from the Parasitic Diseases Drug Service of the CDC) reduce and suppress parasitemia, but parasitemia recurs after discontinuation of these drugs. Diminazene is effective therapy in animals, but its use in one case of human infection was associated with Guillain–Barré syndrome.[96] Diminazene or pentamidine should be considered only if heavy parasitemia is documented. Some authorities suggest the use of trimethoprim–sulfamethoxazole to treat active infection,[94] but the clinical experience is limited.

The most promising therapeutic approach now appears to be the combination of clindamycin, 600 mg qid, and quinine, 650 mg tid.[103] Treatment should be considered with this regimen if parasitemia is documented in immunocompromised hosts.

In summary, it appears that babesiosis has had a minor effect on public health of the world's population despite the fact that there are few places in the world that are free of *Babesia*. Patients with impaired host defenses, such as following splenectomy, or recipients of blood transfusions should be evaluated for the possibility of this infection if they develop fever, chills, hemolysis, and jaundice.

4. Giardiasis

Giardia lamblia has received increased attention as an important cause of diarrhea and malabsorption in both the normal host and patients with immunodeficiency. Like many other modern opportunistic pathogens, it was often regarded as a nonpathogenic commensal organism until the past decade.[104] However, several recent lines of evidence have now established *G. lamblia* as an important cause of GI disease.

1. Although there is a high rate of inapparent infection, *G. lamblia* has been shown to be a major etiologic agent of diarrhea in individuals returning from the Soviet Union, Southeast Asia, and other geographic areas of high endemicity.[104–106]
2. Numerous well-studied epidemics of water-borne diarrheal disease in the United States have been linked to this organism. Indeed, *G. lamblia* is the most frequently documented cause of water-borne epidemic diarrheal disease in the United States.[104,107–109]
3. *G. lamblia* has been convincingly associated with diarrhea and malabsorption in patients with a variety of types of hypogammaglobulinemia.[104,110,111]

4.1. The Organism

Approximately 50 species of *Giardia* have been isolated from primates, other mammals, and even amphibians. In the past, it was assumed that each host species harbored its own *Giardia* species, and hence, each was given its own name and species designation. However, recent studies demonstrating cross-infectivity between human *Giardia* strains in the beaver, the dog, and other nonhuman hosts have led to serious doubts about species specificity.[104,109]

The human strain, *G. lamblia,* may take one of two forms. The motile form, or trophozoite, is responsible for the disease manifestations in the upper small intestine. Cysts, the more hardy infective form, develop as organisms, traverse the bowel, and are subjected to increasing degrees of dehydration. The trophozoite is a 13–19 × 8–11 μm piriform-shaped structure whose most important anatomic feature is a large, bilobed concave sucking disc by which the organism becomes attached to the upper small intestinal epithelium. The cyst forms are thick-walled, oval structures measuring 8–12 × 7–10 μm. Trophozoites may occasionally be found in the stool of patients with a very watery diarrhea and, presumably, a very rapid intestinal transit time. Otherwise, only cysts will be observed in the stool, and duodenal intubation is necessary to demonstrate the trophozoites.[104]

4.2. Epidemiology

The most important means of transmission of *G. lamblia* is by ingestion of contaminated drinking water. Municipal water supplies, rural streams, and individual wells have been implicated as sources of infection. Cysts may remain viable in fresh water for months and are not destroyed by usual chlorination procedures. It has previously been assumed that the source of water contamination was infected human feces. However, increasing evidence suggests that contamination with animal excreta may play a role in the transmission of infection to humans. Only beavers have been definitely implicated as playing such a role. In addition, direct person-to-person transmission may occur via the fecal–oral route and is particularly important among young children, institutionalized subjects, and male homosexuals.[104,108,109,112–114]

The inapparent infection rate appears quite high. Children will manifest symptoms more frequently than will adults with a similar parasite load. Previous exposure may enhance protection against reinfection in immunologically intact individuals.[104,115,116]

4.3. Pathogenesis

Data from both experimental *Giardia* infection and from investigation of water-borne epidemics in which small numbers of *Giardia* cysts were found in quantitative stool studies suggest that the ingestion of as few as 100 cysts will reliably establish infection.[117] After ingestion, excystation takes place in the upper GI tract, yielding the disease-causing trophozoites. An unexplained paradox is that although a low pH appears to be necessary for excystation to occur in vitro, individuals with achlorhydria appear to be at increased risk of symptomatic infection. Once excystation occurs, the motile flagellated trophozoites replicate, move along the surface of the small bowel, and attach themselves directly to the mucosa via their bilobed sucking discs. These events result in shortening and flattening of the villi, elongation of crypts, and increased mitotic activity of the epithelial cells. Electron microscopic studies reveal thinning of the surface coat of the brush border and a variety of changes in the microvilli. The structural abnormalities observed in the microvilli are reflected in the functional defects that are observed in symptomatic individuals: disaccharidase deficiency (the clinically most important being an acquired lactase deficiency) and evidence of protein, fat, and vitamin malabsorption. Only in the minority of the most heavily infected persons do trophozoites actually penetrate the bowel wall. Even without such penetration, however, an inflammatory response may be observed in the underlying submucosa, consisting of variable numbers of polymorphonuclear leukocytes, plasma cells, and lymphocytes.[104,118–122]

Susceptibility to infestation with *Giardia* appears to be increased not only in patients with achlorhydria, but also in those with pancreatic dysfunction[123] and protein-calorie malnutrition.[104] This last may be secondary to immune deficits created by the malnourished state. Overgrowth of the small bowel with colonic flora may occur in giardiasis, and it has been hypothesized that this may play a role in the pathogenesis of the diarrhea, the bacteria causing bile salt deconjugation or, perhaps, direct injury to the small bowel mucosa.[124]

Why some individuals with giardial infestation develop symptomatic disease and others do not is poorly understood. Three observations point toward immunologic factors:

1. Epidemiologic studies indicate that attack rates are higher in children than in adults and in newcomers rather than long-term residents of endemic areas.[104,115]
2. Studies in human subjects suggest an increased incidence and severity of giardiasis in patients with nodular lymphoid hyperplasia of the small bowel, a variety of dysgammaglobulinemias, and hypogammaglobulinemic sprue.[104,110,111,121] This appears to underscore the importance of antibody production in the protection against giardial infection, particularly local secretory antibody in the intestine itself.
3. Studies in the mouse model of giardiasis have demonstrated that resistance to reinfection follows an initial infection that clears. By contrast, athymic mice neither clear their initial infection nor acquire resistance. Resistant mice can transfer resistance to their progeny through breast milk in which anti-

trophozoite antibodies of the IgA and IgG class can be demonstrated.[104,116,125–127]

Thus, although the precise mechanisms of resistance to giardial infection are incompletely understood, there is already ample evidence that specific local and systemic humoral immune mechanisms are involved.

4.4. Clinical Manifestations

An estimated 20–50% of infected individuals develop clinical symptoms 1–3 weeks after initial infection. Characteristically, there is an abrupt onset of midabdominal cramps, watery, foul-smelling diarrhea, abdominal distention, nausea, and flatulence. Chills and/or low-grade fevers may occur at the beginning of the illness but should not persist for longer than a few days. Vomiting, headache, belching, and a generalized malaise are not uncommon. After the first 3–4 days, a variety of patterns of illness may be observed in the untreated patient: in the majority of individuals either persistent or recurring symptoms of moderate severity will occur, with brief episodes of foul-smelling diarrhea, abdominal distention, flatulence, belching, and substernal discomfort being noted. Some patients experience symptoms over weeks to months and have as the major manifestations malabsorption, weight loss, and a general failure to thrive. The last is the major pattern of disease seen in the immunocompromised patient. It is important to emphasize that, particularly in ethnic groups with a predisposition to lactase deficiency, chronic symptoms of diarrhea, flatulence, abdominal distention, and malabsorption may persist for months even after effective therapy has been administered. A lactose-free diet will correct these abnormalities.[104]

4.5. Giardiasis in the Compromised Host

The most firmly established relationship of giardiasis to a host defense defect is that with dysgammaglobulinemias such as occur with the variable immunodeficiency syndrome.[110,111,121] In the best study of this relationship, Ament et al.[111] carefully studied clinical symptoms, multiple small intestinal biopsies, stool samples, and a variety of GI function assays in a group of 39 patients with primary immunodeficiency syndromes with altered γ-globulin production. Their findings are as follows: *Giardia lamblia* was found in eight of nine patients with symptoms of diarrhea, weight loss, vomiting, and anorexia, but in only three of 30 patients who were free of gastrointestinal complaints. Pathologic and functional abnormalities included mild to severe villus abnormalities, malabsorption (often severe) of folic acid and vitamin B_{12}, steatorrhea, lactose intolerance, generalized disaccharidase deficiency, and protein-losing enteropathy. Virtually all these abnormalities were reversed following treatment with metronidazole and eradication of the parasite. Of these eight patients, one had infantile X-linked agammaglobulinemia, and seven were classified as having the variable immunodeficiency syndrome. An extremely important observation was the difficulty encountered in establishing the diagnosis of giardiasis. Stool examination yielded the diagnosis in only 30% of cases; multiple intestinal biopsies revealed the patchy nature of the involvement. Examination of such biopsies and/or Giemsa-stained smears of mucus adhering to the biopsies was required to make the diagnosis in most instances.

