

9

Pulmonary infection in non-HIV infected individuals

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PEM are an important cause of pulmonary disease and the incidence is increasing in areas around the globe. With certain species the disease may have latitudinal preferences (refer to section 9.3). Although the clinical manifestation of pulmonary infection with PEM can resemble TB, the disease is very different because the host acquires infection by exposure to environment sources of these organisms (e.g. the water and soil). In contrast, disease due to MTB is predominantly spread person to person. Thus TB will not be discussed here except when it is pertinent to understanding certain host predisposing conditions for PEM infection and as it relates to situations of prior exposures and cross-immunity.

The causative microbial agents for PEM disease are numerous. The majority of the description in this chapter will deal with infection due to members of the MAC, given the preponderance of this organism causing disease. Unless

otherwise specified, MAC is the prototypical infection referred to in the text. However, when appropriate, other species will be highlighted.

Clinically, PEM infection can range from asymptomatic, indolent disease with minimal clinical symptoms to rapidly destructive pneumonic disease with significant morbidity and mortality. Therapy for these infections is difficult and often associated with toxicity and expense. For reasons that are not understood, extrapulmonary dissemination is strikingly rare when the systemic immune system is intact, despite very advanced pulmonary disease. Because PEM are ubiquitous in the environment, actual pulmonary disease needs to be differentiated from colonization or contamination in the laboratory. Finding environmental mycobacteria in respiratory secretions requires supportive clinical (symptoms, signs and radiographic evidence of disease) and microbiological evidence of infection before treatment is commenced. Practically, the decision to treat is sometimes very difficult and requires a mutual understanding between the patient and the responsible physician. Occasionally, when complex situations arise due to diagnostic or therapeutic uncertainties, the patient can be referred to an institution with experience in treating these infections. In this chapter, data will be presented detailing the geographical variation in environmental mycobacterial infection and potential explanations for these observations will be discussed. This topic is also dealt with in the chapters 2 and 3. For the interested reader, there have been a number of excellent reviews on the subject of pulmonary environmental mycobacterial infections (Griffith 1997, 2002; Iseman 2002).

9.1 CLINICAL ASPECTS

Acquisition of pulmonary infection most likely occurs by the aerosol route. Given the prevalence of PEM in the environment, exposure must be fairly universal. The steps that follow and the determinants of in whom disease takes hold are not completely understood. After disease develops, the symptoms can include cough, sputum production, fatigue, weight loss, sweats, haemoptysis, pleuritic and non-pleuritic chest pain. Occasionally symptoms are out of proportion to the amount of disease seen radiographically. For example, patients with significant disease noted on chest radiograph can be surprisingly asymptomatic or those with minimal changes can have debilitating symptoms. Fever is unusual unless bacterial superinfection occurs; however, the presence of another pathogenic organism in the sputum does not always predict the presence of fever. Often it is difficult to sort out symptoms due to the underlying lung diseases (e.g. dyspnea associated with emphysema or sputum production from underlying bronchiectasis) from those due to the underlying mycobacterial infection.

Confirmation of PEM in respiratory secretions is essential before committing the patient to therapy. If sputum specimens are negative, many clinicians will go on to bronchoscopy for diagnosis. Failure to recognize infection and disease and thus withholding therapy may result in unnecessary lung damage; nevertheless, pulmonary disease needs to be differentiated from colonization, which does not require immediate therapy. *M. kansasii* is thought less likely to be a saprophyte when it is found in patients' respiratory secretions than certain other species, although colonization can occur on rare occasions. In 1997, the American Thoracic Society published criteria for diagnosis and treatment of disease caused by the nontuberculous mycobacteria. These criteria are thought to be best suited for MAC, *M. kansasii* and *M. abscessus*. If a patient presents with the appropriate clinical symptoms and radiographs, one of the following criteria must be met for a microscopic diagnosis to be made (American Thoracic Society 1997):

- A** If 3 sputums/bronchial washings are available in last 12 months:
 - a) 3 positive cultures with negative smears or
 - b) 2 positive cultures and one positive smear

- B** If only 1 bronchial wash available:
 - a) positive culture with 2+, 3+, or 4+ AFB smear or 2+, 3+, or 4+ growth on solid media

- C** If sputum/bronchial wash evaluations are nondiagnostic or another disease cannot be excluded:
 - a) transbronchial or lung biopsy yielding PEM
 - b) biopsy showing mycobacterial histopathologic features (granulomatous inflammation and/or AFB) and one or more sputums or bronchial washings is positive for an NTM even in low numbers.

