Pneumocystis Infection: Seeing beyond the Tip of the Iceberg

Enrique J. Calderón
Instituto de Biomedicina de Sevilla, Hospital Universitario Virgen del Rocío/Consejo Superior de Investigaciones Científicas/Universidad de Sevilla, and Centro de Investigación Biomédica en Red de Epidemiología y Salud Pública, Servicio de Medicina Interna, Hospital Universitario Virgen del Rocío, Seville, Spain

Pneumocystis jirovecii probably is one of the more frequent infectious agents faced by humans in everyday life. However, after the first description of Pneumocystis a hundred years ago, the organism was largely ignored until the dramatic increase in the incidence of Pneumocystis pneumonia (PCP) that occurred with the emergence of the human immunodeficiency virus (HIV) pandemic, which made pneumocystosis a major medical and public health problem in the 1980s [1]. The AIDS epidemic stimulated research on PCP that can be considered just the tip of the iceberg of human Pneumocystis infection [2]. Research interest in Pneumocystis infection accompanied the clinical interest but was thwarted by the inability to culture the organism in vitro. However, the introduction of experimental models and molecular biology techniques have allowed us to overcome some of the limitations imposed by the lack of a reliable culture system [1].

The past 20 years has seen impressive advances in our understanding of the basic biology of Pneumocystis. Molecular and biochemical analyses have provided unequivocal evidence for the placement of Pneumocystis with fungi [3]. Genetic investigations have shown that Pneumocystis organisms derived from different mammalian hosts exhibit considerable chromosomal and gene-sequence divergence, indicating that they are of different species [4]. Now, it is well recognized that each mammalian host is infected by a specific Pneumocystis that cannot infect other hosts [4]. Therefore, efforts are being made to reclassify the various Pneumocystis organisms as separate species. Recently, the organism that causes disease in humans was renamed P. jirovecii [5].

Today, it is established that human PCP in not a zoonotic disease, and this notion has important implications for the epidemiology of P. jirovecii [6]. Even though early studies described the identification of Pneumocystis DNA from the air surrounding apple orchards and the surface of pond water, no Pneumocystis forms were identified in environmental samples by microscopic analysis, and it is uncertain whether there is an ecological niche for Pneumocystis outside mammalian hosts [7–9]. Animal sources of P. jirovecii can be excluded, because the Pneumocystis organisms that infect mammalian species are characterized by strong, close host species specificity [4]. To date, the human being is the only known reservoir host for P. jirovecii, and humans probably acquire the infection only from other humans [10].

Serologic studies have shown that antibodies specific to the pathogen can be detected in most children early in life, indicating frequent exposure to this organism [11, 12]. On the basis of this finding, disease in immunocompromised persons has long been thought to result from reactivation of latent infection acquired in childhood. However, animal and human studies have shown that elimination of Pneumocystis often occurs after infection, implying that the persistence of latent organisms is limited [9]. Recent demonstration of P. jirovecii transplacental transmission may explain the accumulating evidence that primary infection is widely acquired very early in life and supports the commonly held view that human infants are a major natural reservoir of P. jirovecii, since they can remain colonized as their immune response matures [13, 14].

Colonization with P. jirovecii in adults has recently gained attention as an important issue for understanding the complete cycle of human Pneumocystis infection [15]. In general, colonization is defined as isolation of a microbe that does not result in sufficient damage to cause clinical disease but that may alter host homeostasis. For Pneumocystis in particular, coloniza-
tion has been defined as detection of the organism or its DNA in respiratory samples from individuals who do not have signs or symptoms of pneumonia [15].

Detection of *Pneumocystis* colonization has been greatly facilitated by the development of sensitive molecular techniques, such as polymerase chain reaction (PCR). Although colonization can occasionally be detected by traditional staining methods or single-round PCR, nested PCR is generally used because of its greater sensitivity [16, 17].