Whether other immunocompromised patients have an increased incidence and/or severity of giardiasis remains unclear. Clearly, any diarrheal syndrome in patients with known dysgammaglobulinemias, particularly of the variable type, should lead the clinician to suspect giardial infection. Although it has been suggested that apparently normal individuals with giardiasis have a deficiency in intestinal production of IgA,[128] current evidence is against this hypothesis.[129] Therefore, in an apparently normal adult, the diagnosis of giardial infection by itself should not lead to an elaborate evaluation for immunodeficiency. We have observed several cases of symptomatic giardiasis in renal transplant patients, but these instances could have been chance events rather than reflecting increased susceptibility to this infection.

4.6. Diagnosis

The diagnosis of giardiasis can usually be made rather easily by examination of several stool specimens in patients with acute disease. Using direct smear and formol–ether concentration techniques on stool specimens from patients with giardiasis, the

diagnosis can be made on 76% of patients after one stool examination, 90% after two, and 97.6% after three.[104] With more chronic disease, diagnosis by stool examination may be more difficult, particularly since cyst excretion may be episodic. Purging does not appear to increase diagnostic yield. In such instances, particularly in patients with dysgammaglobulinemias in whom stool examinations notoriously give low yields,[111] examination of small bowel contents for trophozoites should be carried out.

The combination of small bowel biopsy and duodenal intubation are the definitive techniques for diagnosing giardial infection. An alternative method is the use of the Entero-test. With this technique, a fasting patient swallows a capsule containing one end of a long string, the other end of which is taped to the patient's face. Over the course of 4 hr, the capsule dissolves, and the string reaches the duodenal–jejunal junction. The string is then withdrawn, and the distal 20–30 cm which is coated with a bile-stained mucus is then examined microscopically for the presence of trophozoites.[130]

In some patients with highly compatible histories, appropriate epidemiologic exposure, and/or an underlying dysgammaglobulinemia, a therapeutic trial of antigiardial therapy is indicated.

4.7. Treatment

Any patient harboring *G. lamblia* with or without symptoms should be treated for the disease. The treatment regimen of choice is quinacrine hydrochloride (Atabrine) at a dose of 100 mg three times a day for 7 days. Possible adverse effects of this regimen include skin rash, GI disturbances, fever, and psychosis. On the whole, however, this regimen is quite well tolerated and is effective in at least 95% of persons. An alternative drug, particularly useful in children because it is available as an oral suspension, is furazolidone, although experience with this agent is much less.[104]

A more controversial form of effective therapy involves use of metronidazole (Flagyl). Metronidazole at a dosage of 750 mg three times a day for 10 days is probably as effective as quinacrine.[131] A simpler regimen consisting of a single dose of 2 g/day for 3 days as a single daily dose appears to be almost as effective.[132] Of concern, however, are reports that metronidazole may be carcinogenic in mice[133] and mutagenic in bacteria.[134] One long-term follow-up study in women who had received this drug for treatment of *Trichomonas vaginalis* infection failed to reveal any excess incidence of tumors.[135] The issue must be regarded as unsettled at the present time.

Whatever regimen is used, the clinician must be aware that relapse rates of 5–20% have been noted. Hence, close clinical follow-up and reexamination of stool and/or small bowel contents may be necessary in patients with only partial or temporary responses to therapy.[104]

5. Toxoplasmosis

Clinical and epidemiologic interest in toxoplasmosis as a disease of the immunocompromised host has paralleled interest focused on *Pneumocystis carinii* pneumonia. Fewer cases of toxoplasmosis relative to pneumocystosis have been reported in the recent literature, but the possibility of toxoplasmosis is often raised in the very same patients suspected of having pneumocystosis. However, *P. carinii* infection is almost always confined to the lung, whereas the causative agent of toxoplasmosis, *Toxoplasma gondii,* infrequently causes pneumonitis alone. More commonly, *T. gondii* affects multiple organ systems and has a special predilection for the central nervous system (CNS) in the immunologically impaired host. The special predilection of clinical toxoplasmosis for the CNS has been borne out by the current AIDS epidemic.[136,137]

Impressive progress in recent years has been made in understanding the epidemiology of toxoplasmosis.[138] Unlike *P. carinii,* which cannot be cultivated in vitro, abundant taxonomic and microbiologic information is available on this common protozoan parasite.

Toxoplasma gondii has been implicated in several clinical problems that are not covered extensively in this chapter: (1) as an infectious agent that can trigger recurrent abortions in humans (a role that is debated); (2) as the cause of a congenital infectious syndrome characterized by encephalitis, mental retardation, and microcephaly; and (3) as an ocular infection, usually retinochoroiditis. In the area of

diagnosis, there are still major problems in interpreting serologic test results. The problems with diagnosis are most important in the immunocompromised host where, like pneumocystosis, *Toxoplasma* infections can be life-threatening medical complications. Many cases of toxoplasmosis in immunosuppressed patients have been diagnosed only at necropsy. This is unfortunate inasmuch as effective therapy for the infection has been available for 30 years and consists of the use of one folate antagonist, pyrimethamine, combined with a sulfonamide such as sulfadiazine.

5.1. History

It is likely that the first description of toxoplasmosis in man dates back to the work of Samuel Darling, a pathologist and parasitologist in Panama who described a case of a young male with an acute illness characterized by fever, headache, and joint stiffness, who had encysted organisms in a muscle biopsy suspected of containing *Trichinella* or *Sarcosporidia*.[139] This report of probable toxoplasmosis published in 1908 took place in the same year that Nicolle and Manceaux identified *Toxoplasma* in a North African rodent, the gondii, and Splendore made the same observations in rabbits in Brazil.[140] Some 15 years later, Janku described retinal parasites in sections taken from a baby who apparently had expired of congenital toxoplasmosis.[141] This was the first suggestion of a relationship between a human illness and the parasitic forms identified in various mammalian and avian species in different continents. It has subsequently become apparent that strains of toxoplasmosis obtained from human and animal sources differ from country to country. In 1939, Wolf and colleagues demonstrated *Toxoplasma* by inoculation into animals of tissues taken from a baby with neonatal encephalitis.[142] Sabin subsequently showed that antibodies to *Toxoplasma* could be demonstrated by mixing organisms with the sera of patients and injecting the mixture into the skin of the rabbit.[143] This was a complex and cumbersome technique for measuring antibodies to *Toxoplasma*. In 1948, Sabin and Feldman described what has become the reference serologic test for detection of antibodies against *Toxoplasma*.[144] The basis for the test is the observation that *Toxoplasma* trophozoites do not stain with methylene blue dye in the presence of specific antibody. Subsequently, complement fixation, indirect hemagglutination, immunofluorescence, and an enzyme-linked immunoabsorbent assay (ELISA) of antibodies have been described.

The identification of *Toxoplasma* as an important etiologic agent of uveitis was initially documented in histopathologic studies. The lymphadenopathic form of toxoplasmosis was described during the early 1950s by Siim[145] and by Gard and Magnusson,[146] and there has been widespread recognition that this is the most common clinical manifestation of toxoplasmosis in man. It has been concluded that approximately 15% of otherwise unexplained lymphadenopathy is caused by toxoplasmosis.[147] Confusion of this finding with lymphoma has been well described, as have cases of coexistent toxoplasmosis and lymphoma.

5.2. The Organism

Toxoplasma gondii are obligate intracellular protozoan parasites. They are considered to be coccidian because of an enteroepithelial cycle and are presently classified among the sporozoa in the suborder *Eimerina*. Three forms exist in nature: trophozoites, tissue cysts, and oocysts. The trophozoites are crescent to oval in shape and are on the order of 3–7 μm in size. They are well stained by readily available Wright or Giemsa stains. This form is present in the acute stage of infection, having the capability of penetrating all mammalian cells except nonnucleated RBCs. Division continues until the host cell lyses or a tissue cyst forms. These trophozoites can be propagated intraperitoneally in a variety of warm-blooded animals, in tissue cultures, or in embryonated eggs (Fig. 5). Tissue cysts vary in size from 10 to 100 μm and may contain thousands of organisms. Tissue cysts are present in the chronic or latent phase of *Toxoplasma* infection and can be identified by PAS stain. These cysts are infective after oral ingestion because disruption of their walls by gastric juice and digestive enzymes frees viable trophozoites that survive long enough to penetrate GI mucosa. Freeze-thawing, heating in excess of 60°C, or desiccation will destroy tissue cysts.