Expert opinion has varied regarding the usefulness of the above guideline but it is included here given the important role these criteria have played over the last few years by providing a standardized mechanism of diagnosis. In addition, the emphasis on the crucial role of a strong microbiological diagnosis is key to management of these patients. Once a diagnosis is made, the clinician may continue to be challenged by many facets of management of these patients. Respiratory secretions from these patients can harbour a complex suite of organisms such as mixed mycobacterial species, fungi and other co-pathogenic bacteria. This, too, can make appropriate antibiotic selection more difficult as

one tries to decide which organism is contributing the most towards pathogenicity.

The natural history of PEM lung disease is quite variable. There are two prototypical descriptions of disease: an indolent (primary) form that tends to occur in older non-smoking females and a more traditional form, which is usually secondary to underlying structural lung disease. Interestingly, there seems to be a shift in disease presentation from the secondary to the primary form.

9.1.1 Primary and Secondary Pathogenic Pulmonary Environmental Mycobacterial Infection

The epidemiology of PEM appears to be shifting from being predominantly a disease of male smokers (usually with underlying emphysema) to one of non-smoking older females with no obvious underlying lung disease. Given this emerging appreciation of different subsets of patients with PEM disease, it is useful to consider the infection as one of primary vs secondary infection in an attempt to sort out the dynamic nature of PEM pulmonary diseases. PEM pulmonary disease will be discussed in this chapter on the basis of “primary” and “secondary” disease based on terminology first proposed by von Reyn, *et al.* (2001). For the purpose of this monograph, primary disease will refer to the absence of a recognized host predisposition for infection (i.e. no pre-existing lung disease). Recently, a distinct syndrome of primary disease presenting as nodular bronchiectasis has been recognized. In this setting, our current understanding is that the mycobacteria are a cause of the disease manifestations in previously healthy hosts. Secondary PEM disease, on the other hand, occurs in the setting of a well recognized lung abnormality where the PEM takes advantage of the host’s pre-existing condition (i.e. it is an illustration of an opportunistic pathogen). Examples of secondary PEM infection include prior structural lung disease such as previous infection, emphysema and bronchiectasis. Secondary disease is relatively common, accounting for up to two thirds of disease in some reports. Regardless of whether PEM cause primary or secondary illness, there is little doubt that PEM can contribute to ongoing clinical illness and pulmonary dysfunction.

9.1.1.1 Primary PEM

In 1989 Prince and co-workers reported a subset of patients with pulmonary PEM disease who had no underlying predisposition (Prince *et al.* 1989). They found that 81% were female, 86% were white and there was a mean age of 66 years. Since the original work by Prince *et al.*, others have described the occurrence of PEM in hosts without obvious predisposing conditions

(Chalermkulrat *et al.* 2002). The patients, similar to the original report, are predominantly female non-smokers, and represent what appears to be a shift from the predominantly male dominated cases of secondary infection presented below (Rosenzweig 1979). These patients do not have obvious abnormalities in lung or immune function. The overwhelming majority are white (or Asian) women who present with nodular-bronchiectatic PEM disease (Reich & Johnson 1992; Kennedy & Weber 1994; Kubo *et al.* 1998; De Groot *et al.* 2001). In fact, the range of female patients in recent studies is between 75-94%. The term Lady Windermere's syndrome has been given to these patients based on the character in Oscar Wilde's play *Lady Windermere's Fan* as her voluntary cough suppression behaviour is considered to be the etiology behind not being able to clear secretions properly (Reich & Johnson 1992). However, this designation has been called into question (Iseman 1996). Whatever the etiology for this process, many experts and reports suggest that there has been a shift to female-predominant primary PEM disease. Most present in the fifth to eighth decade of life. Nodular bronchiectasis is the major clinical presentation in primary PEM disease. Whatever the reasons for this shift, an understanding of the clinical presentation and the gender-specific pathogenesis will be important. Classification of primary disease is based on the exclusion of underlying disorders as we understand them today. As we learn more about these putative predispositions, we may need to re-define these categories.