Among adults, *Pneumocystis* colonization has been demonstrated in both HIV-infected and non–HIV-infected subjects, and certain populations appear to have a higher risk of colonization. Studies have shown that individuals who have an underlying HIV infection or another cause of immunosuppression and those who are not immunosuppressed but have chronic lung disease may often be colonized by *P. jirovecii* and can be a major species-specific reservoir of infection [15].

In this issue of *Clinical Infectious Diseases*, Ponce et al [18] provide evidence that could change the current view of human *Pneumocystis* infection. The investigators found *P. jirovecii* in the lung tissue of approximately two-thirds of the general adult population. Although the high prevalence of *Pneumocystis* carriage in the immunocompetent rat model has long been recognized [19], the high frequency of *P. jirovecii* colonization reported by Ponce and colleagues is surprising. Multiple studies have failed to find the organism on autopsy, in lung tissue, or in respiratory samples from nonimmunocompromised individuals [20–23]. Only one previous study has demonstrated the presence of *Pneumocystis* DNA among adults (at a rate of 20%) who were otherwise healthy [24]. The lower level of colonization found in this study could be explained by varying geographic exposure, but it could also be explained by the method used to collect respiratory samples. However, studies using lung tissue specimens from populations considered to have a higher risk of colonization—such as HIV-infected men dying of causes other than PCP and patients with severe chronic obstructive pulmonary disease (COPD)—also found a lower prevalence of *Pneumocystis* colonization than did Ponce et al [18, 25, 26]. The higher level of detection in the study of Ponce and colleagues might be the result of geographic variation in *Pneumocystis* colonization or differences in PCR techniques. Thus, in a previous study conducted by the same team using the same methods in lung tissue from infants from the same geographic area, a lower colonization prevalence was found in infants than in adults in the current study, although the difference is not statistically significant (51.7% vs 64.9%; *P* = .12) [18, 27]. These findings could be explained by a period effect, but they also could be explained by a cohort effect related to pulmonary reinfections occurring after the primary infection, suggesting that *P. jirovecii* can remain latent within the host for long periods of time. Alternatively, the findings could imply that *P. jirovecii* is ubiquitous in the environment and that exposure in humans is characterized by intermittent colonization [9]. Recently, there have been increasing reports of *Pneumocystis* colonization. This increase could represent an improvement in detection techniques or a growing prevalence of colonization in the community [28].

In studies using single time point identification, as in the study of Ponce and colleagues, there is uncertainty as to whether individuals were truly colonized or had recently been environmentally exposed, because the duration of *P. jirovecii* detectability could not be determined. Thus, the presence of *P. jirovecii* in lung tissue in the absence of overt pneumonia has uncertain significance, both for the individual and in terms of the potential for person-to-person transmission [29].

In any case, results of the study of Ponce and colleagues should be interpreted with caution and requires independent verification; if confirmed, their work would greatly increase our understanding of the size of the human reservoir of *Pneumocystis* infection and support the saprophytic nature of this microorganism in humans.

The accumulating evidence suggests that *P. jirovecii* is a highly adapted parasite that most likely circulates by active horizontal and vertical (aerial or transplacental) transmission mechanisms among human populations and causes mostly mild (although frequent) parasitism in the host’s lungs [3, 4, 10, 30]. Regardless, many important questions remain unanswered about *Pneumocystis* colonization [31]. For instance, are those persons who are colonized by *Pneumocystis* at risk for developing PCP? Could any colonized individual transmit the infection to others, or is transmission dependent on parasite load? How long does *Pneumocystis* carriage last in human populations? Are there factors that can affect the length of *Pneumocystis* carriage? If a colonized individual takes sulfon drugs for a long period of time, is there a risk that a drug-resistant organism may develop, and is this person able to transmit *P. jirovecii* with dihydropteridine synthase gene mutations to others? Animal data indicate that *Pneumocystis* colonization provokes an intense inflammatory response in the lungs. Does a similar response occur in humans, and is it detrimental to the lungs? Might such inflammation play a role in the progression of COPD or other chronic pulmonary diseases?

The answers to these questions have the potential to affect patient care and deserve further investigation to better define the epidemiology and importance of *Pneumocystis* colonization in humans.

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