Oocysts are oval structures 10–12 μm in diameter found only in feline hosts. Following ingestion of either tissue cysts or oocysts, protozoan forms of *T.*

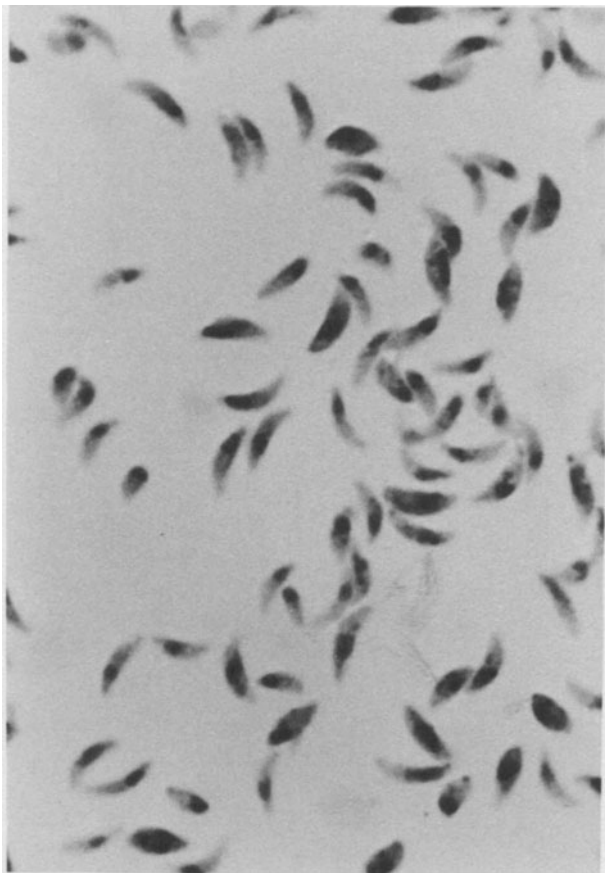

FIGURE 5. Trophozoites of *Toxoplasma gondii* as readily stained by either the Wright or Giemsa technique.

gondii invade the GI tract of the cat, and an asexual cycle (schizogony) is followed by a sexual cycle (gametogony) that results in the development of a noninfectious, nonsporulated oocyst. Cats excrete vast numbers, in excess of millions, of oocysts for approximately 2 weeks. Sporogony occurs outside of the feline host over a period of approximately 5 days and results in an infectious oocyst. These oocysts are highly resistant to physical destruction and remain infectious for more than a year. Ingestion of oocysts has been shown to transmit toxoplasmosis, and oocysts probably play an important role in orally acquired infection. Feline animals, including both domestic and feral cats, appear to be the definitive hosts in the life cycle of the *Toxoplasma* organism, and no other animal has been found to shed oocysts. Transmission between cats and a wide variety of omnivorous, herbivorous, and carnivorous animals occurs via the ingestion of oocysts or ingestion of cysts present in meat. Animals such as cattle, pigs, and birds (including poultry) thus are intermediate hosts that are infected either by congenital transmission, ingestion of oocysts, or through the ingestion of other animals whose flesh contain cysts. Humans acquire the infection primarily by eating the flesh of animals that contain cysts or ingestion of cat excreta contaminated with oocysts.

Surveys of meat available in butcher shops have demonstrated that as many as 10% of mutton, 25% of pork, and less than 1% of beef samples contain tissue cysts that can serve as source of infection if consumed raw or poorly cooked. It is possible that man becomes infected by oocysts through the contamination of human food by coprophagous insects such as cockroaches and flies. The congenital form of disease results when women acquire the acute infection during pregnancy, with the fetus acquiring infection pari passu by the transplacental route. It does not

appear that toxoplasmosis acquired prior to pregnancy can lead to congenitally infected offspring. A small number of infections have been definitely traced to laboratory accidents or by transfusion of whole blood or WBCs. By a variety of serologic techniques, it has been demonstrated that the acquisition of antibodies by humans increase with age, and by midlife an average 50% and as many as 93% of patients surveyed in certain geographic areas have evidence of infection.[140]

Following the usual acquisition of infection by the oral route, *T. gondii* spreads hematogenously to all major organs and tissues. In normals, the generation of host immune response coincides with the disappearance of trophozoites from tissue, and the tissue cyst forms of *T. gondii* take place, i.e., encystation occurs. These cysts contain organisms that are viable for the remainder of the life of the host. Preferred sites of encystment include skeletal and cardiac muscles as well as brain, but tissue cysts may be found in just about every organ. Some investigators believe that monocytes or macrophages can act as a sanctuary for viable trophozoites.

5.3. Pathogenesis

The basic mechanisms underlying host susceptibility to toxoplasmosis have become more clearly defined as a result of both experimental and in vitro studies.[148,149] Evidence for the importance of T-cell-mediated delayed hxpersensitivity immune mechanisms has been presented by Frenkel who demonstrated the ease of infecting nude or congenitally athymic mice with *Toxoplasma*.[150] Unquestionably, therapy with agents that impair T-cell function such as glucocorticoids can reactivate latent *Toxoplasma* infection. Three decades ago Frenkel demonstrated that corticosteroids and irradiation exacerbate *Toxoplasma* infection in hamsters.[151] Such animals develop multifocal central nervous system lesions similar to that observed in immunocompromised humans. Subsequent studies by Strannegard and Lycke have shown that antilymphocyte serum prolongs parasitemia in mice and decreases survival time of infected animals.[152]

These experiments parallel what is observed in man: the tendency for toxoplasmosis to reactivate in patients with Hodgkin disease or AIDS whose T-cell function is especially impaired. More than one-third of patients summarized in Table 5 had Hodgkin disease. Moreover, disseminated disease caused by *T. gondii* appears to occur almost exclusively in patients with underlying hematologic malignancy, recipients of immunosuppressive therapy, or patients with AIDS. Tumors infiltrating tissues that contain cysts seem to have little risk of reactivating infection. Interestingly, some antineoplastic/immunosuppressive agents such as the corticosteroids and cyclophosphamide seem more able to cause recrudescence of latent *Toxoplasma* infection than others, such as vinblastine or bleomycin.

The state of knowledge about specific components of host defense mechanisms against *Toxoplasma* has been enhanced by recent in vitro studies of monocyte–macrophage T-cell interactions. Both experimental mice and humans infected with *Toxoplasma* have stimulated (''activated'') macrophages that inhibit or kill *Toxoplasma* in vitro.[153–155] Human monocytes and monocyte-derived macrophages from normal (unsensitized, nonactivated) or chronically infected patients are unable to kill *Toxoplasma* in vitro. The mechanism by which these macrophages become microbicidal is now being defined. A variety of lymphokines or cytokines, such as γ-interferon (IF_γ) and interleukin-2 (IL-2) are potent activators of mononuclear phagocytes to kill *Toxoplasma*.[156,157] The actual microbicidal mechanisms can be oxygen dependent or independent, depending on specific cell type.[158] The intrinsic susceptibility of the nude mouse to toxoplasmosis gives credence to the primary role of the lymphocyte in the immune process.[150]

lt is not known whether the armed or activated macrophage is solely responsible for the efferent limb of the immune process. Stimulated mouse macrophages kill different organisms (other than *Toxoplasma*) equally well, so there is evidence that the efferent process is non-organism-specific. The presence of *Toxoplasma* organisms intracellularly prevents fusion of the phagolysosome in both normal and stimulated macrophages; therefore, such fusion does not account for inhibition or killing of *Toxoplasma* organisms. Furthermore, macrophage stimulation does not always correlate in time with protection of the host.[159] Attempts to enhance cell-mediated immune function by bacille Calmette-Guérin (BCG) immunization or transfer factor are of undetermined validity in prophylaxis or treatment of toxoplasmosis, and other approaches toward immu-

TABLE 5. Cases of Toxoplasmosis in the Compromised Host—Excluding AIDS[a]

Underlying condition	Cases	Major neurologic presentation or pathology	Treated	Improved
Hodgkin disease	38	25	12	10
Non-Hodgkin lymphoma	10	6[b]	1	0
Leukemia	15			
Acute lymphocytic leukemia	4	1	3	2
Chronic lymphocytic leukemia	4	2	1	1
Acute myelogenous leukemia	2	1	1	1
Chronic myelogenous leukemia	5	1	2	2
Myeloid metaplasia	1	0		
Multiple myeloma	2	2		
Carcinoma				
Breast	3	2	1	1
Ovary	1	0		
Lung	1	1		
Seminoma	1[c]	1[c]		
Melanoma	1	1		
Chromophobe adenoma	1	1		
Neuroblastoma	1	0		
Thymoma	1	1		
Collagen–vascular				
Systemic lupus erythematosus	3	3		
Scleroderma	2	2	2	2
Autoimmune hemolytic anemia	1	0		
Organ transplant				
Bone marrow	9	8	2	1
Kidney	8	6		1
Liver	1	0		
Heart	5	1	3	1[d]
Total	120	65	28	22

[a]Updated from Ruskin and Remington 162
[b]Includes one case with uveitis
[c]This patient also had chronic lymphocytic leukemia
[d]This patient improved secondary to withdrawal of immunosuppression

nologic stimulation or reconstitution have not been investigated.

Toxoplasma infection does not seem to be as common in congenital immunodeficiency syndromes as *Pneumocystis carinii*, and it is not as common in multiple myeloma as in Hodgkin disease despite the tendency to use corticosteroids so commonly in myeloma. This underscores the apparent importance of cell-mediated immune function in defense against this intracellular parasite. The proclivity for *Toxoplasma* to infect the central nervous system in immunocompromised patients suggests that host factors active against *Toxoplasma* are poorly operative in the CNS.

