EDITORIAL COMMENT

Chlamydia pneumoniae Infection and Coronary Artery Disease*

Juan Carlos Kaski, MD, FRCP, FESC, FACC
A. John Camm, MD, FRCP, FESC, FACC
London, United Kingdom

The possibility in recent years that chronic infection with Chlamydia pneumoniae (CPn) may represent a risk factor for atherosclerosis or a mechanism of rapid progression of ischemic heart disease, or both, has attracted the attention of investigators and clinicians all over the world. After the first description of a seroepidemiologic association between antibody titers against the intracellular bacterium CPn and coronary artery disease in 1988 (1), scientific publications (2) from both sides of the Atlantic have confirmed the initial observations. However, more recent reports, one from the U.S. (3) and another from the United Kingdom (4), have cast doubts as to the existence of a true association between CPn seropositivity and coronary events. In contrast, two other large studies also reported this year have found a strong association between immunoglobulin G (IgG) antibodies against CPn and stroke (5) and between anti–CPn IgA antibodies and mortality from ischemic heart disease (6). Thus, at present, controversy exists regarding the postulated relation between CPn seropositivity and ischemic heart disease. One of the possible explanations for the controversial findings may be that in defining “seropositivity,” different authors have used different serologic techniques or have looked at different antibodies, or both. At least three different methods have been used to assess anti–CPn antibody titers—enzyme-linked immunosorbent assay (ELISA), the complement fixation technique and the microimmunofluorescence (MIF) test, which measures specific immunoglobulins (i.e., IgG, IgA and IgM).

Limitations of serology to assess active CPn infection.
Although serology constitutes an important diagnostic tool for the assessment of chronic CPn infection, it is not without limitations. Even the MIF test, which has been considered by some (7) to be the “gold standard” test for assessment of CPn infection owing to its reportedly high sensitivity and specificity, has some drawbacks, including the unknown reproducibility of the technique and the need for highly skilled operators. In addition to the intrinsic limitations of serologic tests, the issue is further complicated by the fact that different investigators have used different antigens or CPn strains and have applied diverse criteria for the definition of chronic versus acute CPn infection. Some of the studies have looked only at IgG or IgA and others have based their conclusions on the presence or absence of circulating immune complexes containing the CPn lipopolysaccharide. The selection of control individuals also varied among studies and earlier reports, in particular, failed to control for confounding factors such as conventional risk factors of coronary disease and social class. There are also other considerations, such as the high prevalence of seropositive cases found in the general population, and this is probably a reflection of the incidence of infection and the duration of raised antibody titers after infection. It is therefore difficult to establish whether seropositivity to CPn indicates a chronic, active infection or simply reflects past exposure. In some patients high antibody titers against CPn may indicate past exposure to the microorganism and not necessarily the need for treatment. In other patients, however, high antibody titers may be the expression of active CPn infection, which may trigger inflammatory mechanisms and contribute to atheromatous plaque disruption. Accurate diagnosis of active infection should require repeated samples to demonstrate rising antibody titers.

Thus, although seroepidemiologic studies have undoubtedly helped to identify an intriguing and potentially important association between CPn infection and atheromatous disease, more accurate techniques are required to characterize active CPn infection in the search for the true role of this organism in ischemic heart disease.

CPn detection in atheromatous tissue—the role of macrophages. Studies have shown that CPn can be detected in atheromatous vessels using techniques such as immunohistochemistry, cell culture and polymerase chain reaction (PCR). Muhlestein et al. (8) have observed that >70% of coronary atherectomy samples obtained from individuals with documented coronary artery disease contained CPn. Importantly, only 4% of specimens obtained from individuals without coronary atheroma showed the presence of the microorganism. These observations have been confirmed and expanded by studies carried out by Maass et al. (9) in Germany, who examined coronary artery specimens in 70 patients who underwent elective or urgent myocardial revascularization for management of their coronary artery disease. Segments from the same atherosclerotic lesions were cultured and also subjected to nested PCR for the assessment of CPn genomic deoxyribonucleic acid (DNA). Viable CPn was cultured from 16% of the 70 atheromatous samples, and CPn DNA was formed in 30% of the samples.
CPn was not found in 17 tissue specimens without atheroma. The DNA sequencing of PCR products revealed no differences between the coronary isolates and respiratory reference strains. This finding confirms previous observations in experimental animal models and indicates that from the respiratory tract, CPn gains access to the circulation carried by alveolar macrophages and subsequently lodges in diseased vascular tissue. The alveolar macrophage thus appears to act both as a reservoir and a carrier for CPn. Recent in vitro studies (10) have shown that peripheral blood mononuclear cells infected with CPn express CPn messenger ribonucleic acid three days after infection and also induce a lymphocyte proliferative response for up to seven days, suggesting that CPn remains metabolically active within monocytes. CPn infection activates macrophages, which in turn produce inflammatory cytokines and metalloproteinases, which may contribute to atheromatous plaque disruption. However, whether CPn plays a causal role in both atherogenesis and progression of coronary artery disease remains a controversial issue. Studies in animals have suggested a possible etiologic role for CPn in the development of neointimal atherosclerotic changes (11). In patients with ischemic heart disease, macrolide antibiotic trials have reported a reduction in the incidence of serious cardiac events (12,13) and reduction of antibody titers to trials have reported a reduction in the incidence of serious cardiac events (12,13) and reduction of antibody titers to trials have reported a reduction in the incidence of serious cardiac events (12,13) and reduction of antibody titers to trials have reported a reduction in the incidence of serious cardiac events (12,13) and reduction of antibody titers to trials have reported a reduction in the incidence of serious cardiac events (12,13) and reduction of antibody titers to trials have reported a reduction in the incidence of serious cardiac events (12,13) and reduction of antibody titers to trials have reported a reduction in the incidence of serious cardiac events (12,13).