Patients with the nodular bronchiectatic primary PEM disease typically present with chronic cough and none, some, or all of the following: fever, fatigue, sweats, weight loss, dyspnea, hemoptysis and chest pain. Time from onset of symptoms to diagnosis can be weeks to years. Chest radiographs demonstrate nodular infiltrates and cylindrical bronchiectasis (Aksamit 2002). The characteristic high resolution computed tomography findings are bronchiectasis and ill-defined small nodules that are centrilobular in distribution. The typical description is "tree-in-bud" appearance, which is felt to represent an inflamed bronchiolar wall with fluid (i.e. mucus and inflammatory cells) within the lumen. Radiographic studies indicate that while disease can involve multiple segments of the lung, a very suggestive pattern consists of disease located in the mid-lung (right middle lobe and lingula) zones (Chalermkulrat *et al.* 2002). The computed tomography scan adds considerable information in the diagnosis of pulmonary PEM disease (Figure 9.1). Studies have shown airflow limitation as well as high residual volumes. Described together they represent distal air trapping in the small airways (Kubo *et al.* 1998). Up to 50% of patients can have normal results on pulmonary function testing (Prince *et al.* 1989; Aksamit 2002;). The disease can be fatal. In primary disease described by Prince and co-workers, death occurred in four patients due to progressive uncontrolled pulmonary infection. Relapse is common after

therapy is stopped. Left untreated the nodular bronchiectatic disease can go on to cavitate and cause severe lung destruction. A certain body phenotype has been described consisting of mitral valve prolapse and pectus excavatum. The patients are often thin and have an abnormal narrowing of the anterior-posterior diameter (Iseman *et al.* 1991). Sufficient data supporting a unifying explanation of a “predisposing phenotype” is currently lacking but is an area of interest for future study. Considerations of the role of bacterial virulence, previous exposure to antimicrobials, immunosenescence, host genetic and environmental factors influencing disease are fruitful areas of future research.

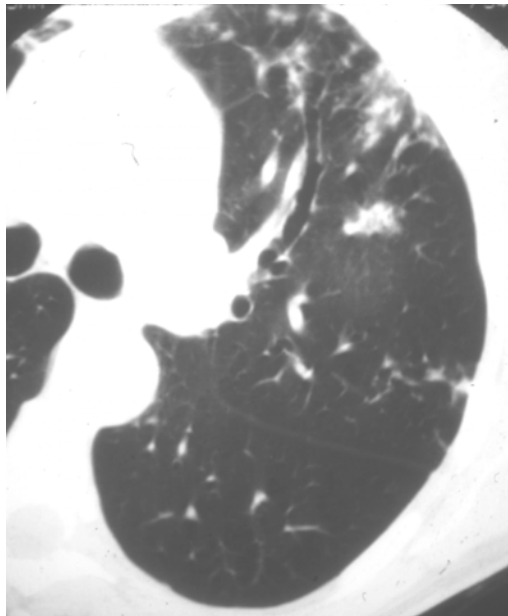


Fig. 9.1 High resolution computed tomography scan of a patient with PEM lung disease demonstrating lingular bronchiectasis, nodules, and infiltrates.

9.1.1.2 Secondary PEM

Unlike primary PEM patients, secondary cases do have some pre-existing lung disease to put them at risk for infection (Aksamit 2002). Secondary PEM disorders can be associated with CF, emphysema, previous TB and histoplasmosis, radiation therapy to the lung, silicosis and other inorganic dust pneumoconiosis, chronic aspiration, malignancies, etc. (refer also to section 9.6). It appears that regional factors in areas of lung damage predispose to

infection. It is postulated that abnormal blood flow, altered anatomy and local impairment of host defences may contribute. Many of these principal disorders result in fibrosis and scarring that is felt to become colonized/infected with PEM. In these cases, disease occurs at the site of previous damage. In smoking-related emphysema, disease is usually in the upper lung zones in areas of pre-existing bullae and can present with fibrocavitary disease. The symptoms can include cough, sputum production, fatigue, weight loss, sweats, hemoptysis, pleuritic and non-pleuritic chest pain. Fever is unusual unless secondary bacterial infection occurs. Often it is difficult to differentiate between symptoms due to underlying lung diseases (i.e. dyspnea associated with emphysema) and those of the superimposed mycobacterial infection. The radiographic manifestations can also include cavities, pleural thickening, nodular infiltrates, consolidation and various forms of bronchiectasis. Since nodules can also signify malignancy (to which smokers are predisposed) it is important to discriminate infection from malignancy. In emphysema related PEM disease, there is a predominance of male smokers. It remains to be seen if the rise in female smoking incidence will be reflected in increased secondary PEM in this group in the future. The patients with secondary PEM pulmonary infection are also in the older age range (sixth to eighth decade). The patients often have grossly abnormal measures of lung function (Aksamit 2002). The patients with secondary PEM tend to have cavities that are thinner walled when compared to those of TB (Prince *et al.* 1989).