5.4. Signs and Symptoms of Toxoplasmosis in Immunologically Intact Patients

There are three well-recognized forms of toxoplasmosis in the normal host: lymphadenitis, uveitis, and congenital infection. Without doubt, the great majority of congenital and acquired infections in patients without immunodeficiency is inapparent. Clinical disease results from either newly acquired infection or from reactivation of latent infection, since organisms remain viable for the life of the host. The latter mechanism appears to be quite common in the uveitis syndrome. Uveitis in the immunologically intact adult is almost always the result of reactivation

of congenital infection rather than of acquisition of a new infection.

In congenital toxoplasmosis, the maternal infection is usually inapparent. Furthermore, not all mothers who acquire toxoplasmosis during pregnancy produce infected offspring; recently published studies suggest that no more than one-third of maternal infections result in congenital infection. There is good evidence that transplacental passage increases with gestational age, but the incidence of clinical disease appears to decrease. Infants with congenital toxoplasmosis may overcome their infection without sequelae, but others may develop chorioretinitis, ocular muscle problems, epilepsy, blindness, psychomotor seizures, and mental retardation. Clinical manifestations of neonatal infection include chorioretinitis, hydrocephalus, microcephaly, cerebritis with sequelae of complications and convulsions, lymphadenopathy, fever, hepatosplenomegaly, jaundice, rash, cerebrospinal fluid (CSF) pleocytosis, and elevated protein.

Asymptomatic lymphadenitis is the most common clinical manifestation of acquired toxoplasmosis. Nontender, discrete lymph nodes are most often enlarged in the cervical areas, but generalized enlargement may be present. Symptoms such as fever, sore throat, myalgias, and malaise may lead to confusion with infectious mononucleosis. A maculopapular rash and hepatosplenomegaly may mimic cytomegalovirus (CMV) infection. Much less common involvement includes pneumonitis, encephalitis, hepatitis, and polymyositis. Pericarditis is possibly more common than myocarditis, and the latter is actually quite rare.[160]

The lymphadenopathic form of toxoplasmosis, when accompanied by systemic symptoms of fever, malaise, and the findings of hepatosplenomegaly, is often suggestive of a lymphoma. There are many anecdotal accounts of patients referred to major cancer treatment centers for therapy of lymphoma or actually started on antineoplastic chemotherapy who were subsequently found to have only toxoplasmosis. Enlarged lymph nodes should be biopsied unless there is an obvious contraindication. Dorfman and Remington believe that lymph nodes, when infected with *Toxoplasma,* exhibit virtually pathognomonic architectural changes.[161] These changes include (1) reactive hyperplasia associated with irregular clusters of epithelioid histocytes in cortical and paracortical zones, and (2) focal distention of subcapsular and trabecular sinuses by monocytoid cells. In a series of cases studied by these investigators, every patient who had *Toxoplasma* lymphadenitis by histologic criteria also had serologic findings (including Sabin–Feldman dye tests, IgM immunofluorescent antibodies, or both) that corroborated the histopathologic diagnosis.[161]

5.5. Toxoplasmosis in the Compromised Host

Infection in the immunocompromised host can clearly result from reactivation of latent infection, but exogenous or neoacquisition of the organism can produce an equally devastating illness. In a previous review, some 81 published and unpublished cases of toxoplasmosis occurring up to 1975 in patients with neoplasia, collagen vascular disease, and recipients of organ transplants were summarized.[162] Table 5 is an attempt to update that summary with additional cases reported during the ensuing 9 years.[163–175] In both the original analysis and in this updated table, it is apparent that neurological manifestations predominate in more than half the patients.

In patients with AIDS, the major clinical findings are almost exclusively limited to the CNS.[136,137] Lymphadenopathy as part of AIDS itself or the clinical prodrome known as AIDS-related complex (ARC) is rarely caused by toxoplasmosis. Toxoplasmosis has been increasingly recognized in recipients of bone marrow transplants[176–178] and in these intensely immunosuppressed patients parasitemia has been documented.[176] In the compromised host, clinical manifestations include diffuse encephalopathy, meningoencephalitis, enlarging central mass lesions, or a combination thereof. Focal signs, seizures, motor impairment, and impaired consciousness are common clinical findings. Examination of the spinal fluid has shown mild reactive pleocytosis with a predominance of mononuclear forms and only a modest elevation in protein. Many patients have had concurrent infections with DNA viruses, and such individuals have usually been more severely ill. Toxoplasmosis may coexist with a neoplastic disorder (particularly lymphoma) and may be confused with a metastatic process in the liver, spleen, lungs, or CNS.[173,174] Noninvasive techniques such as radionuclide scanning,[175] computed

tomography (CT), or magnetic resonance imaging (MRI) are expeditiously required to establish the possibility of infection mimicking tumor. Rarely, now, is arteriography required to distinguish between a vascular mass such as a tumor, and a toxoplasmic abscess. In our experience, mediastinal lymphadenopathy and eosinophilia are not useful in distinguishing lymphoma (e.g., Hodgkin disease) from toxoplasmosis.

Besides newly acquired (per oral) infection or reactivation of dormant infection, toxoplasmosis can be acquired by transfusion of blood products. Siegel and associates described four patients with acute leukemia who developed acute toxoplamosis after receipt of leukocytes from donors with chronic myelogenous leukemia.[179] On the basis of retrospective serologic studies, evidence was obtained implicating donor leukocytes as the source of the parasite. In vitro studies indicate that *T. gondii* remains viable in banked blood for up to 50 days at refrigerated temperatures and that the parasite can be isolated from the buffy coat of totally asymptomatic individuals. Other than the experience of Siegel and colleagues,[179] there are no other well-documented examples of hematogenous or transfusion-associated toxoplasmosis. On the other hand, Raisanen and colleagues have described an experimental model wherein whole human blood inoculated with *Toxoplasma* and stored up to 28 days caused lethal infections in rabbits after transfusion.[180] Thus, transfusion-associated toxoplasmosis appears to be real, but the risk of disease seems very small. It seems impractical at this time to recommend screening of blood products for evidence of prior or recent *Toxoplasma* infection. Correspondingly, some authors have suggested that only *Toxoplasma*-antibody-negative subjects serve as donors of organ allografts to antibody-negative recipients but that such a policy would not appear to be judicious if the recipient had evidence of prior infection (i.e., seropositivity). Toxoplasmosis involving both donor and recipient hearts has been reported after heterotopic cardiac transplantation.[181]

There is little specific information on the incidence of uveitis in the immunocompromised host, and no solid data on the proportion of cases caused by *Toxoplasma*. The differential diagnosis of uveitis in the immunocompromised patient really does not differ from that in the patient with intact host defenses. Thus, in patients with appropriate geographic background, histoplasmosis must be considered high on the list of infectious etiologies causing similar fundoscopic and clinical findings. It can be stated with certainty that uveitis appears to be relatively less common than intracerebral involvement in the cases reviewed in Table 5.

5.6. Toxoplasmosis in AIDS Patients

It is well established that patients with AIDS have a predilection for the development of toxoplasmic encephalitis.[136,137] The risk of CNS disease in patients with positive toxoplasma serology may be as high as 12%.[137] In a study of AIDS patients by Snider and colleagues, *T. gondii* accounted for 38% of known CNS infections and 36% of focal intracerebral lesions.[182] In another hospital study, toxoplasmic encephalitis was the leading cause of death among Haitian patients who were autopsied.[183] In Haitian patients, concomitant infection due to *M. tuberculosis* is common, as are a host of other infectious problems. Other conditions that may be confused with *T. gondii* involving the central nervous system are infections due to *Candida, Aspergillus, Cryptococcus neoformans, Mycobacteria,* progressive multifocal leukoencephalopathy, intracerebral lymphoma, or Kaposi sarcoma. Focal abnormalities are present in most patients and seizures either of a generalized or focal nature may be a presenting finding. Most patients with AIDS have infection localized to the brain, but disseminated disease has also been reported. The presence of intracerebral mass lesions, particularly in the basal ganglia region are quite suggestive of the diagnosis. Since there may be a number of absolute or relative contraindications to brain biopsy, many authorities recommend empiric treatment for toxoplasmosis of CNS mass lesions, even if the diagnosis of *Toxoplasma* infection is not firmly established. The rationale for this recommendation is that the other entities likely to cause a mass lesion(s) are probably not as readily treated and will not respond quickly to therapy. Once successful treatment of the acute episode occurs (and the response may occur within days, particularly when accompanied by a short course of corticosteroids to decrease cerebral edema), it would seem prudent to place the patient on chronic suppressive therapy with

pyrimethamine (25 mg/day) and probably a sulfonamide preparation (2–4 g/day) if the patient can tolerate one of the latter group of agents.

Serodiagnosis of *Toxoplasma* encephalitis in AIDS patients presents a major challenge. Most patients with proven disease do not have elevated *Toxoplasma* titers and seroconversion is infrequent, in marked contrast to the situation in nonimmunosuppressed patients. Parasitemia has been documented in AIDS patients.[184] Some have reported that the *Toxoplasma* agglutination test is a more sensitive indicator of acute infection, particularly in patients with low dye test titers. Remington and colleagues recommend that sera from patients in high-risk groups for AIDS be stored to facilitate specific diagnosis if CNS disease develops later. At that point, changes in titers can be sought.[185] When used in conjunction with the dye test, titers obtained by the agglutination test seem to be the most useful noninvasive indicator of toxoplasmic encephalitis.[186]

5.7. Diagnosis

There are several pitfalls in the accurate diagnosis of clinically significant toxoplasmosis in both the immunocompromised and normal host. Stringent, unequivocal criteria on which to base such a diagnosis include (1) demonstration of trophozoite forms in body fluids or tissues and/or (2) the finding of *Toxoplasma* antibodies of specific type and titer in serums (and/or body fluid).