Viable CPn and coronary artery disease. The finding of Wong et al. (16) reported in this issue of the Journal (16) that “circulating” CPn DNA is more commonly found in men with coronary artery disease than in those without coronary atheroma contributes to the debate regarding the role of CPn infection in ischemic heart disease. The presence of bacterial DNA is likely to represent active infection as previously documented in animal studies and clinical reports.

Only a small proportion of patients (<10%) in Wong’s study had CPn DNA in their blood mononuclear cells. A much larger proportion of patients, however, tested seropositive for CPn. As previously observed in other studies (9), Wong et al. found no correlation between anti–CPn antibody titers and positive PCR results. However, a trend was observed for seropositive subjects to have a higher prevalence of CPn DNA in their circulating monocytes. It is conceivable that the relatively small number of patients showing CPn DNA may account for the lack of statistical significance regarding this finding.

Wong et al. (16) correctly argued that their method may provide a more accurate detection of active CPn infection than serology, as it is difficult at present to equate positive serology with active chronic infection. This is likely to be due to the fact that no agreed serologic criteria exist to define active infection, and also to the fact that more often than not studies have based their assessment on single serologic measurements as opposed to repeated measurements.

The study by Wong et al. (16), like others previously using different methods for the detection of CPn, has shown a positive association between this infectious agent and coronary artery disease. Although no causality can be inferred from the observations in this study, findings in Wong’s report are of interest because they show a relation between viable CPn and the presence of coronary atheroma. The present study is also important because it may set up the basis for well-designed investigations to assess whether antibiotic treatment can affect the load of circulating CPn DNA and result in improved patient outcome. Future studies may use this observation to ascertain what proportion of those patients showing circulating CPn DNA develop atheromatous lesions over time and to establish the true sensitivity and specificity of the test used by Wong et al. In addition, and as discussed by Wong et al., repeated testing will be necessary to determine the reproducibility of their PCR findings. The prevalence of circulating DNA may vary in the individual patient over time, and it would therefore be important to carry out repeated measurements with and without intervention to assess changes in the rate of circulating intracellular CPn.

The intriguing finding that men but not women showed an association between circulating CPn DNA and coronary artery disease requires further investigation. Although this observation may be just the result of the lack of statistical power in this study, owing to the small number of women, it may also represent a differential response of women to CPn infection. Acute-phase reactants and inflammatory cytokines are under the modulation of estrogen, and studies have shown differential levels of inflammatory markers in women versus men. Thus, studies should be designed to address this question specifically.

Wong et al. (16) are probably right to speculate that the presence of circulating DNA, as assessed in their study, may identify patients with active infection who may benefit from antibiotic treatment. However, further studies should be carried out before findings in this observational report can be systematically used for patient risk stratification or to decide which patients are more likely to benefit from antibiotic intervention. We concur with Wong et al. (16) that it is likely that certain patients are more likely than others to benefit from antibacterial therapy, but this has to be objectively demonstrated. Whether the detection of circulating DNA by PCR, as proposed in this study, represents the test of choice to identify patients who will derive benefit from antibiotic treatment needs to be investigated.

At present there is no conclusive evidence that chronic infection with CPn may lead to ischemic heart disease and that antibiotic therapy should be used in patients with coronary artery disease. However, clinical observations such as those made by Wong et al. (16), together with available clinical and experimental evidence, should represent a stim-
ulus to continue the search for a causal link between chronic CPn infection and cardiovascular disease.

The intriguing and challenging hypothesis that CPn may play a role in atherogenesis or rapid coronary artery disease progression, or both, is attractive and clearly deserves further investigation. Although in our view large, well-designed antibiotic eradication trials are important, major efforts and funding should be directed to the identification of specific patient subsets in whom the hypothesized pathogenic mechanisms are more likely to operate and in whom the proposed therapeutic intervention is more likely to succeed.

Reprint requests and correspondence: Dr. J. C. Kaski, Head, Coronary Artery Disease Research Group, St. George’s Hospital Medical School, Cranmer Terrace, London SW17 ORE, United Kingdom. E-mail: jkaski@sghms.ac.uk.

REFERENCES