Of particular interest is the association of PEM and CF. It is felt that the very viscous respiratory secretions of this disease contribute to recurrent pulmonary infections. Prior to the 1990s, the recovery of PEM from sputum of patients with CF was rare. Longevity into the third and fourth decades due to better antibiotics, inhaled medications, nutrition and improved sputum clearance techniques has occurred. With time there has been a rise in the incidence of other important pathogens including drug resistant *Staphylococcus aureus*, *Burkholderia cepacia*, *Achromobacter xylosoxidans*, *Stenotrophomonas maltophilia* and PEM. Olivier *et al.* (2002) found a prevalence of PEM of 13% in the sputum of CF patients. Of these, MAC accounted for 75% of PEM isolates. However, the contribution to respiratory decline in CF patients is disputed. With a follow up of 15 months they found no effect on lung function, but high resolution computed tomography scan of the chest abnormalities suggestive of infection with the PEM was predictive of disease progression. In a large study of multiple CF centres, patients who were infected with the PEM showed no significant differences in gender or frequency of pancreatic enzyme use. Only 37% of the patients met the American Thoracic Society microbiological criteria for disease. Those harbouring an *M. abscessus* were more likely to meet the criteria. Those with PEM were more likely to have

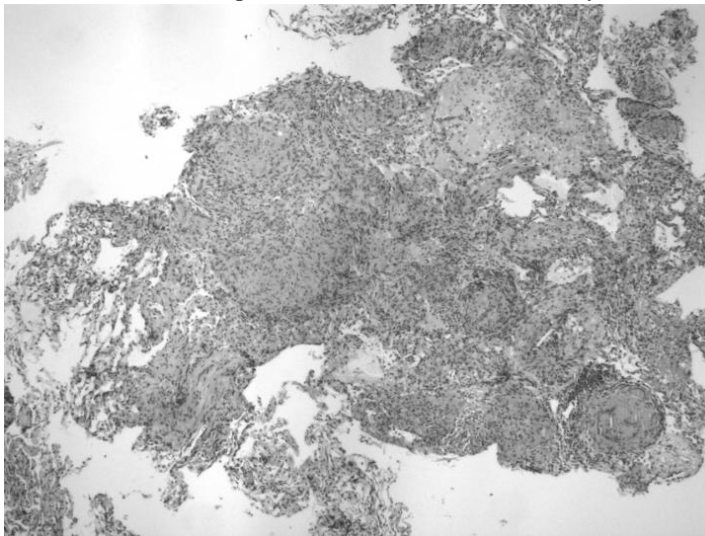
S. aureus infection and have a higher prevalence of *Aspergillus* co-infection. The clinical significance of infection is incompletely defined. There are reports substantiating the important pathogenic role of PEM when found in CF secretions. Autopsy studies have demonstrated granulomas and caseating necrosis felt to contribute to a patient's demise (Boxerbaum 1980). The clinical and radiographic manifestations of PEM in CF patients are similar to those in other cases of pulmonary disease. Chest computed tomography scans demonstrate bronchiectasis as well as cysts (or cavities) consolidated areas and peripheral nodules (including in a tree-in-bud pattern). However, these patients often have another infection (e.g. *Pseudomonas aeruginosa*) that can cause the same symptoms. Experts in the field recommend that multiple positive sputum cultures for the same organism and consistent symptoms and worsening spirometry as well as traditional radiographic findings be present before consideration for treatment (Olivier *et al.* 1996, 2001; Olivier 1998; Ebert & Olivier 2002). In rare circumstances where the patient is stable, a period of close clinical observation can be tried with a low threshold to begin therapy if any parameters change. Given the severity of the disease due to *M. abscessus* and the therapeutic difficulties, these patients are likely to require therapy without delay. The most common isolate in nearly all studies of CF patients is MAC but *M. kansasii* and members of the rapidly growing species (particularly *M. abscessus*) have also been reported. Since infection with the PEM is an indolent process, it is likely that with longer follow up significant clinical impact of infection will be demonstrated.

9.1.1.3 Hypersensitivity pneumonitis

Another form of PEM disease which has been recently recognized is the phenomenon of hypersensitivity pneumonitis seen in immunocompetent people with aerosol exposure to mycobacteria. This form does not have any known gender bias. Typically it is linked to hot tubs and indoor swimming pools and particularly strikes healthy subjects such as lifeguards and those who use these aquatic sources for recreation or work (Embil *et al.* 1997; Mangione *et al.* 2001; Mery & Horan 2002). Aerosols are felt to be the major route of infection in hypersensitivity pneumonitis. Likely due to their hydrophobic nature, these organisms are found in greater quantities in the air above the pool than in the pool water. High hydrophobicity can lead to adherence to surfaces. *M. avium* can also adsorb to air bubbles in water resulting in high concentrations of the organism at the air water interface (discussed in Chapters 2 and 3).