Tissue culture isolation techniques have been used to document parasitemia in patients with toxoplasmosis complicating marrow transplantation[176] and AIDS,[184] and this would provide strong evidence of disseminated disease. However, isolation per se of the parasite following inoculation of human tissue into a susceptible laboratory animal or tissue culture does not provide definitive proof of acute infection because cysts remain viable in such tissues as lymph node, muscle, heart, and brain for life. However, in view of the rarity of cyst forms in enlarged lymph nodes, the isolation of viable *Toxoplasma* from such nodes might be meaningful when combined with the data from serologic studies. Organisms may even be recoverable from blood of asymptomatic patients more than 1 year after initial infection. By the same token, histologic demonstration of the cyst form of the parasite in tissue does not altogether eliminate the possibility that infection is still acute. During the course of active disease, there may be coexistence of cyst forms with trophozoites in the same organs.

There is a growing literature on the serodiagnostic approaches to *Toxoplasma* infection, but no consensus about the most reliable test. Table 6 sum-

TABLE 6. Serodiagnostic Tests for Toxoplasmosis

Test	Acute	Chronic/latent	Comment	Reference
Agglutination	≥1 : 100	1 : 16–1 : 256	For IgG antibodies	186
Complement fixation	≥1 : 32	neg—1 : 8		187
Enzyme-linked immunosorbent-assay (ELISA)	≥1 : 512	≤1 : 256		188
IgM capture	>1 : 256	neg—1 . 64	Lower titers in infants	189
IgM immunoabsorbent				190
Fluorometric immunoassay	>100 signal units			191
Hemagglutination, indirect	≥1 : 100	1 : 16–1 : 256		192
Immunofluorescence				
Antiglobulin second antibody	≥1 : 1000	1 : 8–1 : 1000		193
Anti-IgM second antibody	≥1 : 80[a]	neg after 4 months	Earliest positive, correlates best with acute infection	194
Sabin–Feldman dye uptake	≥1 : 1000[b]	1 : 4–1 : 1000	Requires viable organisms	144

[a]IgM titers ≥1 80 highly suggestive of acute infection
[b]IgG titers 1 16,000 suggestive of acute infection

marizes the commonly used serologic tests for toxoplasmosis with a summary of the ranges of titers in acute disease and the titer ranges seen in chronic or latent infection.[185–194] As with all serologic tests, the most important principle is that the diagnosis of acute toxoplasmosis is established by a rising titer in serial specimens: either from negative to positive or at least a fourfold rise in titer. One of the major problems with the commonly available serodiagnostic methods (Sabin–Feldman dye test, indirect immunofluorescence, indirect hemagglutination, and ELISA), is that titers may remain elevated years after acute infection. Although some advocates of the complement-fixation test claim that titers of 1:16 or greater reflect acute disease, we believe that elevated titers in this range may also persist for years.

Often a patient may be tested for evidence of toxoplasmosis after the peak of symptoms. It may be of value to perform several serodiagnostic tests simultaneously. The earliest test to become positive is one that detects IgM-specific anti-*Toxoplasma* antibodies, followed by the Sabin–Feldman dye test, the indirect hemagglutination test, and the complement fixation test. The IgM–IFA test has been proposed as the most accurate test of acute infection,[193] but problems with this assay have been encountered by several other laboratories. These interlaboratory variations could be because of differences in the fluorescein-labeled anti-IgM antiserum.

IgM detecting methodologies have also been proposed as the most reliable means for distinguishing acute from chronic infection,[189,190,194] but there are notable exceptions to this premise.[185] Patients with AIDS or a disseminated neoplasm may not mount a humoral antibody response.[195] The converse of this conclusion is that some compromised hosts (e.g., heart transplant recipients) may demonstrate significant rises in IgM antibody titers without clinical evidence of active infection.[185] The best working principle is that a detectable immune response, particularly a change in antibody concentrations, is one piece of evidence in establishing a diagnosis, but the absence of change in antibody levels does not exclude the diagnosis of toxoplasmosis.

Several experimental tests have been described that report the detection of free *Toxoplasma* antigens in serum[196,197] or body fluids.[198] Such approaches could be immensely helpful in clarifying confusing clinical and laboratory findings.

Studies for mononuclear–lymphocyte reactivity as well as tests of delayed hypersensitivity have no value in the diagnosis of acute infection in the immunocompromised host. Although lymphocyte transformation occurs in congenitally infected infants,[199] skin tests become positive months after infection and therefore cannot diagnose newly acquired acute infection. Furthermore, such tests are not helpful in evaluating reactivated disease and could well become negative secondary to the effects of immunosuppressive treatment.

5.8. Summary of Diagnostic Approach to Possible CNS Toxoplasmosis

Since CNS involvement is a life-threatening complication, the following steps are reasonable diagnostic measures to be undertaken with celerity.

1. Lumbar puncture should be taken for (a) routine studies (the cell count should be primarily mononuclear, the protein may be modestly elevated, but the glucose may be normal); (b) cytology (the CSF sediment or better yet a cytocentrifuge preparation should be stained with Giemsa and examined by immunofluorescence using *Toxoplasma*-specific antibodies; (c) CSF inoculation into mice (fresh or refrigerated samples should be inoculated into mice intraperitoneally; freezing of the sample will kill trophozoites and should be avoided); and (d) serology (which should be performed and compared to serum titer).
2. Serum serology should be compared with CSF titer. Desmonts[200] proposed a method for assessing antibody activity in the aqueous humor of the eye (for diagnosing ocular toxoplasmosis) by factoring for IgG concentration. The following equation was used.

$$C = \frac{\text{aqueous humor titer}}{\text{serum titer}} \times \text{globulin concentration} \left(\frac{\text{serum}}{\text{aqueous humor}} \right)$$

If $C \geq 8$, antibody production is local. Applied to CSF (in place of aqueous humor), the diagnosis of toxoplasmosis in the CSF should be considered if the titer/IgG is greater in CSF than in serum.

3. CT scan of brain and/or brain scan should be performed.

Definitive diagnosis would be established by brain biopsy if the patient has a mass lesion in an accessible area and there are no contraindictions to surgery. Examination of the specimens by both light and electron microscopy and by immunoperoxidase staining is desirable. However, we would be inclined to initiate empiric therapy (see Section 5.9), depending on the clinical situation (essentially, how unstable the patient's overall condition is) and on the basis of positive serology.

5.9. Therapy

The decision to treat must obviously be based on the constellation of clinical findings, the underlying severity of the patient's illness and a careful weighing of the benefits of drug therapy in view of the limitations and toxic effects of the agents that can be used. There is little question, however, that few clinicians should hold back in therapy if the patient demonstrates CNS involvement, pericarditis, hepatosplenomegaly, or pneumonitis in the presence of elevations in any of the antibody titers listed on Table 6. A more important issue, however, is whether empirical anti-*Toxoplasma* therapy should be started in patients who are awaiting confirmation of the diagnosis by serology, since such specimens are often referred to specialized laboratories, and delay in obtaining results may be experienced. Our policy has been to start empiric therapy in the patient with underlying lymphoma, in recipients of high-dose corticosteroid therapy, and in the individual with AIDS who has evidence of a CNS mass lesion that does not appear to be a tumor by noninvasive studies. There would appear to be little risk associated with pyrimethamine and sulfadiazine in this situation (which would also "cover" infections such as *Nocardia*), and if the patient were to have a brain abscess, the cure would likely require surgical extirpation.

No antimicrobial agent is effective against a tissue cyst form of *T. gondii*. Pyrimethamine, a diaminopyrimidine, and sulfonamide are both active against the trophozoite form and interact synergistically when used in combination. These agents are antagonists of dihydrofolic acid reductase and *p*-aminobenzoic acid, respectively. Both agents are readily absorbed from the GI tract and penetrate lipids and lipid cells in the central nervous system. There is conflicting information about the efficacy of trimethoprim plus sulfamethoxazole as an anti-*Toxoplasma* agent, but recent studies in animals and in tissue culture suggest that it is effective.[201] Unfortunately, human clinical data are lacking, and at this point we do not advise that this agent be used for confirmed *Toxoplasma* infection. The author has seen *Toxoplasma* encephalitis develop in AIDS patients who had recently been given trimethoprim–sulfamethoxazole for pneumocystosis.

For seriously ill patients, a loading dose of 100–200 mg pyrimethamine is recommended during the initial day of treatment in adults followed by a maintenance daily dosage of 1 mg/kg per day. For children, 2 mg/kg should be given as a loading dose for the first 3 days of treatment. There is no problem with the administration of folinic acid (calcium leukovorin) orally or intramuscularly in doses of 2–10 mg/day in conjunction with pyrimethamine to obviate the effect of pyrimethamine on bone marrow DNA synthesis. Fortunately, neither folinic acid nor Baker's yeast (unlike folic acid) inhibits the action of pyrimethamine on *T. gondii,* making this an ideal form of chemotherapy. Reservations about giving folinic acid stem from the concern that in some hematologic malignancies (e.g., leukemia) it might cause proliferation of neoplastic cells. Sulfadiazine or a triple sulfa mixture are the most active sulfonamides against *T. gondii,* whereas other sulfonamides such as sulfisoxazole are less active, and their use should be avoided. Dosage of sulfadiazine in adults is 100 mg/kg per day divided into four hourly doses after a loading dose of 50 mg/kg. For infants, the dosage is 100–140 mg/kg per day PO in four equal portions after a loading dose of 100 mg/kg.

