EDITORIAL

Chlamydia pneumoniae: crossing the barriers?

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Chlamydia pneumoniae has been recognised as a cause of respiratory tract infections and implicated as a potential risk factor or causative agent in different extrapulmonary diseases including atherosclerosis, multiple sclerosis, and Alzheimer's disease [1–3]. Being an obligate intracellular bacterium, *C. pneumoniae* has been detected in circulating monocytes and can activate inflammatory processes in epithelial, endothelial and smooth muscle cells *in vitro* [4].

In the present issue of the *European Respiratory Journal* (*ERJ*), GIEFFERS et al. [5] report an animal model showing that intratracheal infection with *C. pneumoniae* is followed by systemic dissemination of the infection mediated by peripheral blood mononuclear cells (PBMCs). The authors, on the basis of both the animal model and *in vitro* study results, hypothesise a "cellular model" for *C. pneumoniae* dissemination. Infection of the lung is characterised by an early phase dominated by granulocytes, and a late phase dominated by alveolar macrophages. Alveolar macrophages, infected by granulocytes, would migrate through the mucosal barrier, using lymphatic tissue, and gain access to the systemic circulation as PBMCs reaching the spleen and the vasculature. The conclusions are mostly inferred on the basis of cell morphology in the absence of definitive determination of infected cell types in extra-pulmonary tissues. Nonetheless, the proposed "cellular model" hypothesis is intriguing and undoubtedly consistent with other recently published studies.

WARK et al. [6] analysed the relationship between airway inflammation and serological response to *C. pneumoniae* in acute severe asthma. At presentation with acute asthma, the sputum total cell count was increased in *C. pneumoniae* antibody responders compared to nonresponders, and *C. pneumoniae* responders had significantly more sputum neutrophils compared to nonresponders. MOAZED et al. [7] demonstrated that monocytes may act as vectors and systemically disseminate *C. pneumoniae*, and BLASI et al. [8] showed a good correlation between *C. pneumoniae* detection in PBMCs and in atherosclerotic plaques. However, in order to adhere and migrate through the vessel wall, monocytes have to go through a highly coordinated process, which requires the activation of different adhesion receptors in a cascade-like fashion. MAY et al. [9] report that *C. pneumoniae* infection induces rolling and adhesion of macrophages to the noninflamed vessel wall of noninfected, nonatherosclerotic mice. *C. pneumoniae*-infected monocytic cells show enhanced transmigration and attach to the endothelium *via* the activated integrins very late antigen 4 (VLA-4), and the activation of the two β_2-integrins lymphocyte function-associated antigen-1

(LFA-1) and macrophage antigen-1 (MAC-1), involving the urokinase receptor (uPAR). This study demonstrates that *C. pneumoniae*-infected monocytes may be armed to invade noninflamed subendothelium and initiate inflammatory processes. The data indicate that *C. pneumoniae* has the potential to induce a functionally active, adhesive state in monocytic cells by activation of the integrin adhesion receptor system. Therefore, *C. pneumoniae* is not just transported into the subendothelium by monocytes as an innocent bystander, but can actively contribute to the monocyte recruitment to the preferential sites of atherosclerotic lesions. Moreover, this study suggests that *C. pneumoniae*-infected circulating monocytes may have the capacity to induce an adhesive phenotype in adjacent, noninfected monocytes.

Monocytes perhaps also act as the vehicle for trafficking *C. pneumoniae* across the blood/brain barrier. PBMCs may function as a means by which *C. pneumoniae* enters the central nervous system (CNS) to induce neuroinflammation in Alzheimer's disease and in multiple sclerosis [10]. *C. pneumoniae* infection has been shown to stimulate trans-endothelial entry of monocytes through human brain endothelial cells (HBMEC). This entry is facilitated by the upregulation of vascular cell adhesion molecule-1 and inter-cellular adhesion molecule-1 on HBMECs and a corresponding increase of LFA-1, VLA-4, and MAC-1 on monocytes.