The precise pathogenesis is not completely understood, but investigators have attributed the pathology to both a component of bacterial infection and host immune response to mycobacterial antigens. A relatively distinct clinical

syndrome occurs after inhalation. Symptoms include cough, dyspnea, fatigue, impaired exercise tolerance and sputum production. The chest radiograph and computed tomography scans demonstrate hazy or ground glass opacities and peripheral small nodules often with a tree-in-bud pattern (Rickman *et al.* 2002). The AFB smear from sputum is insensitive but the culture is more sensitive (Khour *et al.* 2001). MAC is the most common offender but other species have been implicated. Bronchiolitis obliterans and other features of hypersensitivity, including non-necrotizing granuloma, are seen on pathology (Figure 9.2). Discontinuation of exposure to the source is mandatory. Treatments have varied



between antimycobacterial agents or steroids alone or often a combination of both (Khour *et al.* 2001). Discontinuation of hot tub use has led to prompt improvement in symptoms, pulmonary function and radiographic abnormalities without the use of antimycobacterial agents (Rickman *et al.* 2002). Hypersensitivity pneumonitis secondary to PEM as an occupational lung disorder has been well described and causes a similar syndrome. It has been seen most often in individuals using metal working fluids that are contaminated with mycobacteria (Hodgson *et al.* 2001; Wallace *et al.* 2002a).

Fig. 9.2 Histopathology of a patient with hypersensitivity pneumonitis. Intense inflammation, bronchiolitis obliterans and non-caseating granulomatous infiltrates are seen.

9.1.2 Selected treatment issues

As mentioned above, the criteria for diagnosis of disease usually include clinical signs and symptoms, sputum mycobacteriology and radiographic studies. Failure to recognize infection and disease and thus withholding therapy may result in unnecessary lung damage. Likewise, giving therapy when no evidence of disease is present only puts the patient at risk from unnecessary toxicity. The 1997 American Thoracic Society guidelines on diagnosis and therapy are a useful starting point. Our understanding of the optimal considerations for treatment continues to evolve.

In the era of the newer macrolides such as clarithromycin and azithromycin, greater treatment success is now achievable. Macrolides have the most predictable *in vitro* activity against MAC and have added a great deal to the treatment of infection (Griffith *et al.* 1996; Wallace *et al.* 1996). A typical course of antibiotics includes three or four drugs for many months. Preferred agents include clarithromycin (or azithromycin), rifampin (or rifabutin), ethambutol and possibly an aminoglycoside for a short period of time at the beginning of therapy. Thrice weekly regimens have been studied and are effective (Griffith *et al.* 1998, 2000). For previously untreated PEM disease due to MAC, a regimen of clarithromycin or azithromycin, rifampin, ethambutol and amikacin is often recommended. The amikacin is given for a period of two to three months with careful attention to renal, audio and vestibular toxicity. Toxicities with rifampin include orange discoloration of the secretions and urine, staining of contact lenses, nausea, vomiting, hypersensitivity syndrome (fever, rash), hepatitis, leukopenia, flu-like illness, thrombocytopenia, drug-induced lupus and renal failure. In addition, multiple drug interactions can occur manifested by increased hepatic metabolism of numerous agents by induction of the cytochrome P450 system. Rifampin combined with clarithromycin results in less bioavailability of clarithromycin. Rifabutin has similar toxicities to rifampin and can also cause polymyalgia and polyarthritis. Rifabutin has somewhat less effect on the P450 system. Clarithromycin can cause toxic accumulations of rifabutin due to its ability to inhibit the elimination of rifabutin (which can be associated with uveitis). Ethambutol may cause optic neuritis (loss of red/green colour discrimination and loss of visual acuity) and rash. This side effect is often dose related. Given the complexity of therapy, consultation with experts in the field should be considered.