There is little information on optimal duration of pyrimethamine–sulfa therapy, but a month may be adequate. Patients with chronically impaired host defenses, such as AIDS, may require life-long suppressive therapy.

A number of other antimicrobial agents have been investigated for anti-*Toxoplasma* activity, and

two, clindamycin and spiramycin, have been found active.[185] There are now several reports of successful use of the combination of clindamycin in very high dose (e.g., 1200 mg 3–4 times per day) plus pyrimethamine in the usual dose to treat patients with AIDS.[185] This approach remains experimental and should only be attempted as a last resort in patients with severe hypersensitivity to sulfonamides.

For the patient with uveitis, the additional benefit of corticosteroids given either topically or systematically has been debated, with proponents advocating their use for the antiinflammatory effects and detractors arguing that their use further aggravates host immune defects. Certainly, it is folly to use steroids alone in the treatment of uveitis,[202] and if steroids are employed, they should be given as an adjunct to appropriate anti-*Toxoplasma* treatment.

5.10. Prevention

For seronegative patients perhaps the most appropriate measure is adequate cooking of meat to temperatures in excess of 60°C. Avoidance of cats or areas contaminated with cat feces is probably commendable but of dubious practicality. Also, the determination of the antibody seropositivity of a pet cat is valueless in excluding a potential hazard. Theoretically, avoidance of donors of blood products who are seropositive may be desirable, but is not likely to be feasible; this also applies to selection of donors of organ transplants. Long-term antimicrobial prophylaxis of toxoplasmosis has not been studied in the immunocompromised host, and, indeed, there are appropriate concerns about the effects of such agents on the bone marrow in certain underlying diseases. Prophylaxis might be considered in a clinical situation characterized by high risk of active infection as has been defined for pneumocystosis, but such a situation has not presented itself for study to date.

An unanswered but important question is whether or not all patients with lymphoma or leukemia or prospective organ transplant recipients should be screened for anti-*Toxoplasma* antibodies and possibly given "preventive therapy" if seropositive. Such serologic screening should be part of a routine initial procedure prior to therapy or transplantation (if only to establish a baseline for future comparison). If a patient has elevated or equivocally elevated titers, preventative therapy will probably be of little risk but remains of unproven efficacy. Many patients with neoplastic diseases, recipients of organ transplants, and patients with AIDS receive prophylaxis with trimethoprim–sulfamethoxazole against *P. carinii* infection, and it will be interesting to see whether this has any impact on the incidence of toxoplasmosis.

6. Coccidial Infections

Two agents belonging to the subclass Coccidia within the class of sporozoal parasites have become prominent since 1980 as infectious complications occurring in immunocompromised hosts. These agents are *Cryptosporidia* and *Isospora belli*. Technically, these intracellular parasites are related to the *Toxoplasma* group—tissue cyst-forming coccidial organisms that belong to the suborder *Eimeria*. Both agents can now be readily identified by modified acid-fast staining of fecal specimens (such as the Kinyoun stain), but other more elaborate identification techniques are available.[203] In fact, together they constitute the only acid-fast staining parasites of any clinical significance. The two species can be readily distinguished. Features for *I. belli* include its large size (15 × 25 μm), which characteristically contain two sporoblasts (Fig. 6). In contrast, *Cryptosporidia* oocysts are much smaller (average 5 μm in diameter) and they contain four sporozoites (Fig. 7). However, the small size of *Cryptosporidia* is one reason that they may escape detection unless a special effort is made at diagnosis.

Most clinical attention has been focused on *Cryptosporidia,* a protozoan parasite that completes its life cycle closely adherent to the intestinal surface epithelium of mammals, birds, and reptiles. Cryptosporidiosis was first noted as a human infection following symptoms of overwhelming watery diarrhea in an immunosuppressed host.[204] The clinical prominence of this organism, as well as that of *I. belli,* has been mainly due to the role both have played as causes of severe diarrhea in patients with AIDS. Typically, the diarrhea is watery, profuse, associated with cramps and bloating, and results in severe malabsorption. Perhaps the first report of cryptosporidial disease in an AIDS patient was published in 1981.[205] It concerned a 48-year-old homosexual man treated in December 1979. This patient may well have been one of the earliest cases of AIDS, and the relationship

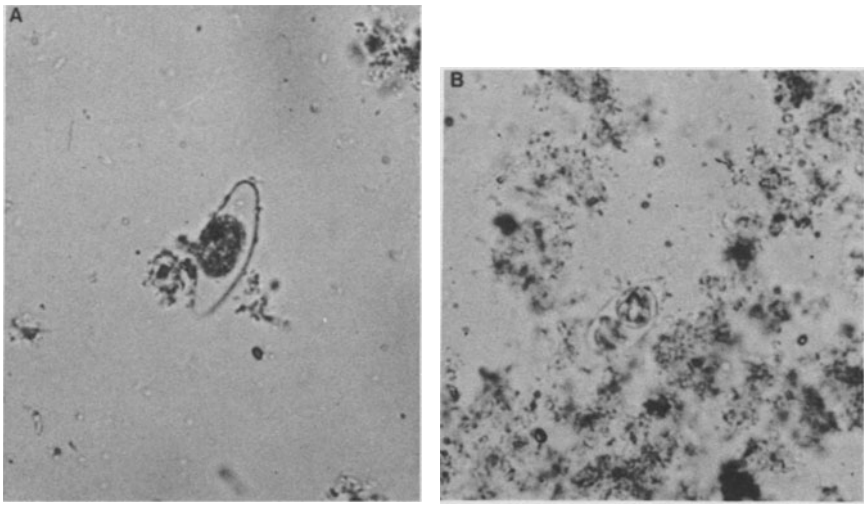

FIGURE 6. The oocysts of *Isospora bella* have a large, distinctive oval shape (25 × 15 μm) containing two intracellular sporoblasts. (A) The latter structures overlap. (B) The latter structures are distinct. (Photos courtesy of Lynne Garcia, UCLA Clinical Microbiology Laboratory.)

of the pathogen to the underlying immunopathologic disorder was not appreciated at the time. As with many subsequent cases, that patient's illness could not be controlled with a variety of antibacterial, antimalarial, and other antiprotozoal medications. Terminally, the patient developed a disseminated CMV infection and died.

Subsequent to the description of cryptosporidiosis in AIDS patients, worldwide attention has focused upon the disease as it occurs in animals.[206] By far and away, this seems to be its greatest importance, particularly in terms of economic ramifications. Besides patients with AIDS, individuals with other types of therapeutic immunosuppression or underlying disorders characterized by defects in cell-mediated immunity are now being described as having cryptosporidiosis: transplant recipients, patients receiving corticosteroids, and patients with IgA nephropathy.[207] With increasing interest focused on this pathogen, more extensive studies of diarrheal illnesses have been carried out with a specific interest in this pathogen. Not surprisingly, this organism has been increasingly detected in stool specimens of normal healthy patients.[208,209] Cryptosporidial disease now appears to be a common cause of diarrhea in developing countries,[210,211] but the reason it has not been more prominent in the clinical literature now seems clear.

First, the normal host is able to overcome or control the infection and the illness is short and self-limited. Recurrent disease has actually been documented in veterinarians,[212] and the evidence for a lasting immunity to this infection has not been established. Secondly, the diagnosis has not been frequently made before the current epidemic of AIDS because the organism is rather small and has not been readily detected by many of the procedures previously used to screen stool specimens. However, when appropriate techniques are used, the organism is identified without difficulty. Extraintestinal cryptosporidiosis has been reported. Ma and colleagues[213] reported three cases of interstitial pneumonia with this pathogen, but such occurrences are uncommon.