An association between *C. pneumoniae* and multiple sclerosis has been proposed, based on the higher frequency of its detection in the cerebrospinal fluid (CSF) of multiple sclerosis patients compared to neurological controls. Multiple sclerosis is an inflammatory demyelinating disease of the CNS of unknown etiology. Current knowledge supports a multi-factorial aetiology in which both genetic and environmental factors (including microbial agents) may concur. Interestingly, experimental autoimmune encephalomyelitis (EAE), the experimental animal model of multiple sclerosis, has been successfully induced using a *C. pneumoniae* peptide analogue of rat myelin basic protein [11]. Although multiple sclerosis and EAE are obviously two different entities, this study provides the first indication of a possible direct contribution of *C. pneumoniae* to the pathophysiology of (experimental) demyelination. The presence of *C. pneumoniae* in human CSF does not actually prove that the organism causes or triggers multiple sclerosis: chlamydial infection of the CNS may just represent an opportunistic, secondary event in the disease. Even in this case, however, the presence of the organism may exacerbate/modulate a pre-existing pathogenic process. This is also supported by the finding that *C. pneumoniae* polymerase chain reaction (PCR)-positive patients have more active lesions than *C. pneumoniae* PCR-negative/patients suggesting a role for *C. pneumoniae* in fostering chronic inflammatory stimulation within the CNS [2]. It can be hypothesised that *C. pneumoniae* might act as cofactor capable of fuelling previously established inflammatory and demyelinating processes and promote a more active disease.

GIEFFERS et al. [5] showed a good correlation between the presence of *C. pneumoniae* in PBMCs both in the lung and in

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the vasculature. These data are consistent with the results reported by BLASI and co-workers [8, 12] who showed that *C. pneumoniae* DNA identification is similar in biopsy specimens (vascular and bronchial) and PBMCs, which suggests that blood PCR may be a useful tool for identifying patients with chronic *C. pneumoniae* infection.

Clinical persistence is probably a key concept in *C. pneumoniae* infection pathogenesis. Microbial persistence is a state of infection during which the host immune response does not eliminate the pathogen, thereby resulting in continuing damage to the host. Persistent infection may amplify airway inflammation in asthma and chronic obstructive pulmonary disease (COPD), but also in extrapulmonary diseases such as atherosclerosis, multiple sclerosis and Alzheimer's disease.

STEPHENS [13] has recently revised the possible pathogenic mechanisms of *C. pneumoniae* infection. He underlines that *C. pneumoniae* can induce an inflammatory process elicited by infected host cells that is necessary and sufficient to account for chronic and intense inflammation and the promotion of cellular proliferation, tissue remodelling and scarring, the ultimate causes of disease sequelae.

The cellular responses of epithelial cells, the primary home for *C. pneumoniae*, can be reliably induced upon acute, chronic and persistent infection. The cellular processes of the epithelial cells, elicited by chlamydial infection, cause the influx of inflammatory neutrophils, T-cells, B-cells and macrophages that are stimulated by the pro-inflammatory cytokine and chemokine environment. These cells become activated in both antigen-nonspecific and, for re-infection, antigen-specific responses to produce their own repertoire of cytokines and growth factors. The induction of host cell cytokines will promote foci of inflammatory responses in addition to promoting cellular proliferation, tissue remodelling and healing processes that, if persistent, result in scarring.

The possible role of chronic-persistent infection is suggested in asthmatic children where persistent clinical features are associated with *C. pneumoniae* infection, indicating that this infection should be investigated and treated in case of persistent asthmatic symptoms [14–15]. On the other hand, in adults with asthma, higher titres of antibodies directed against *C. pneumoniae* are associated with more severe clinical disease [16]. The intense neutrophil influx demonstrated in acute severe asthma is potentially a potent source of proteolytic enzymes with the ability to damage and activate the airway epithelium, whereas neutrophil elastase can cause eosinophil degranulation [6]. The inflammatory changes caused by infection with *C. pneumoniae* have the potential to amplify the inflammation and airway damage present in asthma. Furthermore, standard asthma treatment may potentially enhance this response, given the ability of corticosteroids to reactivate *C. pneumoniae* infection. These data suggest that *C. pneumoniae* infection may influence the clinical course of asthma.

In the same way, preliminary data on chronic infection with *C. pneumoniae* in patients with COPD and its interaction with host cells indicate that this agent may be implicated in the modulation of the natural history of chronic bronchitis and emphysema [12].

The interesting finding of the study by GIEFFERS *et al.* [5] is the demonstration that granulocytes are one of the main target of *C. pneumoniae* infection and that these cells can act as "infecting" cells and reservoir of the pathogen. These data confirm the excellent capacity of *C. pneumoniae* to survive in different immune cells, use the immune cells as carriers for breaching the blood-tissue barriers, and potentially cause chronic/persistent infections.

Chronic infection is probably the real challenge in *Chlamydia pneumoniae* infection and we certainly need new studies addressing the question whether *Chlamydia pneumoniae* long-term survival within immune and nonimmune cells has a role in chronic pulmonary and extrapulmonary diseases.

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