With the exception of clarithromycin, the role and the predictability of *in vitro* susceptibility testing of MAC is an area of controversy (Iseman 2002). In contrast to TB, where susceptibility testing has an undisputed role in management of disease, in PEM disease it is somewhat debatable. The American Thoracic Society statement recommends against testing of agents other than clarithromycin. A study done at the National Jewish Medical and Research Center found that there

was a correlation between treatment response (as measured by consecutive sputum culture negativity for three months) and the number of antimycobacterial drugs that the patient was treated with that had demonstrable *in vitro* susceptibility (Iseman 2002). If a patient has had prior therapy with a macrolide or is failing therapy, *in vitro* susceptibility testing for clarithromycin (at a minimum) should be performed. Testing of rifampin susceptibility for *M. kansasii* and a panel of antimicrobial agents for other mycobacteria, such as the RGM, is recommended (American Thoracic Society 1997). The recommendations for duration of therapy are based on a high rate of relapse if treatment is terminated early (Wallace *et al.* 1996). Anywhere from 12-24 months is the usual suggested duration of treatment, although shorter courses may yet be possible using macrolide containing regimens. Additional details and other variations of these recommendations based on unique underlying patient characteristics can be found in the review by Iseman (Iseman 2002).

Special considerations exist in the diagnosis and management of *M. kansasii* disease. This subject has been extensively reviewed (Griffith 2002). *M. abscessus* may be the most difficult pathogen to treat in patients with and without CF. It is difficult to render patients with this infection culture negative despite the apparent *in vivo* susceptibility of the isolates in the laboratory. It is possible that this has to do with an enhanced resistant state (i.e. a biofilm mode of growth) in the host compared to the planktonic culture used for *in vitro* susceptibility testing (Bardouniotis *et al.* 2003). Treatment recommendations for *M. abscessus* includes amikacin, cefoxitin and usually a third agent such as clarithromycin is added. The duration of intravenous therapy is typically six to eight weeks with a period of longer-term suppressive therapy. Patients with CF have altered drug absorption and pharmacokinetics. Consideration should be given to measuring serum antimicrobial concentrations for patients with and without CF, which may dictate altering drug dosages to avoid toxicity and achieve therapeutic success. Serum concentrations for most of the antimycobacterial agents can be obtained from the Pharmacokinetic Laboratory at the National Jewish Medical and Research Center (+1-303-398-2603). A multicentre treatment trial involving inhaled interferon-gamma is nearing completion and results should soon be available.

Surgical therapy continues to play a role in the management of these patients; however, it is largely reserved for patients who have failed medical therapy, or for those with severe symptomatic disease. When present, localized disease lends itself best to surgical intervention. Predictably, those that have poor preoperative lung function do less well. Complications arising from treatment, such as bronchopleural fistulas, etc., do occur (Iseman *et al.* 1985; Pomerantz *et al.* 1991, 1996; Nelson *et al.* 1998; Shiraishi *et al.* 2002): those that undergo pneumonectomy (especially for right-sided disease) seem to have a higher risk of severe postoperative complications (Pomerantz *et al.* 1991).

9.1.3 Selected microbiological issues

In the clinical context, microbiological data is the foundation of a proper diagnosis. Specimen processing in a laboratory capable of performing up-to-date and timely identification and susceptibility testing is important (refer to 9.4). Isolation of PEM from the sputum of patients with CF is often difficult due to the common finding of overgrowth of more rapidly growing bacteria and fungal species. In order to enhance recovery of PEM, oxalic acid is sometimes added to the decontamination step in specimen processing and can increase the yield of PEM (Bange & Bottger 2002). If a high index of suspicion exists, multiple specimens should be submitted to the laboratory for analysis.

Advances in laboratory methodologies have enabled more rapid and reliable differentiation of mycobacterial species. Also, new and emerging species have recently been described that cause pulmonary disease. Additional insights into PEM infections are gleaned from applying molecular typing tools. In a study of pulmonary nodular bronchiectasis infection involving *M. intracellulare* isolates that had the same colony morphology on the plate, PFGE demonstrated multiple different fingerprints (i.e. a polyclonal infection) (Wallace *et al.* 1998a). In patients with secondary PEM infection, especially disease due to underlying emphysema with fibrocavitary disease, there was often only a single isolate over time. If therapy with a macrolide was continued long enough in those with nodular bronchiectatic disease and relapse, the relapse strain was macrolide susceptible and represented acquisition of a new strain (Wallace *et al.* 2002). Considering the relatively high numbers and wide diversity of *M. avium* and *M. intracellulare* in the environment (i.e. drinking-water and soil), it is not surprising that some patients would be infected by more than a single strain.