Illustrative Case 3

The patient was a 42-year-old homosexual man who had been in generally good health all his life, except for several episodes of gonorrhea and one episode of hepatitis B. Following a slow convalescence from hepatitis B, the patient then began to notice an onset of increasing watery diarrhea, weight loss, and low-grade fever Over a period of 6 weeks he became severely anorectic with bloating, diffuse abdominal cramps, and severe anorexia Except for clear liquids and gelatinlike foods, he was unable to eat. After approximately 3 months of watery diarrhea and a 30-lb weight loss, he sought medical attention. Several examinations of the stool revealed the acid-fast oocyst of *Cryptosporidia* species The

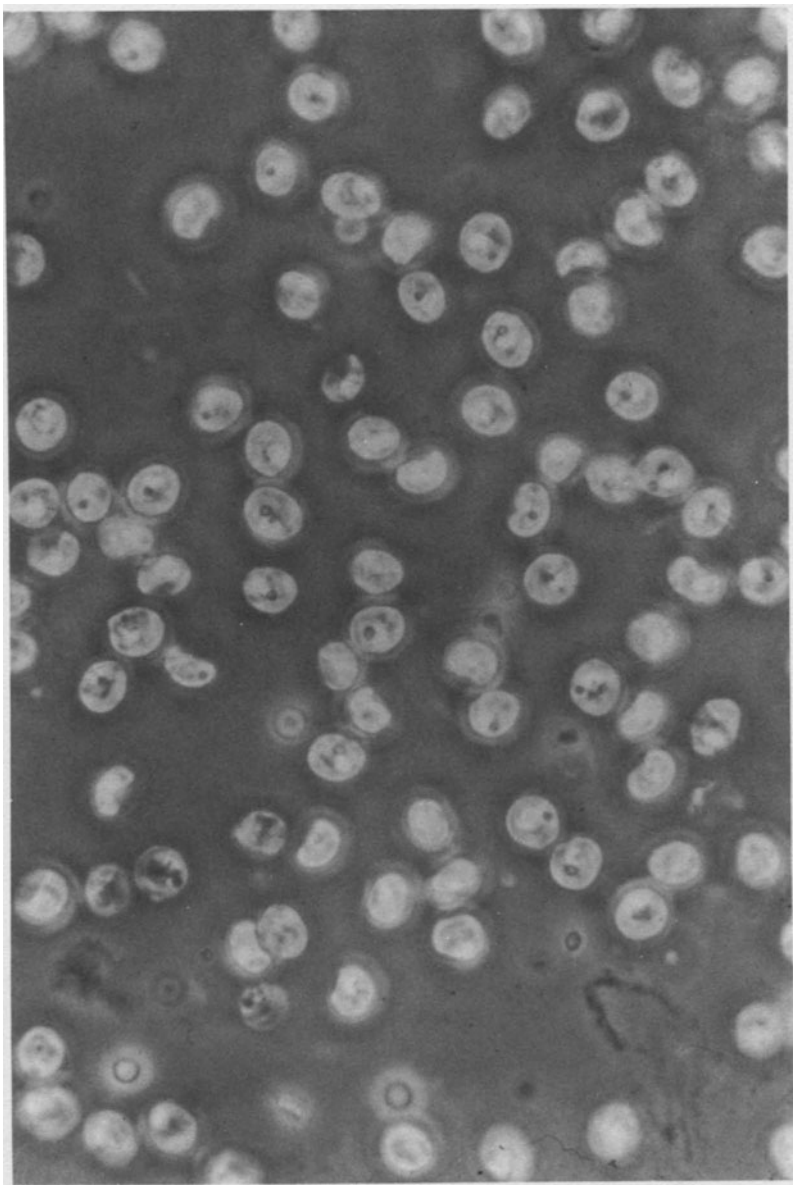

FIGURE 7. Like *Isospora, Cryptosporidia* are acid-fast but much smaller (4–6 μm in diameter) and roundly shaped. They may be tightly adherent to intestinal epithelial surfaces. (Photo courtesy of Lynne Garcia, UCLA Clinical Microbiology Laboratory.)

patient was treated with a variety of antidiarrheal agents, including bismuth-containing compounds, and opiate-containing medications without success A program of systemic hyperalimentation through a Hickman catheter was initiated This had no effect on the diarrhea (characterized by production of up to 20 thin, watery, explosive, diarrheal stools per day) but did result in some modest gain in weight. Serum albumin gradually rose and liver function tests, improved slightly. In an attempt to control the diarrhea, a variety of medications were used. This included chloroquine and clindamycin plus quinine. There was modest response to the latter combination but the latter two agents were extremely unpalatable. The dose of quinine and clindamycin was varied somewhat, but the patient was unable to take his medications consistently. Finally, naproxen was given in a dosage of 200 mg four times per day. Within about 24 hr there was a dramatic reduction in stool output. The patient was able to gain considerable symptomatic relief with naproxen and subsequent stool examination using a modified acid-fast stain identified no cryptosporidial organisms Some half-dozen follow-up stool examinations were carried out. When naproxen was discontinued, however, there was a return of watery diarrhea, cramping and bloating. Despite some stabilization on naproxen and hyperalimentation, the patient continued to lose weight and then developed low-grade fevers and lung infiltrates The infiltrates were consistent with *Pneumocystis carinii* pneu-

monitis, and the patient was treated empirically with trimethoprim–sulfamethoxazole. Lung infiltrates responded to this empiric therapy. Subsequently, however, he developed disseminated infection due to *Cryptococcus neoformans* with meningitis and positive blood cultures. The patient subsequently expired while being treated with amphotericin B, and no autopsy was obtained.

Comment. This patient presents with a fairly typical clinical progression of infections as seen in AIDS patients. Other patients with cryptosporidial or *I. belli* infection that we have encountered since the beginning of the outbreak have had marked weight loss, fluid and electrolyte loss, and severe inanition. The organism was missed on initial stool examinations but was subsequently identified with an acid-fast stain. The case report demonstrates that the major clinical management problem is fluid and electrolyte loss and it may become necessary to begin hyperalimentation simply in order to keep that patient alive

With regard to therapy, a variety of symptomatic and perhaps more specific antiparasitic medications were used in this unfortunate patient. Clindamycin plus quinine was tried because of its reported success in other protozoan infections.[103] Interestingly, *Cryptosporidia* were not seen in stool after the clindamycin plus quinine therapy, but a small bowel biopsy and autopsy examination were not performed in order to validate clinical efficacy Unfortunately, this is a major problem of evaluating response to therapy in patients with AIDS or similar immunologic disorders. What did give the patient symptomatic relief was a prostaglandin inhibitor, naproxen. Other medications such as indomethacin had been used with similar success. As has occurred in so many AIDS patients, one serious life-threatening infection such as cryptosporidiosis was supplanted by several other processes and the actual cause of death could not be precisely determined.

With regard to therapy of cryptosporidiosis, there has been some reported success with an experimental macrolide, spiramycin, in terminating the cryptosporidial diarrhea of AIDS patients.[214] However, the overall prognosis is extremely poor Therapeutic responses are somewhat better in patients with *I belli* infection Trimethoprim–sulfamethoxazole therapy is usually rapidly effective with termination of diarrhea in 48 hr,[215] and treatment should be continued for 2–4 weeks Nonetheless, there is a high rate of recurrence, and while long-term suppressive therapy has been advocated for AIDS patients, this may not be feasible because of drug intolerance. Alternative medications include metronidazole and pyrimethamine–sulfadiazine

7. Strongyloidiasis

In healthy subjects, infection caused by the intestinal threadworm *Strongyloides stercoralis* is usually clinically inapparent or manifested by mild GI symptoms of epigastric pain and diarrhea. As with toxoplasmosis, infection can rapidly progress to a fatal outcome in immunocompromised hosts. The causative organism is fairly common throughout the world, and the diagnosis should be suspected in patients with diffuse pneumonitis, eosinophilia, and GI symptoms. Some of the most severe systemic infections caused by this parasite are also associated with sustained or unexplained bacteremia caused by *Enterobacteriaceae* or *P. aeruginosa*. However, the parasitic organism may not be found in routine stool examinations, and infected patients may not have eosinophilia.

7.1. The Organism

Strongyloides stercoralis is an intestinal nematode with a complex life cycle. Three different life cycles may occur at various times. The parasitic female lives in the intestine in a parthenogenic state. Eggs are released and hatched in the mucosa, and rhabdoid larvae are passed in the feces. Similar to what occurs in hookworm, rhabdoid larvae develop in the soil into filariform larvae that can penetrate human skin on contact, migrate through the bloodstream, and pass into the lungs. In the lungs, the larvae break out of the capillaries into the alveoli, migrate up the tracheobronchial tree to the pharynx, and are swallowed, whereby they again colonize the GI tract. A second life cycle involves the transformation of rhabdoid larvae in the soil into free living male and female adults which mate; these eggs hatch into larvae which may become infective for man. The third or so-called autoinfection cycle can take place entirely within the gastrointestinal tract of the host. Rhabdoid larvae molt and change into filariform larvae which are invasive. These filariform larvae can penetrate the intestinal mucosa or the skin of the perianal areas as they are passed in the stool. By this mechanism, clinical systemic infection can occur many decades after a patient leaves an endemic area.

7.2. Clinical Manifestations

Disseminated, occasionally fatal strongyloidiasis may occur in subjects whose immune mechanisms are presumably intact. But the most common setting for systemic infection is one of immunodeficiency resulting from either an underlying neoplastic disease or from immunosuppressive treatment. In those infections initiated by penetration of the skin by filariform larvae, the presence of a rash characterized by erythematous, maculopapular, or serpiginous lesions associated with pruritis is an early manifesta-

tion of infection. Urticaria may also be present. This pruritic reaction occurs directly at the site of larval skin penetration and usually appears within 24 hr. In those patients with autoinfection secondary to GI carriage, such lesions may be seen in the area of the lower abdomen, buttocks, perineum, or upper thighs. These lesions contain filariform larvae. During the early stage of active infection, blood eosinophilia is a common but not necessarily consistent finding. With persistence of the infection, eosinophilia may disappear, but in chronic strongyloidiasis associated with eosinopenia, the prognosis has been poor. Granulomatous reactions of pseudotumors around degenerating larvae or adult worms have been described in the intestine of animals and humans.