9.2 OVERALL BURDEN OF DISEASE

The precise incidence of PEM is difficult to ascertain. MAC is estimated to have a range of infection of 1-2.5/100 000 population and is rising (Marras & Daley 2002). The upward trend is not due exclusively to AIDS, because PEM occurred before and independently of the appearance of HIV. There are multiple potential reasons for an increase in PEM disease. These include, but are not limited to: 1) more sputum cultures are being ordered; 2) more computed tomography lung scans are being ordered and when the classic radiographic pattern is seen (i.e. bronchiectasis, tree and bud), cultures are more likely to be requested; 3) the use of more liquid media resulting in higher recovery rates compared to the use of Lowenstein-Jensen agar media. The organisms most commonly implicated in human infections are MAC, *M. kansasii*, and the RGM especially *M. abscessus*. In some regions, *M. kansasii* is one of the most common PEM isolated. It has

annual infection rates of 0.5-1/100 000 population. In certain areas it has higher rates (i.e. 2.4-17.6 per 100 000). It occurs in geographic clusters and affects primarily white men but it can occur in any race or at any age.

9.3 DISTRIBUTION

PEM lung disease has been reported from all over the globe. MAC has been most commonly studied and is found in nearly all regions of the world where it has been looked for. However, it has been found in the primary disease form particularly in patients from the south-eastern United States coastal states of the gulf (M. Iseman, G. Huitt, personal communication). In a study done by the CDC, the rates of MAC isolation tended to be highest in states bordering the Atlantic ocean and the Gulf of Mexico (> 4.8/100 000) (Good & Snider 1982). *M. kansasii* tends to occur in an inverted "T" in the United States with a broad area of disease in Texas, Oklahoma and up into the Midwest. *M. xenopi* is an endemic problem in Britain and France; however, sporadic cases have been described elsewhere, including in the United States. *M. malmoense* is also more prevalent in Europe. Other slowly growing mycobacteria, including *M. simiae*, appear to be on the rise as well. The distribution of disease due to this species and to some of the rarer species is unknown.

9.4 DESCRIPTIVE EPIDEMIOLOGY

The epidemiological study of pulmonary infections is challenging. Growth of a PEM in culture can result from contamination in the laboratory or from any point in the process of specimen collection. In order to reliably discern the incidence and prevalence of disease associated with each isolate, clinical data is required to understand more comprehensively the true incidence of disease. In contrast to infection caused by *M. tuberculosis*, disease due to the environmental mycobacteria is not reportable in the United States. Thus data is not available to ascertain the precise incidence and prevalence.

In the early 1980s O'Brien *et al* (1987) carried out a survey of the state and city health departments. Disease due to the PEM occurred in 1.8/100 000. Limitations of the study include incomplete sampling from across the population of the United States and possible under-recognition of the nodular bronchiectatic form of disease at the time the survey was done. However, other national surveys, such as one performed in Switzerland, showed fairly consistent profiles of disease prevalence of around 1/100 000 population (Debrunner *et al.* 1992). A superb analysis of the world's literature has recently been published (Marras & Daley 2002). The authors have analysed data where laboratory based surveys also contained clinical data allowing some

ascertainment of presence or absence of disease. Nearly all studies performed in Europe, North America, Asia, Africa and Australia found a rising proportion of potentially pathogenic mycobacteria in laboratory-based surveys. For instance, in one province of British Columbia the annual incidence of PEM pulmonary disease rose at an exponential rate (from 0.08/100 000 in 1960 to 0.6/100 000 in 1980). In Massachusetts in 1972 non-*M. tuberculosis* species accounted for 12% of mycobacterial isolations; by 1983 they accounted for 70%. The incidence rates in recent decades range from 1.7-4.5 per 100 000 people. In Europe, reports of disease rates range from 0.3-18/100 000 people. African, Asian and Australian studies have shown a rate between 0.3-78/100 000 in certain areas. In all regions, most reports would suggest that concomitant with the decline in TB, the incidence of PEM has increased.

9.5 CAUSALITY AND ASSOCIATED MICROBES

It is important to accurately identify the organism to ensure proper diagnosis and to make proper treatment selections. Advances in laboratory methodologies have enabled more rapid and reliable differentiation of mycobacterial species and new species with the potential to cause pulmonary disease are being described all the time. In the United States MAC was most commonly seen followed by *M. kansasii* (20%) and rapid growers (10%) (Marras & Daley 2002). MAC includes *M. avium* and *M. intracellulare*. These two species are indistinguishable using routine laboratory criteria such as biochemical testing and morphology. Newer methods based on sequencing and chromatographic patterns of mycolic acids are employed in reference labs. Molecular tools have allowed distinction on the basis of 16SrRNA sequences using commercially prepared kits (GenProbe, Accuprobe). Each species should be described separately as they are likely to have different biologies in the environment, unique antibiotic susceptibilities and particular virulence characteristics. Recently, by sequencing the *hsp65* gene of representative isolates, it was shown that there are unique pathogenic capabilities of human isolates compared to the environmental strains (Smole *et al.* 2002).