In the intestinal phase of strongyloidiasis, the major clinical manifestations are burning or colicky abdominal pain associated with diarrhea. The stool may be watery and contain mucus or blood. In severe infections, a malabsorption syndrome may develop as the patient loses large amounts of fat and protein in the feces. Bowel lesions resembling ulcerative colitis may develop and lead to systemic manifestations including systemic toxicity and fever. Severe malabsorption, dehydration, electrolyte imbalance, and paralytic ileus may ensue. Immunosuppressive therapy and corticosteroids in particular may inhibit cell-mediated immune mechanisms against invading larvae or suppress the humoral antibody response to these antigens. The mechanical effect of a paralytic ileus resulting from other therapy or from an obstructing tumor may alter the host–parasite relationship in favor of the parasite. Massive intestinal infection with *Strongyloides* may itself cause paralytic ileus. Decreased intestinal motility from any cause may result in the 24–48-hr time interval required for the transformation of rhabditiform to invasive filariform larvae which then migrate out of the gut into the bloodstream.

Overwhelming infection by *Strongyloides* has received relatively little attention in the current literature on opportunistic infection. Scowden et al. have reviewed this subject comprehensively and pointed out more than 20 conditions associated with systemic strongyloidiasis, including Hodgkin disease, acute and chronic leukemias, leprosy, nephrotic syndrome, renal transplants, lupus erythematosus, protein-calorie malnutrition, and chronic renal failure.[216] Scowden et al. use the term hyperinfection syndrome to designate massive invasion of the gastrointestinal tract and lungs, whereas disseminated strongyloidiasis is used to refer to extensive parasitic invasion of other tissues. Hyperinfection is an exaggeration of the normal life cycle of the parasite, and patients present with a combination of respiratory and gastrointestinal symptoms. Disseminated strongyloidiasis implies involvement of organs not part of the ordinary life cycle of the parasite. In such an extension of disease, involvement by large filariform larvae can cause direct injury to heart, liver, skeletal muscle, adrenals, pancreas, kidney, and CNS. Because of the ready availability of chest radiography, perhaps the most commonly recognized site of involvement in both the hyperinfection syndrome and disseminated disease is the lung. Hemorrhagic pulmonary infiltrates or diffuse bilateral alveolar infiltrates with or without GI complaints have been reported.[217] Strongyloidiasis has also been associated with the onset of the adult respiratory distress syndrome (ARDS).[218]

In the gastrointestinal tract, severe ulcerating hemorrhagic enterocolitis may serve as a port of entry for bacterial and fungal superinfection. Both the hyperinfection syndrome and disseminated disease may be associated with sustained gram-negative bacteremia, and bacteremic pneumonia may be confused with parasitic infection in the lungs. This pattern of sustained bacteremia should be a clue to consider the diagnostic possibility of strongyloidiasis, particularly in the patient with eosinophilia or abdominal symptoms. Similarly, gram-negative bacillary meningitis can be an important concomitant process. It is not certain, however, that the parasitic infection always directly involves the brain. Meningitis probably results from bacteremia, but several authors have speculated that bacteria are (1) transported through the circulation and the brain in a piggyback fashion, i.e., adherent to the worm itself, and (2) excreted by migrating larvae. The manifestations of CNS disease can range from abnormalities in mental status to stupor and deep coma.

Illustrative Case 4

A 57-year-old Puerto Rican man was diagnosed as having polyarteritis nodosa in 1971. He was maintained on an average

daily dose of 60 mg prednisone for the next 7 years He then developed diarrhea and both a *Salmonella* species, group E, was isolated from stool, and larvae of *Strongyloides stercoralis* was detected in the same specimen. The *Salmonella* was untreated and the *Strongyloides* treated with 1.5 g thiabendazole bid for 2 days. Five months later, he experienced return of fevers and polyarthralgias, and his stool was again positive for *Strongyloides*. A lumbar puncture was performed after mild headache developed and the CSF found to contain 4030 WBC/mm^3 of which half were neutrophils. The protein content of this sample was 180 mg/dl, and the glucose 77 mg/dl. All cultures and serologic tests on the CSF were negative. The patient was treated with 18 million units of penicillin, and his condition gradually improved. On two subsequent occasions, the patient was found to have larvae of *Strongyloides* swarming in his stools. With the second of these episodes, blood cultures were positive for *Escherichia coli* and *Pseudomonas aeruginosa* and the latter was isolated from CSF, which showed an elevated protein and WBC count. The patient responded to intravenous gentamicin and carbenicillin. Before therapy was completed, the patient developed progressive back pain and was found to have compression fractures of the L4 and L5 vertebrae. Tomograms revealed erosive lesions of both vertebral bodies, and bone scan showed increased radionuclide uptake. The diagnosis of vertebral osteomyelitis was confirmed by an open biopsy and recovery of a *P. aeruginosa* with the same antibiogram as the blood isolate.

Comment. Although there is no proof of systemic invasion by the round worm, this case represents a probable example of disseminated infection caused by *Strongyloides stercoralis*. In retrospect, the first episode of bacteriologically sterile meningitis may have represented invasion of the meninges by *Strongyloides*. Polymicrobial gram-negative bacillemia and metastatic bone infection are commonly associated with disseminated *Strongyloides* infection in immunosuppressed hosts.[216] In patients with repeatedly positive stool examinations containing *Strongyloides* and systemic bacterial infections, long-term intermittent suppressive therapy is recommended (Section 7.4)

7.3. Diagnosis

The unambiguous diagnosis of *Strongyloides* involves detection of rhabdoid larvae in feces, secretions (e.g., sputum), or tissue. Freshly passed feces may contain active rhabdoid larvae, but stool may be negative in more than 70% of cases despite careful examination.[217] Refrigeration of stool causes rapid disappearance of larvae. Ova can rarely be detected in stools, and multiple repeat stool examinations may be necessary, since larvae may only appear several weeks after institution of immunosuppressive therapy.[218] Caution must be urged in the examination of older clinical specimens because any hookworm eggs present in the feces for any length of time could hatch and yield rhabdoid larvae that are morphologically similar. If stools are negative, but a strong clinical suspicion remains, aspirated duodenal contents should be examined for the presence of larvae. Recently, greatly improved detection has been observed using the Entero-test method for sampling of duodenal contents (this procedure is described in Section 4.6).

Larvae have also been reported present in sputum, peritoneal fluid, and urine. Tracheal or transtracheal aspiration may be necessary for detection in respiratory secretions, and sputum specimens should preferably be concentrated. Filariform larvae may be present in biopsies of skin lesions. Larvae have also been identified in pleural fluid, peritoneal fluid, lymph nodes, and urine and are best identified by a wet-mount examination and/or Giemsa stain. Eosinophilia exceeding 30% is common in infection with *Strongyloides* in intact hosts and has been present in perhaps half of immunosuppressed patients.[216] Patients with disseminated disease receiving corticosteroids, however, may not have this valuable laboratory clue. Other aspects of laboratory diagnosis, including skin testing and serologic methods, have not been helpful, particularly in immunocompromised hosts.

Radiologic studies of the small intestine may suggest the diagnosis.[219,220] Findings consistent with the diagnosis of Strongyloidiasis include dilated loops of bowel with thickened mucosal folds. These findings reflect worm penetration of the intestinal mucosa, lymphatic obstruction, and allergic edema of bowel wall.

7.4. Treatment

Patients with intact host defenses still have the potential for accelerated autoinfection, and it has been recommended that every patient with *Strongyloides* infestation be treated. This is no less urgent than in the patient with altered host defenses. Because the results of treatment of disseminated strongyloidiasis in the compromised host have varied from fair to poor, every effort to make the diagnosis in patients who have appropriate geographic histories prior to the start of immunosuppressive therapy is indicated. Although definitive proof is lacking, early and appropriate treatment in these hosts may avert a potentially catastrophic bacterial or fungal superinfection.

The agent of choice for *Strongyloides* is thiaben-

dazole in doses of 25 mg/kg twice daily for 2–3 days in normal hosts. An alternative agent may be mebendazole, given in doses of 100 mg bid for 3 days. These agents are quite effective in normal hosts with only minimal adverse reactions consisting of nausea and GI upset. In the treatment of the immunosuppressed host, a significantly longer course of therapy seems necessary, and the agent has been given as long as 15 days with a mean of about 7 days. There is little danger of drug toxicity. More often, shorter but multiple courses of therapy are given. For those patients with repetitive episodes of sepsis–meningitis, it may be best to give thiabendazole 25 mg/kg 2–3 days every month.[216] Another method of suppression is to give a 5-day course of thiabendazole followed by a 3-day course of mebendazole. This appears to interrupt the cycle of recurrent systemic larval infection. The best guide to adequacy of treatment is the careful serial monitoring of feces or duodenal aspirates as well as following the course of extraintestinal manifestations (e.g., the chest radiograph). Persistent or recurrent disease has been associated with anatomic bowel abnormalities such as blind loops or diverticula and may require longer therapy or direct instillation of drug. Eosinophilia may take a longer period of time to resolve, perhaps as long as 2 months. Corticosteroids should be avoided, even in the face of high blood eosinophil counts. When polymicrobial infection is present, such as characterized by recurrent or breakthrough bacteremia, the bacteremia may cease only after effective antiparasitic treatment is given.

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