9.6 RISK FACTORS

Risk factor analysis needs to consider host factors, microbial factors and environmental factors.

While the precise details are not well worked out, it is generally believed that patients acquire mycobacteria from their environment, including waters and soils. The number of mycobacteria isolated from drinking-water can be very high: up to 100 000 cfu/cm² can be found in water distribution systems (refer to Chapter 3). Since the combined length of water pipes in an average distribution system can be hundreds

of miles, the potential human exposure is immense. In addition, the mycobacteria are relatively resistant to chlorination, which may explain their success and persistence in drinking-water.

Pulmonary infections disproportionately affect the elderly. Some of the areas with the highest incidence of PEM disease are also the areas with the greatest proportion of older adults (data from the United States Census Bureau). The proportion of people over 65 years of age was 1 in 20 in 1930, it increased in the mid eighties to 1 in 10, and projections for 2020 are that 1 in 5 individuals will be 65 years and older. With an ageing population, we may see more pulmonary PEM disease. It may also be that the current trend in some countries for elderly people to retire to warm, moist environments may put these people at risk. Additional risk factors include those with underlying host conditions such as patients with CF and smoking-related emphysema. These and additional risk factors for pulmonary disease are listed in Table 9.1.

Table 9.1 Risk factors for PEM disease (adapted from De Groote & Iseman 2003)

Traditional lung disorders	Heritable Conditions	Body Habitus	Prior lung infections	Aspiration
emphysema	CF	pectus excavatum	TB	gastro-oesophageal reflux
dust pneumoconiosis	disordered ciliary motility	scoliosis	histoplasmosis	achalasia
fibrosis, ankylosing spondylitis, rheumatoid arthritis	tracheo-broncheomegaly	slender body type	coccidioidomycosis	swallowing disorders
radiation lung injury, alveolar proteinosis, pulmonary embolism	alpha-1 anti-trypsin disorders	mitral valve prolapse	aspergillosis	mineral oil ingestion

Host defences against mycobacterial infections include intact epithelial surfaces, the mucociliary lining of the respiratory tract, neutrophils, macrophages and lymphocytes. Factors that negatively affect any of these defences predispose patients to mycobacterial infections.

9.7 PREVALENCE OF ASYMPTOMATIC DISEASE

Most of the data regarding asymptomatic disease comes from skin test surveys. This test depends on the development of a delayed hypersensitivity to skin test

antigens derived from PPD of mycobacteria. In two separate and large studies, a consistent and striking epidemiological finding was the geographical differences in exposure, with particular high rates in the south-east United States (Marras & Daley 2002). The first used the PPD of *M. tuberculosis* at low and high doses (5 and 250 TU). A positive response to the 250 TU skin test was felt to be related to exposure to antigenically similar mycobacterial species. Those subjects whose residence was in zones encompassing Texas, North and South Carolina, Georgia, Florida, Tennessee, Alabama, Mississippi, Louisiana, Arkansas, Georgia and Oklahoma had the highest degree of reactivity. Similarly, skin test surveys done in the 1960s using a battery of skin test antigens including the Battey bacillus (MAC) revealed a higher percentage of reactivity in Navy recruits from the south-eastern United States. Von Reyn *et al.* (2001) have recently shown a high incidence of skin test positivity in the United States with higher rates among adults in the southern states. Limitations in interpreting this data related to asymptomatic disease include absence of supportive clinical data to rule out active disease. Both large studies were done on presumably healthy young people, so this is likely just a theoretical concern. Secondly, there is non-specificity in the antigenic response to antigens in these preparations and cross reactivity can occur. Cross immune protection to MTB and other environmental mycobacteria probably occurs. Newer tests using gamma interferon production by whole blood lymphocytes may yield additional discriminative information.

9.8 KEY RESEARCH ISSUES

There are a number of research priorities to be considered in dealing with the issue of PEM pulmonary infections. Some of these are listed below:

- design prospective, safe, multicentre controlled studies of natural history, risk factors, treatments and predictors of outcome;
- develop and standardize methods for unique identification to the species level and study microbial correlates to pathogenesis and outcomes;
- define the nature of exposure in the environment to advise high-risk individuals how to avoid exposure;
- define the role of pharmacokinetic and *in vitro* susceptibility studies in the routine management of PEM.